

Port of San Diego Student Research Project Update

Submitted: March 8, 2007

Project: Assessing contaminant levels and health effects in the San Diego Bay green turtles

Investigators:

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Dr. Rebecca Lewison, PhD., Associate Professor, San Diego State University

Dr. Peter Dutton, PhD., Head of Marine Turtle Research, Southwest Fisheries Science Center



Summary:

The first portion of this research project consisted of developing and validating field and laboratory protocols, collection of samples, and conducting preliminary data analyses. The field season for turtle sample collection began in November 2007 and will conclude in May 2008. To date, scute and blood samples have been collected from 17 and 13 turtles, respectively. Blood biochemistries and complete blood cell counts have been conducted on all blood samples collected to date. Contaminant analyses of the blood samples are scheduled to begin in April 2008. Trace metal analyses have been conducted on the scutes of 14 turtles via ICP-OES. Mercury analyses of these samples are currently being completed. Food web samples (eelgrass, algae, sediment, water, and invertebrates) have been collected bi-monthly beginning in August 2007 and analyses of these samples is currently being conducted.

Scute ICP-OES results

Trace metal analysis via ICP-OES yields data on the concentration of the following elements: Ag, Al, As, Cr, Cu, Fe, Mn, Ni, Pb, Se, Sr, Ti, V, and Zn. We first wished to analyze these metals and assess their variability within the individual and the population. We took eight to ten samples from different scutes on each turtle and analyzed 86 total scute samples from 14 individuals captured from November 2007 to February 2008. This analysis found that aluminum (Al), iron (Fe), and zinc (Zn) were by far the most prevalent trace elements stored in the scutes (Figure 1A and 1B). This is not surprising because two of these metals, Fe and Zn are known to be essential elements necessary for various metabolic processes. The high levels of Al, however, is very concerning because it is one of the few abundant elements that have no known function in living cells. Aluminum competes with calcium for absorption, so its toxicity can be traced to increased deposition in bone and the central nervous system, particularly in the presence of reduced renal function.

One of the goals for collecting ten samples per individual was to investigate intra-individual variability in contaminant loads and compare the variability to that of the population. However, our preliminary results have indicated a large amount of variability both within and between individuals, and variability found in Al concentrations depicted below (Figure 2) was also displayed in most other elements. Further investigation will be conducted upon the analysis of the second set of scute samples scheduled to be run at the end of the field season in May 2008.

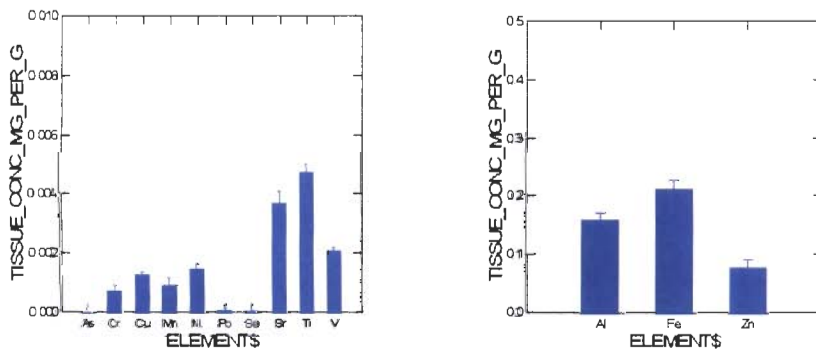


Figure 1A and 1B: Arithmetic mean tissue concentrations of trace elements found in the scutes of 14 individuals from the San Diego Bay. Note that Al, Fe, and Zn are presented in a different graph due the difference in scale of their concentrations from the rest of the elements.

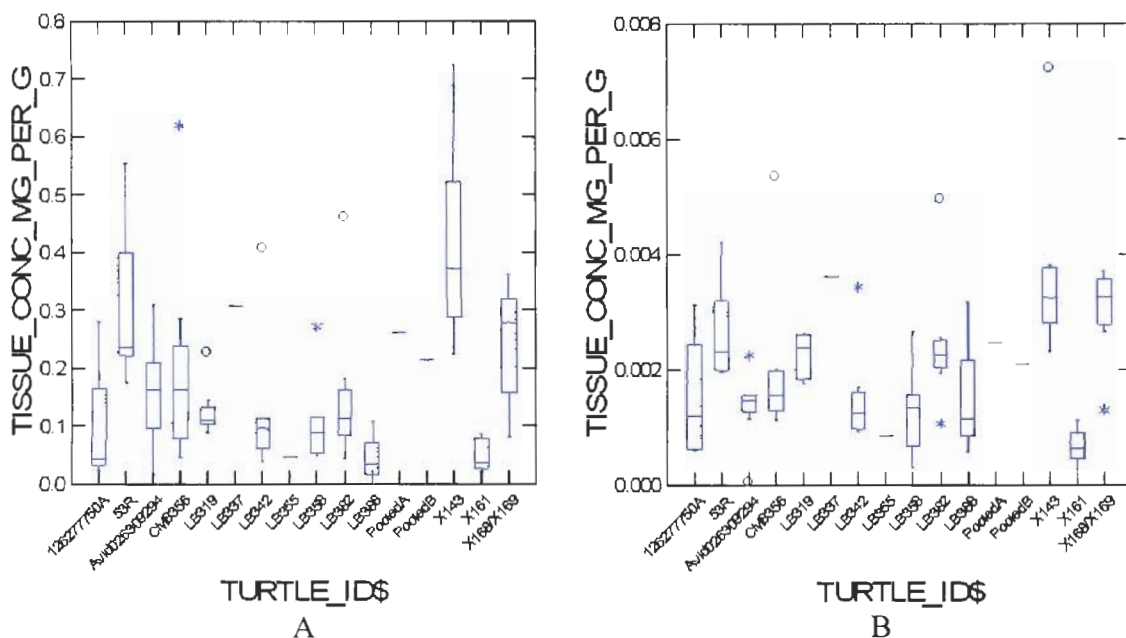


Figure 2: A box plot displaying the variability of Aluminum (A) and Vanadium (B) concentrations within and between individual turtles. Note that pooled samples A and B do not have a range because only one replicate was run as a check for standards.

There have been some reports of significant positive or negative contaminant load relationships with sea turtle size, which is used as a proxy for age. However, linear regressions between the curved carapace length (CCL) and contaminant concentration did not reveal any strong correlations between any elements and CCL. ANOVA's were also run for each element with CCL, and did not report any significant relationships. Further investigation of this will be conducted as well in May 2008 with the complete data set. A compilation and comparison of trace metal values found in other populations of green turtles is currently being conducted and will also be included in the final data analysis to compare the contaminant loads of the San Diego Bay turtles to other populations of this species.

Health Assessments

Health assessments were attempted on all turtles from which it was possible to draw blood. Initial problems with the collection hematology protocols inhibited the evaluation of several samples. The development and verification of these methods was completed in late February 2008, and it is expected that it will be possible to attain adequate samples from all future captures to conduct these tests. The relevant preliminary data attained to date is listed below (Table 1). The parameters listed are commonly used as indices to assess the liver and kidney function, immune response, and other measurements of internal health in vertebrates. The last row show a list of average reference values attained from the University of Florida green turtle research project. Unfortunately this is the only adequate data set currently available for comparison of our samples with a "healthy population" of the same species. Unfortunately this source does not have values for several of the parameters we are measuring in our population. Our research team is currently in pursuit of another reference range source that will allow us to compare all of the parameters we are measuring. It is obvious, however, that there is a large amount of variability in our population. As expected, there does seem to be some different patterns of parameters depending on sex (Figure 3). However, a complete analysis will not be conducted until an ample sample size is attained (May 2008).

Turtle_ID	Sex	CCL (cm)	Weight (kg)	%White Blood Cell	Heterophil	%Lymphocytes	H:L Ratio	%Hematocrit	AST (IU/L)	BUN (mg/dL)	
126277750A	Juv	54.5	19	4	59	39	1.5		178	27	
Avid26309294	M	93.5	119	10	20	80	0.3		171	18	
X143	M	106.4	140	6	40	50	0.8		121	30	
X168/X169	M	91.7	113	6	30	52	0.6		133	17	
132316632A	M	100	135	10	21	67	0.3	52	183	10	
X161	M	107.5	151	6	16	61	0.3	45	131	19	
LB358	Juv	79.6	59	10	18	79	0.2	35	141	19	
LB319	F	109.2	181	10	30	57	0.5	44	169	20	
126461461A	Juv	52.1	19	6	0	0		38	178	16	
LB342	F	114.2	202	3	72	4	1.8	69	369	10	
123919454A	Juv	52.7	17		26	67	0.4				
X143	M	106	140		10	45	0.2	33			
UFlorida Ref									28.70869565	274	19.08695652

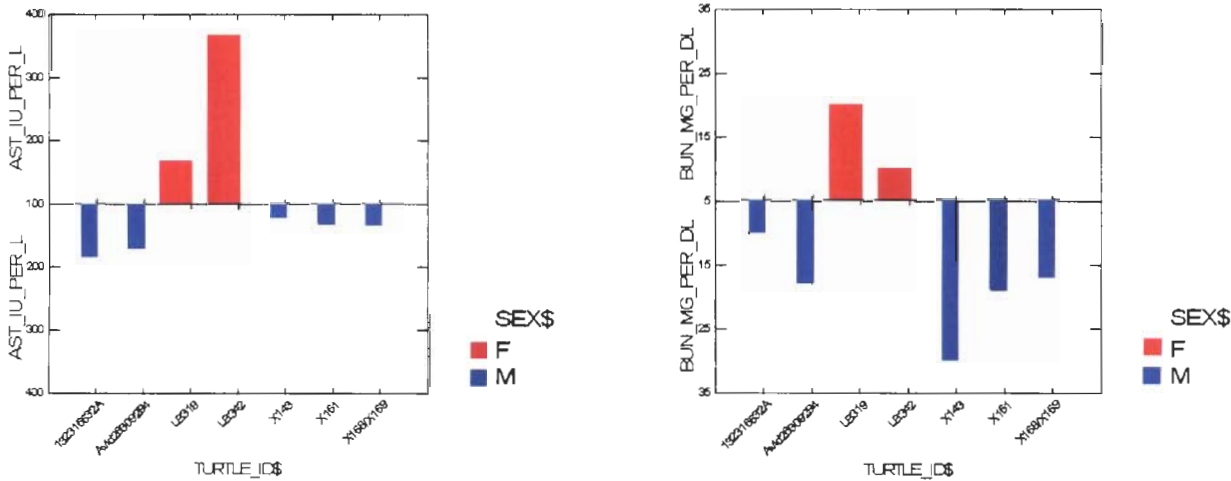


Figure 3. A. AST is a measure of liver function. B. BUN is a measure of kidney function.

Current and Future Analyses

We are currently collecting the rest of our turtle samples, and will continue to collect our food web samples bimonthly through the summer 2008. Also during that time, we will be conducting the laboratory analyses systematically to complete our proposed project. We are currently fine tuning the protocol for analysis of algae and eelgrass using ICP-OES at Scripps Institute of Oceanography. Once this is complete we will be running these analyses and then move on to the analysis of the turtle blood samples, as well as the water and sediment samples. We expect to complete the trace metal analyses of all samples by the Fall of 2008. We plan to collect a second season of samples for the turtles beginning in November 2008.

As outlined in our budget, the first third of the student grant we received was used to purchase supplies for the project, boat fuel, conduct the preliminary analyses. We are now requesting the second third in order to be able to continue to purchase boat fuel to collect our samples and pay for the laboratory costs to run the ICP-OES and the mercury analyses (CVAFS) on the samples we have collected. If any further information is needed in order to complete this request, please contact me at lisa.komoroske@gmail.com. Thank you very much for your consideration and aid in completing this research.



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3/13/08

Interim Progress Report – Statement from Advisor

Project title: Heavy metal contaminants in Endangered green turtles in the Port of San Diego

Grantee: Lisa Komoroske

Dear Eileen,

I am writing to express my support and confirmation of the progress Lisa Komoroske is making on her Port-funded project “**Heavy metal contaminants in Endangered green turtles in the Port of San Diego**”. Lisa has been collecting and analyzing samples and is making progress relative to the goals of her project.

Please don't hesitate to email or call me if you have any questions.

Sincerely,

Rebecca Lewison, PhD.

Assistant Professor

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