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Crocodile Bones as archives of Pollution Exposure: Lead Contamination in Kakadu National Park, and what's in Sweetheart's Osteoderms?

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Investigations of biological archives using secondary ion mass spectrometry (SIMS) has been extended from freshwater bivalves to other long-lived fauna with laminated calcified structures, such as estuarine crocodiles, that have been exposed to metal contaminants in their aquatic environments.

Estuarine crocodiles (*Crocodylus porosus*) in some sections of Kakadu National Park (KNP) are known to consume fauna shot with lead ammunition, leading to concerns that they may be contaminated with lead. A program of sampling both crocodile osteoderms and edible flesh, over a wide size range of animals from the three main catchments of KNP, provided samples for analysis of lead (Twining et al., 1999). Analysis of the lead in the osteoderms and flesh of 40 crocodiles sampled throughout KNP showed elevated levels ($P < 0.001$) in both tissues from individuals in the two exposed habitats, relative to all other individuals. Analysis by SIMS of the Pb-208/Ca-42 signal ratios across the osteodermal laminations of lead-exposed crocodiles confirmed they were elevated and relatively constant (Twining et al., 1999). These results suggested that: (a) the crocodilian digestive system can retain lead ammunition following ingestion and solubilise and absorb it into the body, and (b) crocodiles were exposed to elevated lead levels during most of their life, that they could apparently tolerate.

An experimental study undertaken at Crocodylus Park, northern Australia, demonstrated the following. After ingestion of lead shot the lead concentration in the blood immediately increased, appearing to have reached an equilibrium concentration in 30-40 days of ca. 350 mg dL-1, which was an increase above background of more than an order of magnitude (**Figure 1**). This result confirmed the ability of the digestive system of C. porosus to solubilise lead from the ingested shot and absorb it into the blood at a rapid rate. The subsequent lavage and radiography of the stomachs of exposed crocodiles retrieved or identified more than 70% of the individual ingested lead shot pellets, confirming their ability to retain lead shot (Jeffree et al., 2001).

These experimental findings made it then possible to evaluate the hypothesis that the osteodermal laminations would record enhanced blood lead concentrations resulting from the ingestion of lead shot. At about 140 days following lead shot ingestion, two osteoderms were removed from each exposed and control animal. SIMS analysis of Pb-208 and Ca-42 signal intensities was then performed on sections that were prepared and analysed using similar methods previously used on field-collected specimens (Twining et al., 1999).

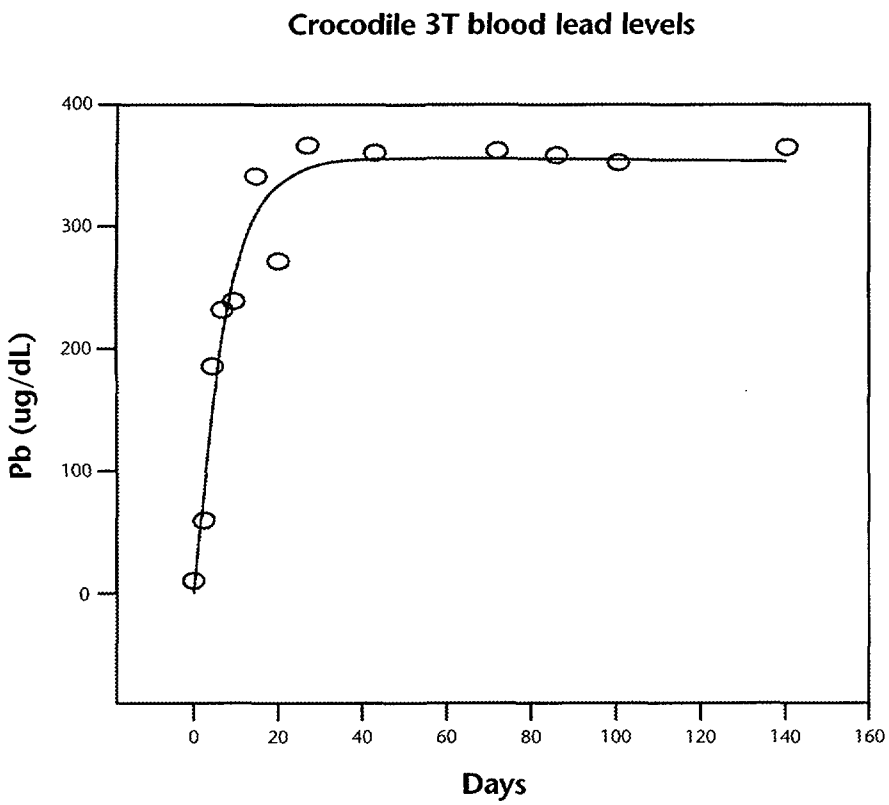
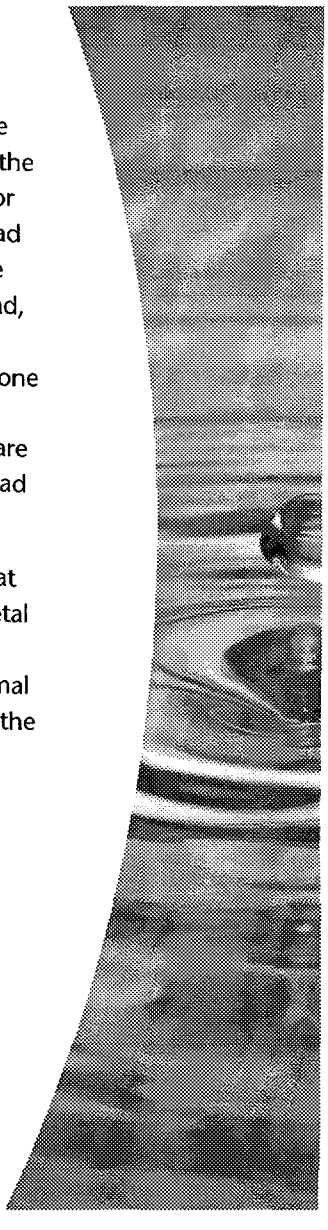
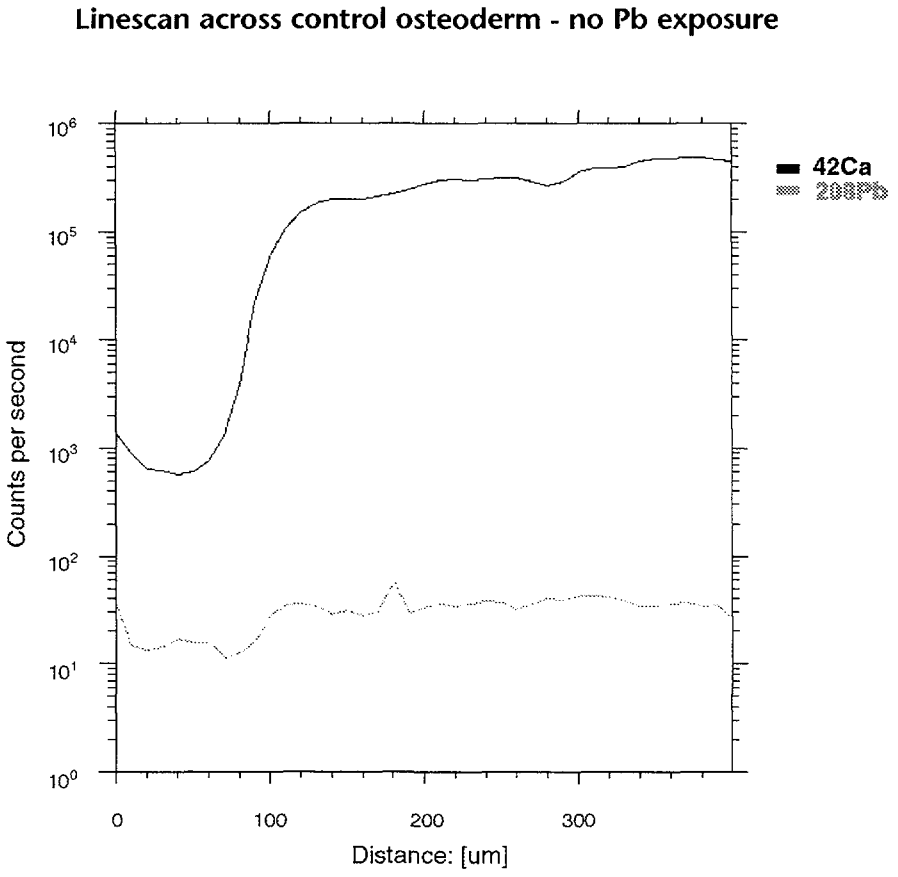


Figure 1. Pattern of lead concentration ($\mu g\ dL^{-1}$) in the blood of *C. porosus* plotted against time (days) following ingestion of lead shot pellets (five).

Figure 2 shows SIMS line scans for these two isotopes, that begin in the skin (0), in which the osteoderms are constructed, and move through the skin/bone interface (~100 μm), through the most recently deposited laminations to those progressively constructed at greater periods prior to experimental treatment (>100 to 200 μm), for both a control specimen (i) and one that had ingested five lead shot (ii). For both specimens, the Ca signal increases through the skin/bone interface to remain elevated through the rest of the osteodermal laminations. However for lead, the signal is relatively constant throughout the control section but the lead-exposed animal shows an increase in signal intensity by more than an order of magnitude through the skin/bone interface and most recently constructed laminar material, before declining by an order of magnitude in the previously constructed laminations of the osteoderm. These initial findings are consistent with the hypothesis that incremental laminations of the osteoderm will archive a lead signal that responds to enhanced levels of lead in the animal's blood during its life.

These results indicated that the osteoderms from the skin of 'Sweetheart', that was captured at Sweet's Lookout in the Finniss River in 1978, and was at least 50 years of age, may record metal concentrations in its aquatic environment during both the pre-mining and mining phase, particularly that period before contaminant loads were measured. Accordingly, two osteodermal cores were obtained from the mounted skin of Sweetheart in the Museum and Art Gallery of the Northern Territory, Darwin. These were prepared for scanning electron microscopy with microprobe EDS and SIMS analysis, using techniques similar to those described in Twining et al. (1999).





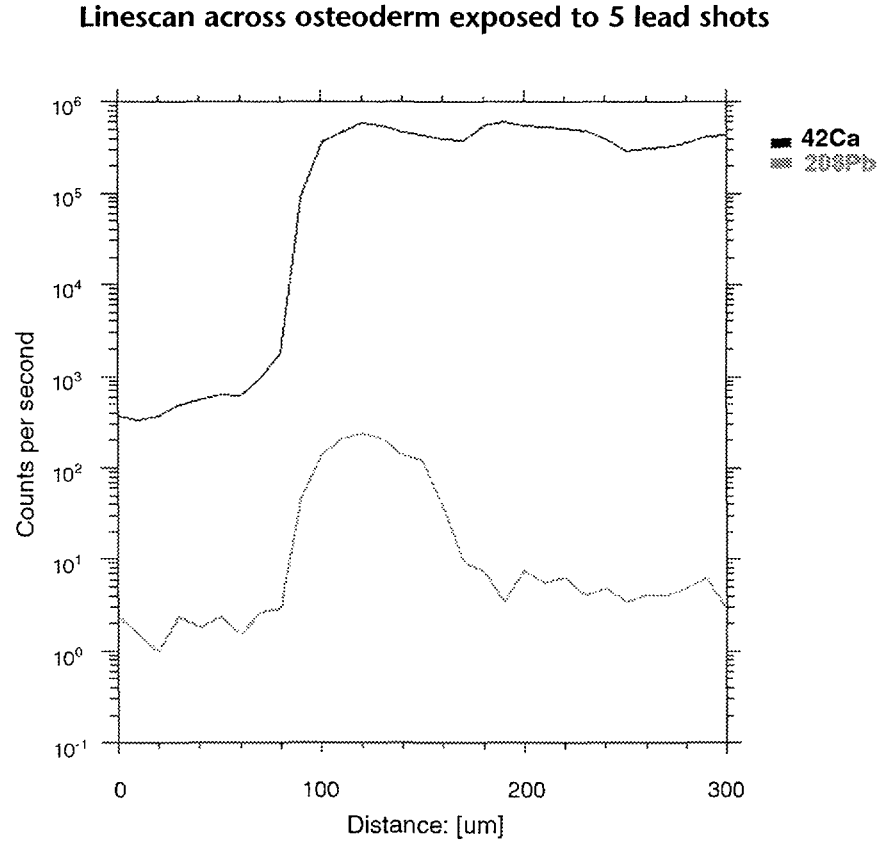


Figure 2. SIMS line scans of Ca-42 and Pb-208 signals across the skin and laminated osteoderm from *C. porosus* (i) unexposed and (ii) exposed to lead shot ammunition previously ingested with food.

SEM analysis identified the laminated structure of the osteoderm, however microprobe analysis did not detect appreciable amounts of even Ca and P, although the organic matrix was obviously present. It is proposed that this anomalous result is due to the preparatory tanning of the skin, in acid solution, that could be expected to leach elements from the organic matrix. An analogy is drawn with the decalcified skeletons in the bodies of the Druid sacrifices due their deposition in acid swamp waters in the UK.

The challenge is now to sample osteoderms from large crocodiles in the Finniss River, preferably in close proximity to the Rum Jungle mine site, where any archived pollution signal would be more intense, and then repeat this analytical investigation of the osteodermal history of contaminant loadings.

References

Jeffree, R.A., Markich, S.J., Hammerton, K.M. and Russell, R. 2001. Accumulation of Radionuclide and Metal Contaminants in Flesh and Osteoderms of Estuarine Crocodiles (*Crocodylus porosus*): Pathways and Histories of Catchment-Specific Exposure. ECORAD 2001 Congress, 3-7 September, Aix-en-Provence, France.

Twining, J.R., Markich, S.J., Prince, K.E. and Jeffree, R.A. 1999. Osteoderms of estuarine crocodiles record their enhanced lead exposure in Kakadu National Park. Environ. Sci. Technol. 33, 4396-4400.