POLYCHLORINATED BIPHENYLS, ORGANOCHLORINE PESTICIDES, AND PERFLUOROOCTANESULFONATE IN THE BLOOD OF LOGGERHEAD SEA TURTLES

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Introduction

Monitoring environmental contaminants is an important aspect in wildlife health assessment studies, especially for species with reduced or declining populations such as sea turtles. The loggerhead sea turtle (*Caretta caretta*) is a threatened species; therefore, nonlethal sampling procedures are necessary. The goal of this study was to assess the use of blood from juvenile loggerhead sea turtles for monitoring persistent organic pollutants (POPs).

Blood offers many benefits for monitoring contaminants. Reasonably large volumes can be collected safely and relatively non-invasively from live turtles and can be sampled repeatedly for continuous monitoring. Contaminant concentrations in blood may better represent the exposure levels of target tissues. In addition, several studies have shown that blood POP concentrations are correlated to concentrations present in fatty tissues¹⁻⁴. In all of those studies, statistically significant, positive correlations were observed between blood and fatty tissue concentrations of POPs. Boon *et al.*⁵ provides data on harbor seals that support a kinetic model, in which POP concentrations in the blood are in a dynamic balance with fatty tissues.

One significant drawback to using blood for monitoring contaminants is that blood concentrations may fluctuate more than those in fatty tissues as a result of recent dietary intake or lipid mobilization. For example, POP concentrations in the blood of humans and seals increase following dramatic weight loss^{6,7}. Overall, these prior studies suggest that blood POP concentrations will fluctuate during lipid mobilization, but in many situations, and with many wildlife species, blood can be used to successfully monitor ambient POP concentrations in wildlife.

The current study investigated the concentrations of polychlorinated biphenyls (PCBs) and organochlorine (OC) pesticides in paired samples of blood and adipose tissue from juvenile

loggerhead sea turtles. The relationships of concentrations between the two tissues were determined. We also investigated the effect of lipid mobilization on blood POP concentrations by comparing the percent lipid measured in adipose biopsy samples to the blood POP concentrations, assuming that turtles recently mobilizing lipid stores would have lower lipid in their fatty tissues. Next, we examined temporal variability of POP concentrations in blood using turtles that were recaptured. Finally, blood/plasma samples were analyzed for perfluorooctanesulfonate (PFOS) to examine the concentrations of fluorinated surfactants that have been measured in other wildlife⁸.

Methods and Materials

During August 2000 and July 2001, 44 juvenile loggerhead sea turtles were captured using the pound-net fishery in Core Sound, North Carolina. Blood samples and adipose biopsies were collected from each turtle for PCB and OC analysis⁹. Six of these turtles were captured again at a later date and another blood sample was taken. Blood or plasma samples from an additional 11 turtles captured between July 2000 and October 2001 were used to measure PFOS.

Approximately 5 g of whole blood and 0.4 g to 4 g of adipose tissue were analyzed for 55 PCB congeners and 24 OC pesticides as described elsewhere⁹. Briefly, blood was extracted using a liquid:liquid extraction technique employing formic acid and 1:1 MTBE:hexane. Adipose samples were minced and extracted using a pressurized fluid extractor. Cleanup of the extracts was performed using alumina columns for blood and gel permeation chromatography for adipose. PCB congeners were fractionated from the pesticides in both blood and adipose extracts using an aminopropylsilane column⁹. Analysis was performed using gas chromatography (GC) with dual micro-electron capture detectors for all adipose extracts was performed using GC with low resolution mass spectrometry operating in the electron-impact mode and using selected ion monitoring.

PFOS was measured in blood/plasma samples as described elsewhere⁸. Briefly, tetrabutylammonium hydrogensulfate was added as an ion-pairing reagent to 1 ml of blood/plasma and the analyte ion pair is partitioned into methyl-tert-butyl ether. The instrumental analyses were performed using a HPLC interfaced with an electrospray mass spectrometer by monitoring the primary ion characteristic of PFOS.

Results and Discussion

The relationships of POP concentrations between the blood and adipose are shown in Figure 1 (\sum PCBs and \sum DDTs are shown). Significant Spearman Rank correlations were observed between the blood and adipose concentrations of \sum PCBs ($r_s = 0.63$), \sum DDTs ($r_s = 0.67$), \sum chlordanes ($r_s = 0.72$), mirex ($r_s = 0.63$), dieldrin ($r_s = 0.58$), and heptachlor epoxide ($r_s = 0.44$). The turtle with the highest concentrations appeared to be a strong component of the correlations, but the correlations remained significant even after removing this turtle. These relationships suggest that the POP concentrations in blood reasonably predict the concentrations in fatty tissue.

In mammalian wildlife, lipid mobilization during weight loss results in an increase of POP concentrations in the blood, which may affect the utility of using blood to monitor contaminants⁷. We investigated this possibility in sea turtles by comparing the lipid content of loggerhead sea turtle adipose tissue to their blood POP concentrations (Figure 2). We assumed that turtles

undergoing lipid mobilization would have lower percent lipid in their fat stores. Weak, but significant, negative Spearman Rank correlations were observed between the percent lipid in the adipose tissue and blood concentrations of $\sum PCBs$ ($r_s = -0.40$), $\sum DDTs$ ($r_s = -0.36$), \sum chlordanes ($r_s = -0.67$), mirex ($r_s = -0.55$), and heptachlor epoxide ($r_s = -0.43$). These relationships suggest that POPs may be released into the blood during lipid mobilization, thus increasing exposure. It is important to note that these relationships are weaker than the correlations observed between blood and adipose POP concentrations, suggesting that blood POP concentrations relate more strongly to the POP concentrations in adipose tissue than to the percent lipid in adipose tissue.

Figure 1: Correlations between adipose biopsies and blood for $\sum PCBs$ and $\sum DDTs$ concentrations



Figure 2: Correlations between % lipid in adipose and blood POP concentrations



In order to examine the temporal variability of POP contaminants in blood, we analyzed a second blood sample from six recaptured loggerhead turtles. The duration between sampling ranged from 17 to 403 days, and all turtles grew during that period. The blood POP concentrations varied considerably. Some doubled while others decreased by a half (Figure 3). POP concentrations increased in four of the six turtles, while a decreasing trend was observed in the other two turtles. The changes did not correlate to the length of time between sampling events. Conclusions based on this small sample size are limited, but it is evident that blood POP concentrations can change dramatically over short intervals of time. Unfortunately, adipose biopsies were not collected upon recapture, so the changes in adipose POP concentrations are unknown. This fluctuation in blood

concentrations, though, should be considered when designing a monitoring study that relies solely on blood.





* Numbers above columns indicate duration (days) between blood sampling.

PFOS was detected in 7 out of 11 turtle blood/plasma samples. The concentration of PFOS in the sea turtle blood (<1 - 27.5 ng/mL) was much lower than concentrations measured in plasma of bald eagles from the Great Lakes region (<1 - 2030 ng/mL), but similar to serum of albatrosses from Midway Atoll (3.0 - 34 ng/mL)⁸. These preliminary measurements suggest that PFOS is low, but measurable in most loggerhead sea turtles.

Conclusions

PCBs, OC pesticides, and PFOS are detectable in the blood of most loggerhead sea turtles from the nearshore waters of North Carolina. Blood concentrations of POPs can predict the concentrations stored in the adipose tissue and most likely represent the concentrations reaching target tissues. However, monitoring contaminants with blood does have its limitations because of significant fluctuations in POP concentrations.

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