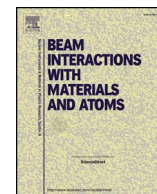




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Provenance of whitefish in the Gulf of Bothnia determined by elemental analysis of otolith cores

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ABSTRACT

The strontium concentration in the core of otoliths was used to determine the provenance of whitefish found in the Gulf of Bothnia, Baltic Sea. To that end, a database of strontium concentration in fish otoliths representing different habitats (sea, river and fresh water) had to be built. Otoliths from juvenile whitefish were therefore collected from freshwater ponds at 5 hatcheries, from adult whitefish from 6 spawning sites at sea along the Finnish west coast, and from adult whitefish ascending to spawn in the Torne River, in total 67 otoliths. PIXE was applied to determine the elemental concentrations in these otoliths. While otoliths from the juveniles raised in the freshwater ponds showed low but varying strontium concentrations (194–1664 µg/g), otoliths from sea-spawning fish showed high uniform strontium levels (3720–4333 µg/g). The otolith core analysis of whitefish from Torne River showed large variations in the strontium concentrations (1525–3650 µg/g). These otolith data form a database to be used for provenance studies of wild adult whitefish caught at sea. The applicability of the database was evaluated by analyzing the core of polished otoliths from 11 whitefish from a test site at sea in the Larsmo archipelago. Our results show that by analyzing strontium in the otolith core, we can differentiate between hatchery-origin and wild-origin whitefish, but not always between river and sea spawning whitefish.

1. Introduction

Otoliths are often the last remnants of decayed fish and can be preserved for millions of years [1]. They can pass through the digestive system of animals and have been used to determine the number, species and weight of fish consumed e.g., by birds [2]. The shape of the otolith is specific for each fish species and the layered structure, the growth rings, will reveal the age of the fish. Even daily increments can be seen in otoliths [3].

The otoliths aid the fish in balance and hearing and consist of calcium carbonate and organic material of protein origin. The protein content is only a few percent and the rest is mainly calcium carbonate which most often occurs as aragonite crystals in Teleostian fish. Strontium can replace calcium in the aragonite structure. The strontium content in otolith is known to reflect the strontium content of the ambient water. Otolith microchemistry has been used to study anadromous species that migrate from the sea into freshwater to spawn [4].

Whitefish (*Coregonus lavaretus*) in the Baltic Sea can be divided into two ecotypes according to their spawning strategies: one spawning in

shallow bays along the coastline and one spawning upstream rivers [5]. However, the construction of hydropower plants and weirs may obstruct the whitefish migration to many of their spawning grounds in rivers. Furthermore, the increased eutrophication has a negative effect on the spawning grounds. The wild river spawning whitefish is endangered and is in risk of becoming extinct [6]. To compensate for the decline of whitefish, huge amount of one-summer old fingerlings has been stocked along the Finnish coastline [7]. However, the proportion of stocked adult fish in the sea is unknown.

Our attempt was to use otolith microchemistry to differentiate between wild and stocked fish and to determine the provenance of the stocked fish. By elemental characterization of otoliths from whitefish juveniles from freshwater ponds at hatcheries, from local adult sea spawning whitefish along the Finnish west coast, and from adult river spawning whitefish, we built a database that helped identifying the provenance of the adult whitefish captured at sea. A low strontium concentration in the core of an otolith from an adult whitefish indicates that the fish has spent most of its first year in fresh water, e.g., a freshwater hatchery pond. Furthermore, by comparing the strontium

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concentration in the otolith core with concentrations found in otoliths from juvenile fish raised in ponds, the hatchery may be identified. The database was evaluated in a case study of whitefish captured during spawning time at sea at the Larsmo archipelago. At this site whitefish juveniles raised in fresh water ponds in Taivalkoski have been annually stocked during many years and the presence of fish originating from Taivalkoski is therefore expected.

The present study is part of a larger project on the development of methods for differing between hatched-origin and wild-origin fish, but also for differing between the two ecotypes of whitefish along the Finnish west coast. Besides the elemental analysis of otoliths with ICP-OES (bulk analysis), LA-ICP-MS and PIXE (polished otoliths), methods like gill raker counts and microsatellites in DNA have been applied [8–13].

2. Experimental

2.1. PIXE analysis

The otoliths from the whitefish were analyzed with particle induced X-ray emission spectrometry (PIXE). The irradiations were performed in air with a 3 MeV collimated proton beam from the Åbo Akademi MGC-20 cyclotron [13]. The diameter of the extracted proton beam was 0.5 mm and the beam current was about 2 nA. The integrated charge at the sample needed for quantification was obtained by measuring the light emission from nitrogen molecules in air during irradiation [14]. The emitted X-rays were measured with an intrinsic germanium planar detector. The GUPIX software package was used to calculate the elemental concentrations from the obtained spectra [15]. The calibration was evaluated by analyzing the following certified reference materials: Limestone, Argillaceous, (SRM 1d) from NIST, and FEBS-1 (Otolith certified reference material for trace metals) and CACB-1 (calcium carbonate), both from NRCC, Canada.

2.2. Otoliths from juveniles raised in hatcheries – building a database

Juvenile whitefish were collected in 2015 and 2016 from 6 hatcheries in Finland - 5 fresh water ponds and one at sea – where they had spent the whole life in the same water habitat (Fig. 1). Otoliths from these juveniles are therefore considered to have an even strontium distribution and can be analyzed without advanced sample preparation. The otoliths were extracted from the fish and rinsed in distilled water. After drying the otoliths were glued onto a polycarbonate backing. Only a slight polishing of the surface was performed before the proton irradiation. A single spot was irradiated on the polished surface of these otoliths (Table 1). The weight of the juveniles was around 5 g and the length about 100 mm. The otolith weight was around 2 mg.

2.3. Otoliths from sea spawning populations – only small differences in strontium concentration

For comparison, whitefish was also sampled at sea along the Finnish west-coast and around the Ålands Islands during spawning (2012–2016). As these sea spawners only migrate short distances [16], the distribution of strontium in the otoliths should not show large variation between the annual rings. Only a small decrease in otolith strontium in the south-north direction of the Gulf of Bothnia is suspected in these otoliths due to the small latitudinal decrease in salinity [10]. After the extraction of the otoliths, they were glued to a backing and slightly polished to obtain a smooth surface, but not exposing the core, before irradiation. There was no effect in the strontium concentrations due to the variation of salinity between the sampling sites (Table 2).

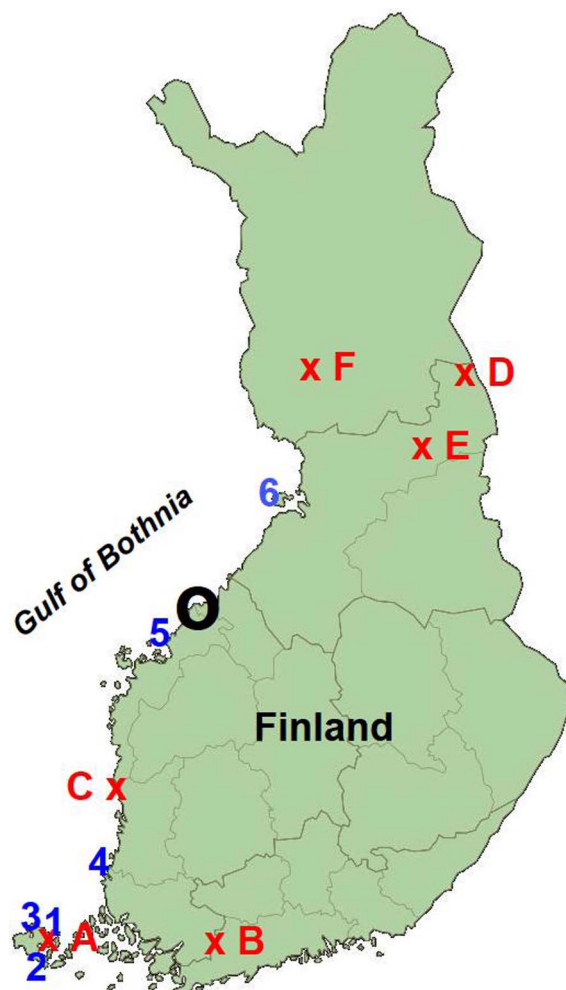


Fig. 1. Whitefish hatcheries in Finland from where juvenile otoliths were obtained (A–F) and sites where local sea spawning whitefish were sampled (1–6). The juveniles were raised in freshwater ponds before stocking (Table 1) with the exception for the juveniles from Merikarvia (site C) which were hatched in freshwater but raised in the sea.

2.4. Otoliths from whitefish migrating upstream in the Torne river

Torne River is the most important spawning river for whitefish in Finland and was therefore included in this study. The whitefish was captured on their way to the spawning grounds in the river in October 2013. Otoliths from the adult fishes were prepared for the analysis by embedding the otoliths in epoxy resin followed by grinding and polishing until the core region was exposed. The polished otoliths were mounted on glass backings. Two spots were irradiated on each otolith (Table 3); one in the core region of the otolith reflecting the water conditions at the breeding location and one close to the edge, clearly outside the first annual ring, reflecting the water conditions at the feeding grounds (on the dorsal side about 1.5 mm from centre).

2.5. Wild or stocked fish – looking into the otolith core of adult fish

To evaluate the relevance of the database, whitefish with unknown provenances was sampled in the sea at the Larsmo archipelago on the Finnish west coast (black circle in Fig. 1). The mature whitefish was caught at their spawning site in October 2016. Otoliths from the adult fish were prepared and polished in a similar way as the otoliths from the river spawning whitefish. Two spots were irradiated on each otolith (Table 4); one in the core region of the otolith and one close to the edge (on the dorsal side about 1.3 mm from centre, Fig. 2).

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