METALS IN TISSUES OF DIAMONDBACK TERRAPIN FROM NEW JERSEY

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Abstract. Relatively little is known about contaminants in reptiles, particularly turtles. The distribution of metals in eggs, liver and muscle of diamondback terrapin (*Malaclemys terrapin*) was examined from Barnegat Bay, New Jersey as part of an aquatic study to understand movement of contaminants in the bay. Lead and cadmium were relatively low in all tissues. There were significant differences among tissues for all metals, except lead. Where there were significant differences, levels were highest in the liver, except for chromium. Levels of mercury were 6.6 times higher in the liver than muscle, and manganese levels were 4 times as high. The levels of metals in muscle of diamondback terrapin are below those that might cause effects in consumers, including humans who eat them in stews. However, the level of mercury in liver is sufficiently high to be problematic for consumers and scavengers that eat liver.

Keywords: Barnegat Bay, biomonitoring, consumers, metals, turtles

1. Introduction

Environmental and coastal managers are interested in assessing the health of marine and coastal environments, especially as more people concentrate along coasts, and participate in a variety of activities in coastal waters. Overall, the majority of people in the world live within 100 km of bays and estuaries (Norse, 1993; NRC, 1995). Marine organisms are exposed to a variety of pollutants that enter estuaries from surface runoff, airborne deposition, and natural geochemical cycles. While anthropogenic contaminants from urban, industrial and agricultural runoff are a major issue, levels are augmented by natural geological processes, and global transport (Mailman, 1980; Fitzgerald, 1989).

Because of bioaccumulation and bioconcentration, organisms at intermediate and higher trophic levels are exposed to higher concentrations of chemicals (Van Straalen and Ernst, 1991; Burger *et al.*, 1992; Burger, 1993). In marine systems, most contaminants work concentrates on fish because of their importance in the food chain, including human consumers. Relatively little attention is devoted to other marine vertebrates, such as turtles.

In this article the levels of arsenic, cadmium, chromium, lead, manganese, mercury and selenium in the eggs, liver, and muscle of diamondback terrapin

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(*Malaclemys terrapin*) are reported from Barnegat Bay, at Tuckerton, New Jersey to better understand movement of contaminants in the bay, and to provide baseline data for trend analysis of metal levels. Of particular interest was whether terrapins are accumulating mercury, as are some of the birds that forage on fish from these same waters (Burger and Gochfeld, 1997). For example, the eggs of Forster's tern (*Sterna fosterii*) nesting nearby have some of the highest levels of mercury (mean 2.1 ppm, wet weight) of any birds in the bay, well above levels known to cause adverse effects (Eisler, 1987). Further, with energy deregulation, mercury levels in the bay may increase due to atmospheric deposition from coal-burning power plants in the mid-west. Diamondback terrapins are still caught and eaten in New Jersey and elsewhere along the Atlantic coast. Thus it is important to determine the status of mercury levels in terrapins.

Diamondback terrapins live in estuarine waters from New England to Texas, and would thus be useful for biomonitoring contaminants over a wide geographical range. They are the only turtle to specialize in estuarine environments, with a diet consisting of invertebrates (snails, crabs and worms), as well as some vegetation (Palmer and Fowler, 1975, unpubl. data). In the waters from New Jersey to Florida the terrapins are numerous, and nest in the hundreds on some nesting beaches.

2. Study Area and Methods

Under appropriate state collecting permits, 11 adult female diamondback terrapins were collected along coastal New Jersey from the south end of Barnegat Bay at Tuckerton (Figure 1). Terrapins had a mean carapace of length of 14.3±0.7 cm, well within the range of adult breeding females for the region (Montevecchi and Burger, 1975). New Jersey is a microcosm for examining coastal issues because it is the most densely populated and one of the most highly industrialized states, and yet it has important natural resources, including important fisheries (Burger, 1996; MacKenzie, 1992).

Specimens were frozen immediately and transported to the Environmental and Occupational Health Sciences Institute. Terrapin were dissected using standard dissection tools except for heavy shears for the carapace. Tissues removed for analysis included liver, leg muscle, and eggs (where they were present, N = 8). Soft tissues (2 g wet weight) were digested in ultrex ultrapure nitric acid (2 mL) in a microwave (MD 2000 CEM), using a digestion protocol of three stages of ten min each under 50, 100 and 150 pounds per square inch (3.5, 7.0 and 10.6 kg cm^{-2}) at 70X power. Digested samples were subsequently diluted with deionized water to a final volume of 20 mL and a final acid matrix of 10%. Mercury was analyzed by cold vapor atomic absorption spectroscopy, and all other metals were analyzed by graphite furnace atomic absorption spectroscopy. All concentrations in tissues are expressed in parts per billion (ng g^{-1} on wet weight).

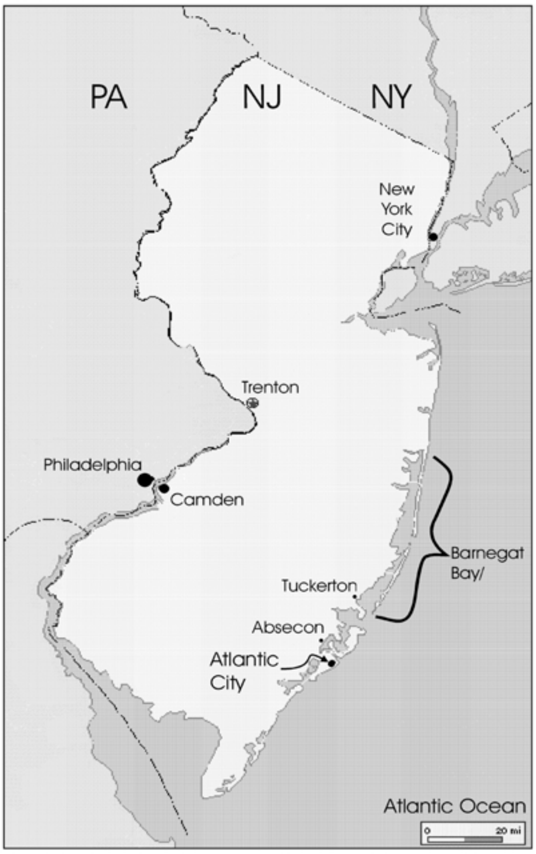

Figure 1. Map of NJ showing collection sites.

Detection limits were 0.02 ppb for cadmium, 0.08 ppb for chromium, 0.15 ppb for lead, 0.09 ppb for manganese, 0.2 ppb for arsenic and mercury, and 0.7 ppb for selenium. All specimens were run in batches of 35 that included blanks, spiked samples, an initial calibration verification (NIST, bovine liver), and after each ten samples there is a continuous calibration verification. The recoveries for spikes ranged from 93% (mercury) to 101% (arsenic). The coefficient of variation (CV) on replicate, samples ranged from 4–7%. Further quality control included periodic blind analysis of samples for each metal (varied by less than ±8%). Duplicate samples of each tissue were run and the means used for analysis.

Non-parametric Kruskal-Wallis One Way Analysis of Variance was used to compare concentrations among tissues, followed by Duncan multiple range test to examine differences among tissues (SAS, 1995). Tissues with a different letter on Table I are significantly different from one another. Kendall tau correlations were used to examine relationships among metals.

TABLE I

Concentration of elements in diamond back terrapins (Tuckerton, NJ). Results are expressed as mean and SE (wet weight) for each tissue. Geometric means are indicated below arithmetic means. Means with the same letters are not significantly different

Metal	Egg	Liver	Muscle	$X^2(P)^a$
Arsenic	12±6 15 (B)	562±168 370 (A)	728±190 551 (A)	6.44 (0.04)
Cadmium	0.26±0.24 0.07 (B)	66±19 52 (A)	18±7 12 (C)	11.29 (0.003)
Chromium	390±255 254 (A)	69±6 67 (B)	297±40 267 (A)	14.74 (0.0006)
Lead	40±30 21 (A)	90±21 72 (A)	62±13 39 (A)	1.70 (NS)
Manganese	248±141 117 (B)	2750±801 1982 (A)	665±143 524 (B)	10.49 (0.005)
Mercury	35±10 32 (B)	1139±473 271 (A)	172±38 123 (A,B)	8.23 (0.02)
Selenium	498±25 497 (B)	1621±260 1503 (A)	507±116 394 (B)	12.12 (0.002)

[a] X^2(P): Analysis of variance test.

3. Results

There were significant differences among tissues for all metals except lead (Table I). Both arithmetic and geometric means are given in Table I to facilitate comparisons with other studies in the literature. For all tissues, cadmium and lead levels were relatively low. Muscle to liver ratios were as follows: arsenic = 1:0.77, cadmium = 1:3.8, chromium = 1:0.23, lead = 1:1.5, manganese = 1:4.1, mercury = 1:6.6, selenium = 1:3.2. Thus the relative storage in the muscle varies, with a relatively higher proportion of arsenic and chromium in the muscle than in the liver.

There were relatively few correlations among metals within muscle, although there were significant correlations between cadmium and chromium (r = 0.62, $P < 0.008$), cadmium and lead (r = 0.56, $P < 0.02$), chromium and lead (r = 0.73, $P < 0.002$), chromium and manganese (r = 0.54, $P < 0.02$), and manganese and lead (r = 0.59, $P < 0.01$). There were fewer significant correlations among metals in the liver: cadmium and mercury (r = 0.64, $P < 0.03$), manganese and chromium (r = 0.57, $P < 0.05$), and manganese and lead (r = 0.57, $P < 0.05$).

4. Discussion

4.1. Tissue comparisons

Very few studies with reptiles compare levels of metals in different tissues, but often they frequently examine whole body levels (see Campbell and Campbell, 2000, 2001). In the present study, levels were higher in the liver than muscle for most metals, except chromium and arsenic, which were slightly higher in the muscle. Thus, as with other vertebrates, the liver is the organ where metals concentrate, with the exception of lead, which concentrates in bone (Ma, 1996).

One objective was to determine which metals are transferred from the female to the egg, and to some extent, all metals were transferred (refer to Table I). Transfer of metals to reptile eggs has been shown for a number of species, including pine snakes (*Pituophis melanoleucus*, Burger, 1992) and slider turtles (*Trachemys scripta*, Burger and Gibbons, 1998).

4.2. Comparisons with other reptiles

There is relatively little information on metal levels in liver and muscle of reptiles, other than alligators, lizards, and snakes (Burger *et al*., 2000; Campbell and Campbell, 2000, 2001). Lizards are useful because of their potential as bioindicators because they are frequently common and widely-dispersed (Campbell and Campbell, 2000). Despite the fact that in many regions people eat turtles, there is little information on contaminant levels in muscle.

More data are available on metal contamination in eggs of reptiles. Mercury levels for snapping turtle egg contents from the St. Lawrence River in Canada ranged from 50 to 180 ppb for 6 pooled samples (Bonin *et al*., 1995). Mercury in 16 eggs of slider turtles from the Savannah River Site, near Aiken, South Carolina averaged 40 ± 15 (Burger and Gibbons, 1998). Metal levels in sea turtle (*Caretta caretta*) eggs varied by nesting beaches, with average mercury levels ranging from 635 ppm to 1391 ppb (Stoneburner *et al*., 1980, Table II). Thus the mercury levels in eggs of diamondback terrapin were relatively low.

Other than sampling on slider and sea turtles, there is relatively little information on other metals in turtle eggs, and almost none on arsenic. In general, the levels of metals in the eggs of diamondback terrapin were similar to or lower than those reported for other turtles (Table II).

4.3. Ecosystem considerations and human risk

Increases in metal levels in higher trophic levels have been noted among many different taxa (e.g. Hothem and Ohlendorf, 1980; Furness, 1996). Mercury and cadmium are key contaminants of concern in marine ecosystems because of their high concentrations in seawater. Monteiro *et al*. (1998) demonstrated a clear relationship between levels of mercury in prey organisms and mercury levels in seabirds,

TABLE II

Comparison of metals in the eggs of turtles. Given are ranges (means) in ppb

Source	Diamondback Terrapin This study	Sea Turtle Stoneburner *et al.*, 1980	Slider Turtles Burger and Gibbons, 1980
Cadmium	0.01–0.74 (0.26)	26–195	3.0–444 (67)
Chromium	120.00–899 (390)	1043–1149	56.0–353 (139)
Lead	6.00–99.0 (40)	1134–2185	173.0–1852 (687)
Manganese	14.00–502	–[a]	1903.0–8224 (4477)
Mercury	21.00–53 (35)	635–1391	0.4–240 (40)
Slenium	469.00–547 (498)	–[a]	131.0–1041 (417)

[a] = Not analyzed, ND = non detected.

and presumably this applies to other vertebrates and their prey. Kim *et al.* (1996) recently suggested that some pelagic seabirds (albatrosses and petrels) are capable of demethylating methylmercury in the liver, and storing it as an immobilizable inorganic form, a chemical pathway which should be examined for estuarine and marine turtles that have accumulated relatively high mercury levels. In diamondback terrapins mercury levels were relatively high in the liver (1139 ppb), but were relatively low in the muscle (172 ppb), well below the limit allowed for interstate commerce in fish muscle tissue (normally 0.5 or 1.0 ppm, FDA, 1987; Lange *et al.*, 1994). Further, risk from consumption is a function of both contaminant levels and consumption rates, and diamondback terrapins are not a significant part of the diet of people.

It is important to know whether the existing levels of metals are high enough to cause a problem for terrapin populations or for predators and scavengers that consume terrapins. Although humans are not apt to consume the liver of diamondback terrapins, other animals are, particularly scavengers. Arsenic residues of over 10 ppm (wet weight) in the liver or kidney of birds are indicative of poisoning (Goede, 1985), and levels of 5 to 10 ppm in liver or kidney of domestic livestock are indicative of poisoning (Vreman *et al.*, 1986). Thus arsenic concentrations in the livers of terrapin appear to be at non-toxic levels. Cadmium levels were also sufficiently low in the terrapin to suggest no problems for consumers. Cadmium damage is first noted in the kidney of mammals (Cooke and Johnson, 1996), suggesting it might also be the organ of concern in terrapins. Sublethal toxic symptoms have been noted at kidney levels of 30 ppm in mammals (wet weight, Chmielnicka *et al.*, 1989).

Hexavalent chromium is a mutagen, teratogen, embryotoxin, and carcinogen, and tissue levels in excess of 12 ppm (wet weight) indicate chromium contamination in a wide range of vertebrates (Eisler, 1986; Domingo, 1994). The chromium levels in terrapin were well below this.

The lead levels in the terrapin were sufficiently low not to pose a problem. Lead levels of 23 ppm in the liver, 32 ppm in the kidney, and 1.2 ppm in the brain (wet weight) have been associated with acute lead poisoning in dogs (Forbes and Sanderson, 1978), and 25 ppm is the critical kidney level for small mammals (Ma, 1989), but there are no comparable data for turtles.

While the levels of cadmium, chromium, lead and arsenic were relatively low in all tissues, levels of manganese, mercury and selenium were higher, particularly in the liver (which would be eaten by many consumers and scavengers). The dietary threshold for selenium, where wildlife that consume them would be affected, is 1.6 ppm (wet weight) for fish skeletal muscle, and 3 ppm (wet weight) for fish liver (Lemly, 1993), suggesting that the levels of selenium in muscle and liver of terrapin are below a toxic level. There is information on the doses of manganese that are toxic in mammals (Domingo, 1994), but there are no studies of the tissue levels associated with toxicity in reptiles, although some effects similar to lead have been found in birds (Burger and Gochfeld, 2000).

Mercury, however, could pose the biggest risk to consumers of terrapin. Tissue concentrations of over 30 ppm (wet weight, liver and kidney) are lethal in some animals, and levels of 1 ppm are associated with adverse effects in some mammals (Eisler, 1987; Beyer *et al.*, 1996). It is not unreasonable to assume that if consumers or scavengers eat terrapin livers containing levels found in this study (1.1 ppm), they might receive a sublethal toxic dose.

5. Conclusions

Levels of most metals were higher in the liver than in muscle, except for chromium and arsenic, where the levels were similar. Selenium, chromium, and manganese were sequestered in the eggs of diamondback terrapin at higher levels than were other heavy metals. Overall, the levels of metals were not sufficient to cause problems for the terrapin or for consumers eating their muscle, although the levels of mercury in liver may be sufficiently high to cause some sublethal effects.

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References

Beyer, W. N., Heinz, G. H. and Redmon-Norwood, A. W.: 1996, *Environmental Contaminants in Wildlife*, CRC Press, Inc., Boca Raton, Florida.

Bonin, J., DesGranges, J. L., Bishop, C. A., Rodrigue, J., Gendron, A., and Elliott, J. E.: 1995, 'Comparative study of contaminants in the mudpuppy (Amphibia) and the common snapping turtle (Reptilia), St. Lawrence River, Canada', *Arch. Environ. Contam. Toxicol.* **28**, 184–194.

Burger, J.: 1992, 'Trace element levels in pine snake hatchlings: Tissue and temporal differences', *Arch. Environ. Contam. Toxicol.* **22**, 209–213.

Burger, J.: 1993, 'Metals in avian feathers: Bioindicators of environmental pollution', *Rev. Environ Toxicol.* **5**, 302–311.

Burger, J.: 1996, *A Naturalist Along the Jersey Shore*, Rutgers Univ. Press, New Brunswick, NJ.

Burger, J. and Gibbons, J. W.: 1998, 'Trace elements in egg content and egg shells of slider turtles (*Trachemys scripta*) from the Savannah River Site', *Arch. Environ. Contam. Toxicol.* **34**, 382–386.

Burger, J. and Gochfeld, M.: 1997, 'Risk, mercury levels, and birds: Relating adverse laboratory effects to field biomonitoring', *Environ. Res.* **75**, 160–172.

Burger, J. and Gochfeld, M.: 2000, 'Effects of lead on larids: A review of laboratory and field studies', *J. Tox. Environ. Health. Part B.* **3**, 59–78.

Burger, J. and Montevecchi, W. A.: 1975, 'Aspects of the reproductive biology of the northern diamondback terrapin *Malaclemys terrapin terrapin*', *Am. Midl. Nat.* **94**, 166–178.

Burger, J., Cooper, K., Saliva, J., Gochfeld, D., Lipsky, D. and Gochfeld, M.: 1992, 'Mercury bioaccumulation in organisms from three Puerto Rican estuaries', *Environ. Monit. Assess.* **22**, 181–187.

Campbell, K. R. and Campbell, T. S.: 2000, 'Lizard contaminant data for ecological risk assessment,' *Rev. Environ. Contam. Toxicol.* **165**, 39–116.

Campbell, K. R. and Campbell, T. S.: 2001, 'The accumulation and effects of environmental-contaminants on snakes: A review,' *Environ. Monit. Assess.* **70**(3), 253–301.

Chmielnicka, J., Halatek, T. and Jedlinska, U.: 1989, 'Correlation of cadmium-induced nephropathy and the metabolism of endogenous copper and zinc in rats', *Ecotoxicol. Environ. Safety* **18**, 268–276.

Cooke, J. A. and Johnson, M. S.: 1996, 'Cadmium in Small Mammals', in W. N. Beyer, G. H. Heinz and A. W. Redmon-Norwood (eds), *Environmental Contaminants in Wildlife*, Lewis Publ., Boca Raton, Florida, pp. 377–388.

Domingo, J. L.: 1994, 'Metal-induced developmental toxicity in mammals: A review', *J. Toxicol. Environ. Health* **42**, 123–141.

Eisler, R.: 1986, *Chromium Hazards to Fish, Wildlife, and Invertebrates: A Synoptic Review*, U.S. Fish and Wildlife Service Rep. 85 (1.6), Washington, D.C.

Eisler, R.: 1987, *Mercury Hazards to Fish, Wildlife, and Invertebrates: A Synoptic Review*, U. S. Fish and Wildlife Service Rep, 85 (1.10). Washington, D.C.

FDA (Food and Drug Administration): 1987, *Compliance Policy Guide*, Food and Drug Administration, Washington, D. C.

Fitzgerald, W. F.: 1989, 'Atmospheric and Oceanic Cycling of Mercury', in J. P. Riley and R. Chester (eds), *Chemical Oceanography*, Vol. 10, Academic Press, New York, NY, pp. 151–186.

Forbes, R. M. and Sanderson, G. C.: 1978, 'Lead Toxicity in Domestic Animals and Wildlife', in J. O. Nriagu (ed.), *The Biogeochemistry of Lead in the Environment, Part B. Biological Effects*, Elsevier/North Holland Biom. Press, Amsterdam, pp. 227–277.

Furness, R. W.: 1996, 'Cadmium in Birds', W. N. Beyer, G. H. Heinz and A. W. Redmom-Norwood (eds), *Environmental Contaminants in Wildlife: Interpreting Tissues Concentrations*, Lewis Press, Boca Raton, FL, pp. 389–404.

Goede, A. A.: 1985, 'Mercury, selenium, arsenic and zinc in waders from the Dutch Wadden Sea', *Environ. Pollut.* **37A**, 287–309.

Hothem, R. L. and Ohlendorf, H. M.: 1989, 'Contaminants in foods of aquatic birds at Kesterson Reservoir, California, 1985', *Arch. Environ. Contam. Toxicol.* **18**, 773–786.

Lange, T. R., Royals, H. E. and Connor, L. L.: 1994, 'Mercury accumulation in largemouth bass (*Micropterus salmoides*) in a Florida lake', *Archiv. Environ. Contam. Toxicol.* **27**, 466–471.

Lemly, A. D.: 1993, 'Guidelines for evaluating selenium data from aquatic monitoring and assessment studies', *Environ. Monit. Assess.* **28**, 83–100.

Ma, W.: 1989, 'Effect of soil pollution with metallic lead pellets on lead bioaccumulation and organ/body weight alterations in small mammals', *Arch. Environ. Contam. Toxicol.* **18**, 617–622.

MacKenzie Jr., C. L.: 1992, *The Fisheries of Raritan Bay*, Rutgers Univ. Press, New Brunswick, NJ.

Mailman, R. B.: 1980, 'Heavy Metals', in J. J. Perry (ed.), *Introduction to Environmental Toxicology*, Elsevier, NY, pp. 34–43.

Monteiro, L. R., Granadeiro, J. P. and Furness, R. W.: 1998, 'Relationship between Mercury levels and diet in Azores seabirds', *Mar. Ecol. Progr. Ser.* **166**, 259–265.

National Research Council: 1995, *Understanding Marine Biodiversity*, National Academy Press, Washington, DC.

Norse, E. A. (ed.): 1993, *Global Marine Biological Diversity Strategy: Building Conservation into Decision Making*, Center for Marine Conservation, Washington, DC.

Palmer, E. L. and Fowler, H. S.: 1975, *Fieldbook of Natural History*, McGraw-Hill Book Co., New York, NY.

Stansfield Jr., C. A.: 1983, *New Jersey*, Westview Press, Boulder, Colorado.

Stoneburner, D. E., Nicora, M. N. and Bloud, E. R.: 1980, 'Heavy metals in loggerhead sea turtle eggs (*Caretta caretta*): Evidence to support the hypotheses that demes exist in the Western Atlantic population', *J. Herpetol.* **14**, 171–175.

Van Straalen, N. M. and Ernst, E.: 1991, 'Metal biomagnification may endanger species in critical pathways', *Oikos* **62**, 255–256.

Vreman, K., Veen, N. G., Molen, E. J. and De Ruig, W. B.: 1986, 'Transfer of cadmium, lead, mercury and arsenic from feed into milk and various tissues of dairy cows: Chemical and pathological data', *Neth. J. Agric.* **34**, 129–144.