Comparison of the elemental composition of different hard parts of freshwater fish (otoliths, scales, fin rays, vertebrae and eye lenses) using ICPQMS

Leo Kirchmaier[1], Andreas Zitek[1,2], Johanna Irrgeher[1], Thomas Prohaska[1]

[1] University of Natural Resources and Life Sciences, Department of Chemistry, Division of Analytical Chemistry, VIRIS Laboratory, Konrad-Lorenz-Straße 24, 3430 Tulln, Austria

[2] University of Natural Resources and Life Sciences, Department of Water, Atmosphere and Environment, Institute of Hydrology and Aquatic Ecosystem, Max-Emanuelstraße 17, 1180 Vienna

Introduction

Fish hard part structures grow continuously throughout the life of a fish and incorporate trace elements from their surrounding environment. Therefore, these structures provide a powerful tool to reconstruct the environmental life history of individuals by measuring their trace element contents for instance by solution based ICPQMS (inductively coupled plasma quadrupole mass spectrometry). While otolith (ear stone) trace element analysis is well established in fishery science, we still lack information on alternative structures (eye lenses, vertebrae and for non-lethal sampling: scales, fin rays).

This study is the **first to our knowledge that provides information** on the **trace element content** in absolute values **comparing the following five structures of different matrices**

- hydroxy apatite (HAp): scales, vertebrae, fin rays
- calcium carbonate: otoliths
- stable proteins (crystallins): eye lenses

of a typical **cyprinid fish species of the Danube River**, cactus roach (*Rutilus virgo*, Heckel 1852 according to Kottelat & Freyhof (2007)).

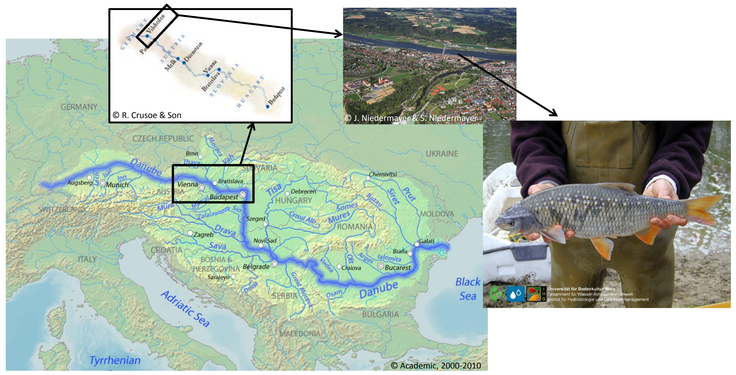


Fig. 1: Danube catchment area showing the sampling site in Vilshofen and the species of interest: *Rutilus virgo*

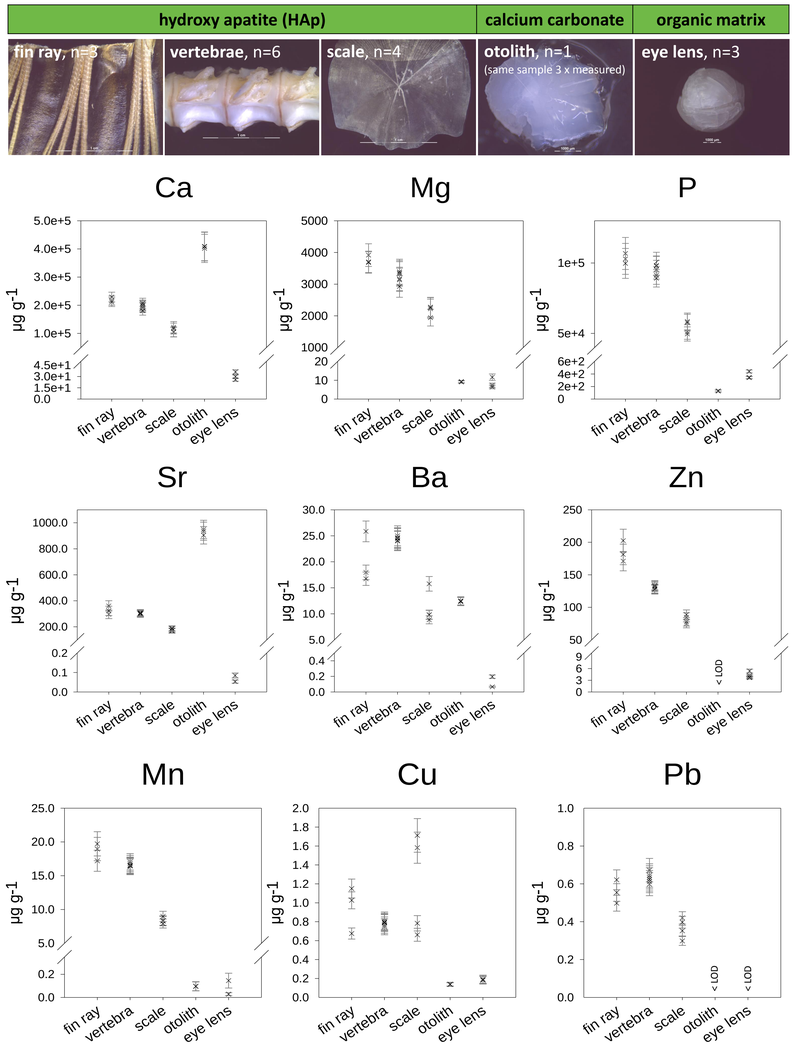


Fig. 2: Elemental mass fractions of fish hard parts; error bars : U_c (k=2)

Results (Fig.2)

- **Ca** dominates in calcium carbonate (40 % (w/w) in otolith) and HAp structures (ranging from 11 % in scales to 20 % in vertebrae and fin rays)
- **Sr/Ca:** mean ratios were similar for HAp structures ranging from 0.00149 to 0.00159 while those for otoliths and eye lenses ranged from 0.00206 to 0.00229
- **Pb:** detectable above LOD only in HAp structures
- **Mn, Cu:** highest levels in HAp structures
- **Zn:** highest levels were found in fin rays (170 – 200 µg g^{-1})
- **Ba** levels were comparable between HAp structures and otoliths
- **Sr:** highest levels were found in otoliths (900 – 950 µg g^{-1})
- **Mn, Zn:** similar distribution behaviour in all tissues
- **Zn:** not above LOD in otoliths but at levels of 4.0 – 5.5 µg g^{-1} in eye lenses

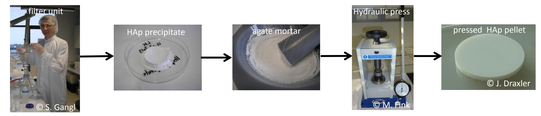
Summary and Conclusion

- Significant differences in trace element concentrations between hard parts
- Sr serves as proxy for Ca for all tissues
- HAp structures: higher degree of calcification in vertebrae and fin rays compared to scales
- HAp structures might better serve as tracers for environmental pollution for Pb, Zn, Mn, Cu because of their higher levels found compared to otoliths
- In otoliths concentrations well above LOD only for Ca, P, Sr, Mg and Ba
- Generally low elemental mass fractions in the organic matrix of eye lenses

Analytical procedure

- Fish Sampling at Danube River site in Vilshofen (Fig. 1) – species: cactus roach
- Hard part preparation – cleaning procedure using ultrasonic bath and ultrapure water followed by microwave assisted acid digestion (HNO_3 and H_2O_2), Multiwave 3000 (Anton Paar GmbH, Austria)
- Elemental analysis – ICPQMS using the NexION 300D (PerkinElmer, USA)
- Method validation was done using the following certified reference materials (CRMs): Riverine water (NRC SLRS-5), Bone ash (NIST SRM 1400), Bone meal (NIST SRM 1486), Fish otolith (NRC FEBS-1)
- Calculation of combined uncertainties U_c (k=2) using a Kragten approach

Outlook



- Time resolved analysis of fish hard parts using laser ablation (LA)-ICPQMS
- Validation of in-house prepared matrix-matched HAp reference materials for LA-ICPQMS
- Sampling of fish from a second location in the Danube catchment - Nötschbach, Carinthia with elevated Zn and Pb levels in water

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CONTACT: Leo Kirchmaier (leo.kirchmaier@boku.ac.at)