NOT TO BE CITED WITHOUT PRIOR
REFERENCE TO THE AUTHOR(S)

Northwest Atlantic Fisheries Organization

Serial No. N4488 **NAFO SCR Doc. 01/100**

SCIENTIFIC COUNCIL MEETING – SEPTEMBER 2001
(Deep-sea Fisheries Symposium – Poster)

Otolith Microchemistry as a Means of Identifying Stocks of Deep-water Demersal Fishes (OTOMIC)

by

J.D.M. Gordon[a], S.C. Swan[a], A.J. Geffen[b] and B. Morales-Nin[c]

[a] Scottish Association for Marine Science, Dunstaffnage Marine Laboratory, Oban, Argyll, Scotland, PA34 4AD. E-mail jdmg@dml.ac.uk or scs@dml.ac.uk
[b] University of Liverpool, Port Erin Marine Laboratory, Breakwater Road, Port Erin, Isle of Man, UK, IM9 6JA.
[c] Department of Natural Resources, IMEDEA, Miguel Marqués 21, 07190 Esporles, Spain.

Abstract

The deep-water fisheries of the North Atlantic and the Mediterranean have developed rapidly as traditional shelf fisheries decline. Most of the currently exploited species have very wide geographical ranges, which may place them within one or more of the designated management areas such as ICES, NEAFC and the Mediterranean. Many are also caught in international waters and come within the remit of 'straddling stocks and highly migratory species'. The assessment and management of these fisheries has many problems, including a lack of knowledge on stock identity. This problem is compounded because catch and effort data tend to be reported by statistical areas that were devised for shelf fisheries.

If a fish lives in one or more water masses during its life then its otoliths will carry a chemical signature representative of these water masses. This chemical signature has been used to discriminate between stocks of shelf fishes. The objective of the OTOMIC project (EC FAIR 98/4365) is to investigate the use of otolith microchemistry for stock discrimination in four deep-water fish species of the Atlantic and Mediterranean. Three of the species, the smooth grenadier (*Nezumia aequalis*), the bluemouth (*Helicolenus dactylopterus*) and the hake (*Merluccius merluccius*) are present in both the Atlantic and Mediterranean and have differing depth ranges and life history characteristics. The fourth species, the roundnose grenadier (*Coryphaenoides rupestris*), is widely distributed throughout the North Atlantic. Elemental composition of the otoliths was determined using solution-based and laser ICP-MS. This poster describes some of the preliminary results of the project.

Introduction

Fisheries for the deep-water fishes of the continental slopes and ridges in the North Atlantic and the Mediterranean are becoming increasingly important, as pressure on the stocks of the continental shelf increases. The development of deep-water fisheries has, in most cases, been extremely rapid and very little is known about the biology and life histories of these species. Many that are currently being exploited have very wide geographical ranges or are caught in international waters. At the present time, the exploitation of most deep-water fishes is unregulated. For the future assessment and management of the stocks, knowledge of stock identity is essential.

The objective of this project is to assess the use of otolith microchemistry as a possible tool for defining the stocks of different deep-water fish species of the Atlantic and Mediterranean. This method relies on the assumption that otoliths incorporate elements from the environment throughout the life of the fish and are metabolically inert. If a species lives and grows in a discrete area, it is reasonable to suppose that the elemental signature in its otolith would be distinct. Alternatively, if the species has its early life in one area or depth zone and moves to another later in its life cycle, then it should be possible to distinguish changes in the elemental signature in samples from different parts of the otolith. Three of the four species chosen for this study have a commercial value and have been selected to provide examples most likely to have distinctive elemental signatures. They represent a wide range of habitats and life history characteristics and it is hoped that some of the processes of element incorporation might be elucidated.

Study Areas and Species

The deep-water environments of the Atlantic and Mediterranean provide some interesting contrasts. Both areas are comprised of a complex layering of different water masses and each water mass has a characteristic chemical signal. Both areas have seasonal and permanent thermoclines, but below the thermocline the temperature regime is markedly different. In the Mediterranean the temperature remains at about 13^{o} C, whereas in the Atlantic temperature generally decreases steadily with depth. These differing temperature profiles could mediate the uptake of trace elements into otoliths. There is an interchange of water through the Straits of Gibraltar. High salinity Mediterranean outflow, with its characteristic density, extends as a plume into the Atlantic at about 1000 m depth. One consequence of Atlantic inflow into the Mediterranean is that it creates a series of fronts in the Balearic region associated with local topography. The Algerian Basin acts as a reservoir for water of Atlantic origin and its influence extends to the Balearic Slope. Figures 1 and 2 show the sampling locations for all four of the study species.

European hake (*Merluccius merluccius*) is an important commercial species in the Atlantic and the Mediterranean and the stocks are considered to be seriously over-exploited. It is of interest to this study because it is generally considered to be both an outer shelf and an upper slope species. The bluemouth (*Helicolenus dactylopterus*) is a scorpaenid fish that is widely distributed in the eastern Atlantic and the Mediterranean. It is an important commercial species in southern Europe, the Mediterranean and at the Azores. It is also landed as a bycatch of the deep-water trawl fisheries of northern Europe. The smooth grenadier (*Nezumia aequalis*) is a small macrourid fish that has a wide distribution in the eastern North Atlantic and the Mediterranean. It is of small adult size and hence is of no direct commercial importance. However, it can be locally abundant at mid-slope depths and is probably an important component of the food chain. Roundnose grenadier (*Coryphaenoides rupestris*) is an important commercial species, which has a range extending throughout the whole of the North Atlantic, but is not present in the Mediterranean. Although this species is rarely caught in the colder waters of the Nordic Sea, small populations exist in the Skagerrak and in some Norwegian fjords. In addition, roundnose grenadier has a very wide depth range from about 500 to 2000 m and ontogenetic changes in depth distribution have been reported for some areas. Spawning has been reported from all areas, including the Skagerrak. Generally, deep-water fish species of the Mediterranean tend to be smaller than their Atlantic counterparts.

Materials and Methods

Otoliths are composed of about 96% calcium carbonate by weight. The remainder consists of about 3-4% organic matrix and <1% non-organic trace impurities. With the exception of strontium, the minor elements (>100 ppm) such as sodium and potassium are likely to be under physiological regulation via the endolymph, which bathes the otolith. This does not appear to be the case for the trace elements. It is these trace elements, which may provide a useful record of the environment to which the fish was exposed (Campana, 1999).

Elemental concentrations in dissolved otoliths were determined using a VG Elemental Plasma Quad 3 (S-Option) Inductively Coupled Plasma Mass Spectrometer (ICP-MS). Using ICP-MS, it is possible to measure the concentration (to parts per billion) of a large number of elements simultaneously. The otolith solutions were analysed at two separate dilutions (200 000 times for calcium and strontium, and approximately 1000 times for all other elements). The method of standard addition was used to minimize possible matrix effects. Data from the analysis of whole otoliths were analyzed using discriminant analysis, which tests whether the assumed membership of an otolith sample to a categorical group (area) is justified, using some explanatory variables (element concentration). Significant effects of fish length on element concentration were taken into account prior to analysis.

Laser-ablation ICP-MS was used to examine a continuous line from the ventral to the dorsal axis of sectioned roundnose grenadier otoliths. All element counts were ratioed to calcium, which takes into account the varying amounts of otolith material ablated by the laser.

Results

European hake

The total length (cm) ranges of the fish sampled for this analysis are shown in Figure 3. Figure 4 shows a scatterplot of the first two discriminant scores based on the concentrations of 12 elements. There was considerable overlap in element concentrations between the Atlantic and Mediterranean groups, causing some misclassification of samples. As a result of this, statistical analyses were carried out for these two areas separately. Romsdal Fjord samples were characterized by higher concentrations of lithium, rubidium, strontium and yttrium. Higher concentrations of magnesium and manganese, and lower concentrations of rubidium, strontium and yttrium were found in Gulf of Lions samples. Lithium and aluminium levels were higher in Catalan Slope samples. Rubidium, strontium and yttrium levels were higher in Balearic Slope samples. Catalan Slope samples were more similar to those from the Balearic Slope than the Gulf of Lions.

Bluemouth

Figure 5 shows the total length (cm) ranges of the fish sampled. Figure 6 shows a scatterplot of the first two discriminant scores based on the concentrations of 19 elements. As with hake, there was some overlap in element concentrations between the Atlantic and Mediterranean groups. Lithium and rubidium concentrations were high in both Catalan Slope and Hebridean Terrace samples. Hebridean Terrace samples also had higher levels of chromium. Alboran Sea samples had higher concentrations of strontium, yttrium and barium. The most similar groups were Portuguese Slope (N) and the Azores, the most dissimilar were Portuguese Slope (N) and the Catalan Slope.

Smooth grenadier

The pre-anal length (cm) ranges of the fish sampled are shown in Figure 7. Figure 8 shows a scatterplot of the first two discriminant scores based on the concentrations of 18 elements. High concentrations of lithium, aluminium, zinc and cerium were found in samples from both the Balearic Slope and the Reykjanes Ridge. Reykjanes Ridge samples had the highest levels of chromium, yttrium and strontium. Balearic Slope samples were found to be high in manganese, rubidium and lead.

Roundnose grenadier

Figure 9 shows the pre-anal fin length (cm) ranges of the fish sampled for this analysis. Figure 10 shows a scatterplot of the first two discriminant scores based on the concentrations of 11 elements. The discriminant analysis showed a good separation between the samples from the Norwegian fjords and the Skagerrak, and those from all other areas. The Norwegian samples were characterized by having the lowest concentrations of barium and highest levels of rubidium, strontium and yttrium. Skagerrak samples had the highest concentrations of lithium and Storfjord samples were high in manganese. Samples from the Mid-Atlantic Ridge and Porcupine Seabight were most similar.

Figure 11 show manganese: calcium profiles across two roundnose grenadier otoliths obtained from sample sites in Storfjord and the Rockall Trough. The otoliths were extracted from fish of a similar size and with an approximate estimated age of 20 to 25 years. In the Storfjord sample, manganese concentration increased towards the edge of the otolith, whereas it was concentrated in the otolith core for the Rockall Trough sample.

Discussion

It was not possible to obtain fish samples of a similar size range for individual species from all areas. Some significant length effects were identified using analysis of covariance (ANCOVA), with fish length as the covariate. These were corrected for, by using the common linear slope (Edmonds *et al*., 1989) prior to discriminant analysis. However, for smooth grenadier, only small fish were obtained from the Balearic Slope area and it is difficult to ascertain to what extent this causes the separation of these samples from other Mediterranean samples.

The results obtained from the laser-ablation ICP-MS analysis of sectioned roundnose grenadier otoliths showed very different profiles for manganese across the sections. These differences were reflected in the results obtained using solution-based ICP-MS for this species. Levels of manganese were high in the Storfjord samples compared to all other areas and the concentrations were lowest in the otolith core, when the fish would have been higher up in the water column. These high concentrations may be a result of manganese cycling in a fjordic environment.

Conclusions

Laser-ablation ICP-MS analysis of otolith sections indicated major differences in the concentrations of some elements, depending on the region of the otolith under examination. Elemental differences in the otoliths of a single species sampled at a variety of locations were also reflected in the results obtained from solution-based ICP-MS analysis of whole otoliths. The results indicate that otolith microchemistry may have some value for stock discrimination in deep-water fish species. Ongoing research using laser-ablation ICP-MS analysis of otolith cores may enhance the value of the technique.

Acknowledgements

The authors acknowledge the geochemistry group at SAMS for their guidance in the interpretation of the ICP-MS results.

References

CAMPANA, S. E., 1999. Chemistry and composition of fish otoliths: pathways, mechanisms and applications. *Marine Ecology Progress Series*, **188**: 263-297.

EDMONDS, J. S., M. J. MORAN, N. CAPUTI, and M. MORITA. 1989. Trace element analysis of fish sagittae as an aid to stock identification: pink snapper (*Chrysophrys auratus*) in Western Australian waters. *Canadian Journal of Fisheries and Aquatic Science*, **46**: 50-54.

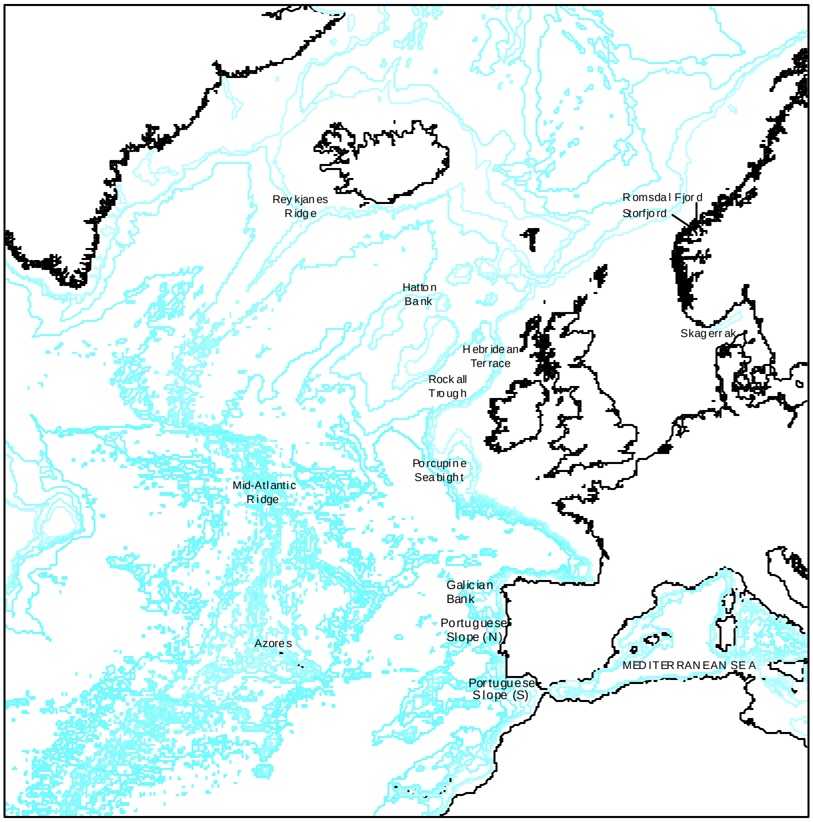


FIGURE 1. Location of sites throughout the North East Atlantic from which samples were obtained.

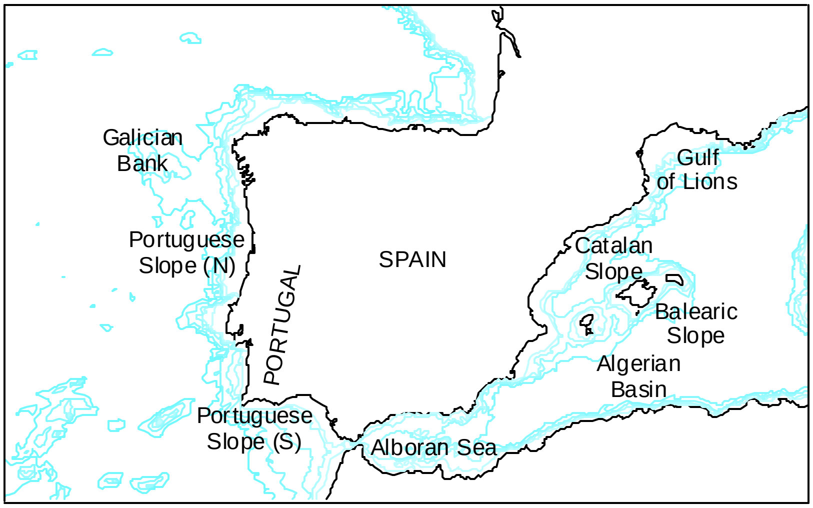


FIGURE 2. Location of sites throughout the Mediterranean from which samples were obtained.

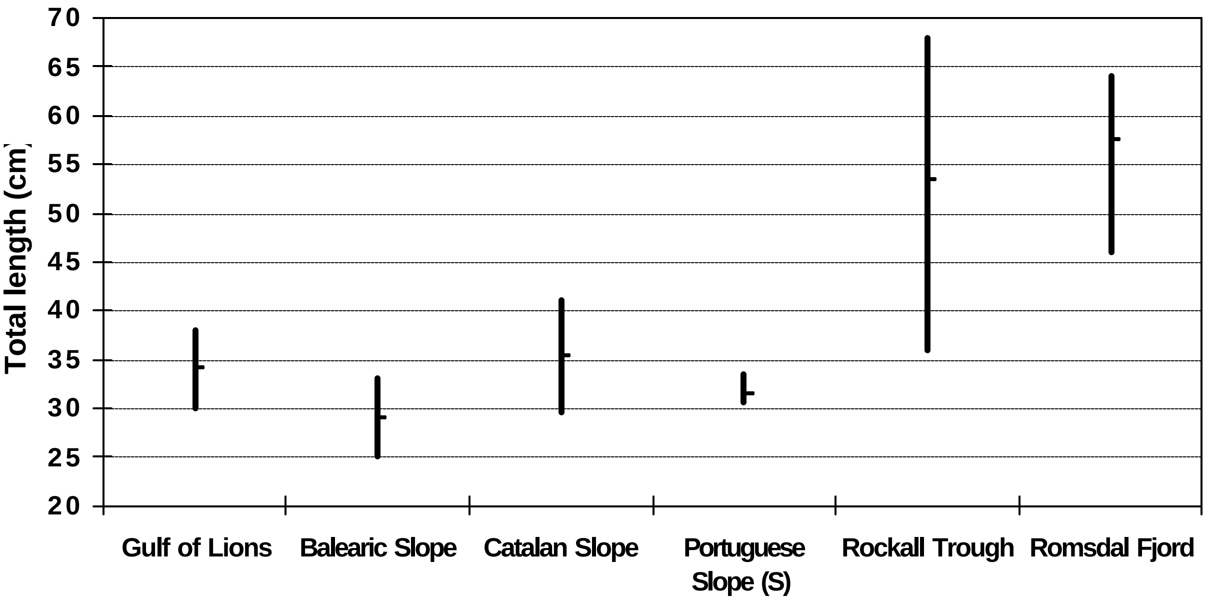


FIGURE 3. Length ranges and mean total lengths of hake samples by area.

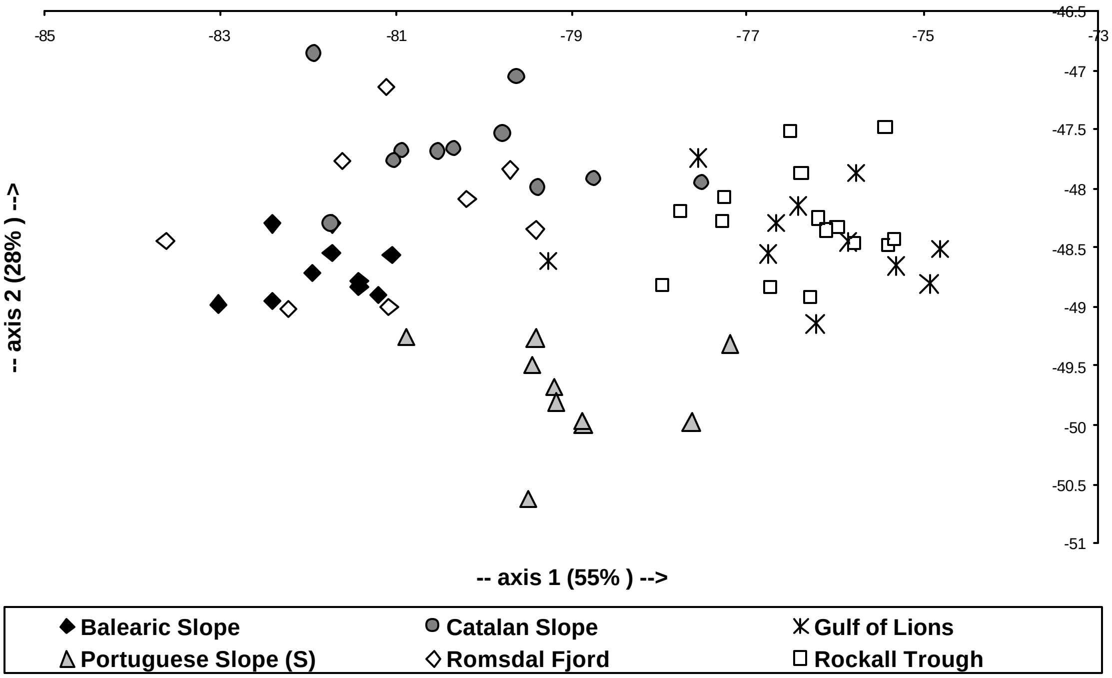


FIGURE 4. Discrimination between hake otolith samples based on the concentrations of 12 elements.

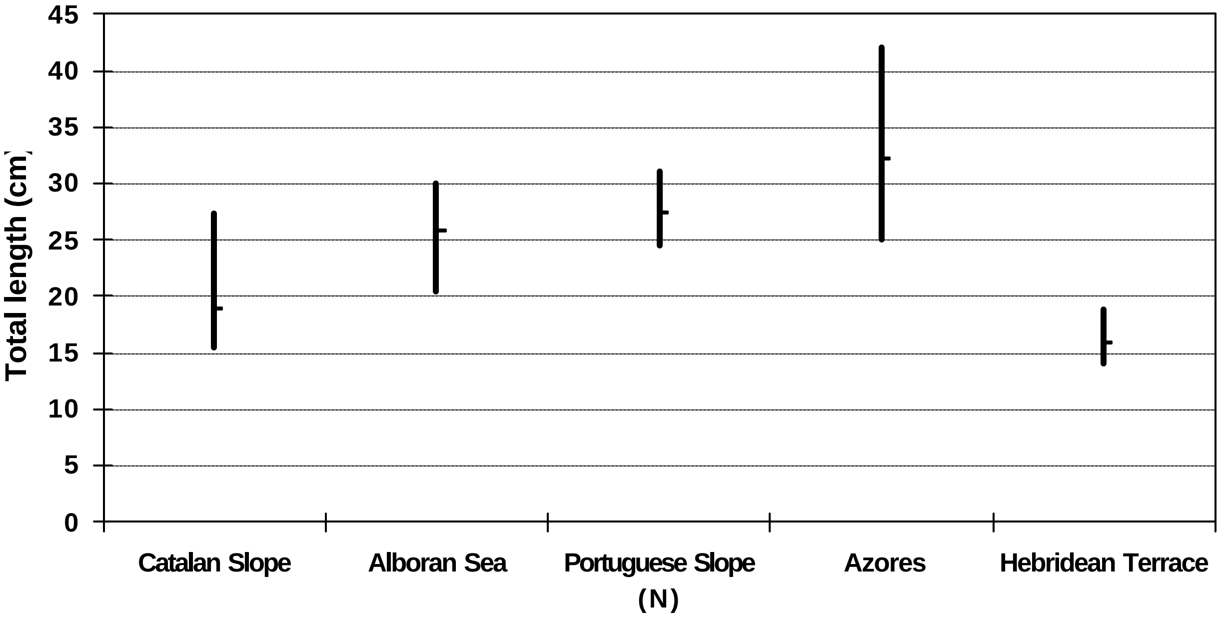


FIGURE 5. Length ranges and mean total lengths of bluemouth samples by area.

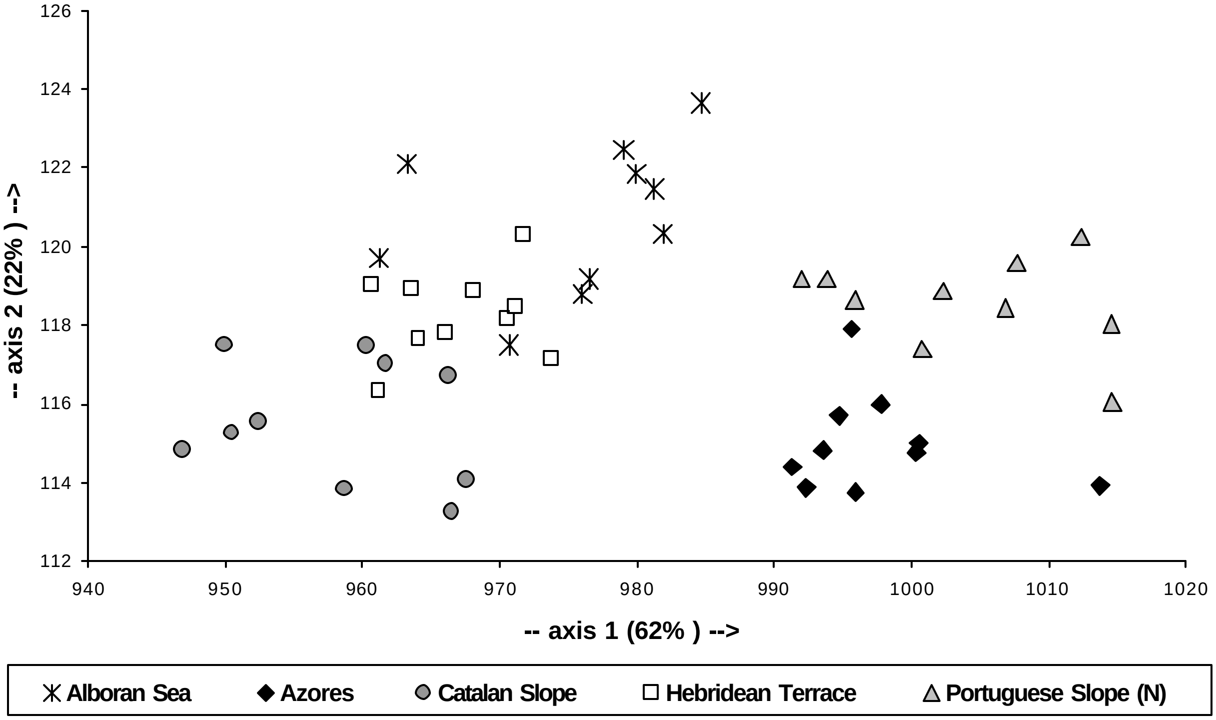


FIGURE 6. Discrimination between bluemouth otolith samples based on the concentrations of 19 elements.

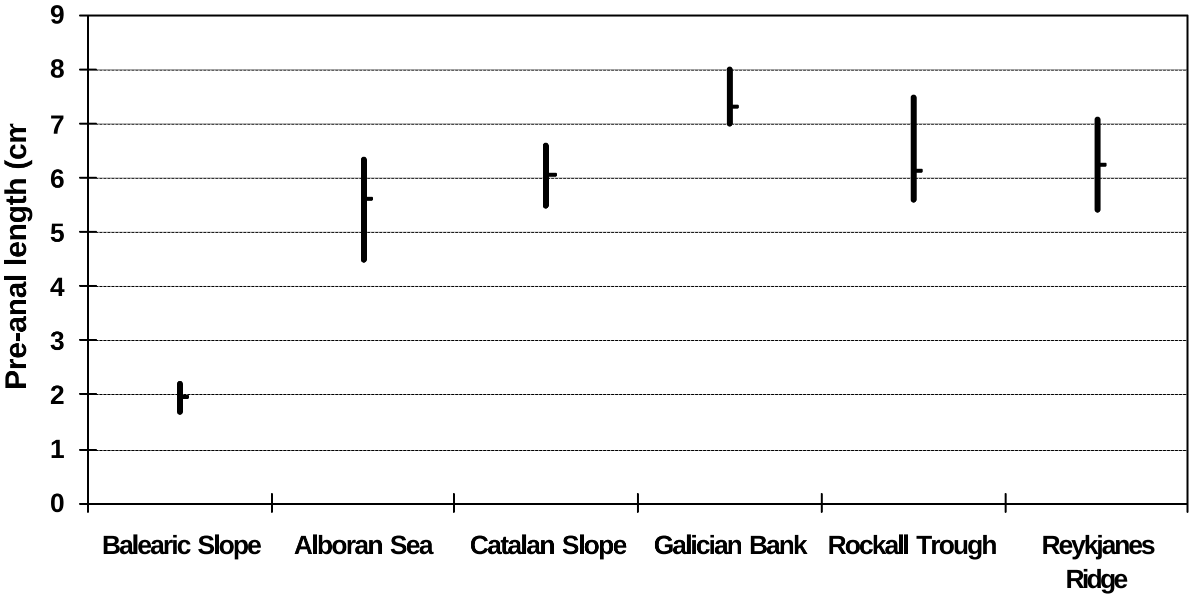


FIGURE 7. Length ranges and mean pre-anal lengths of smooth grenadier samples by area.

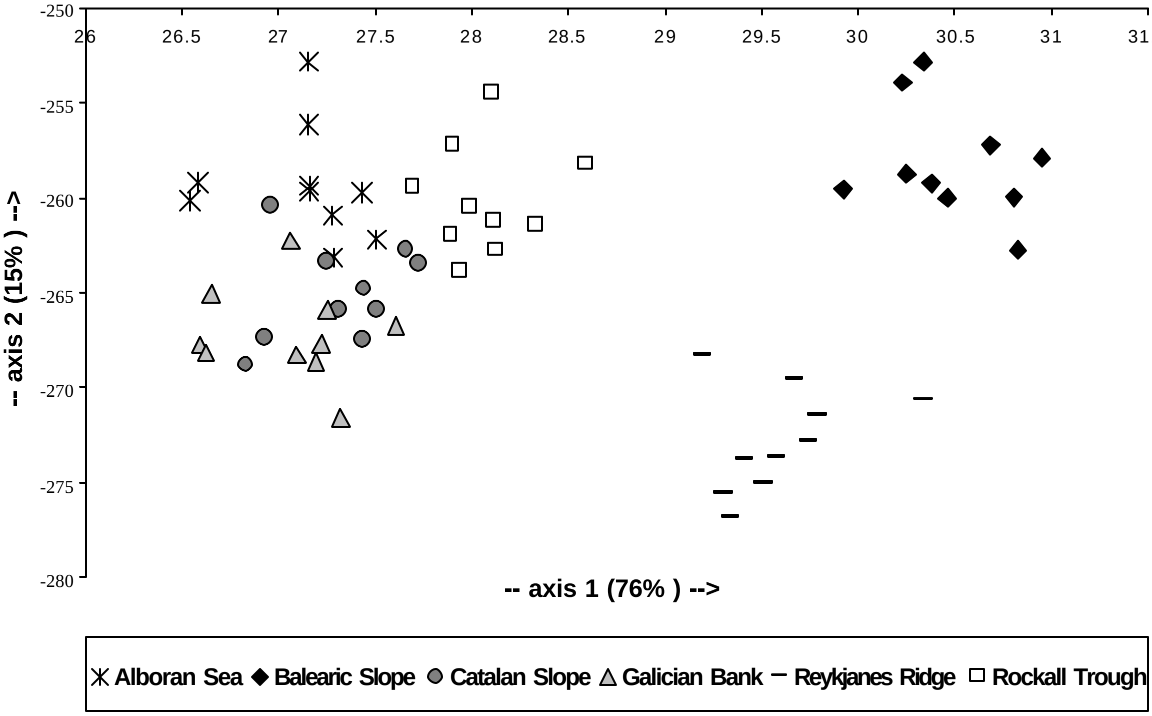


FIGURE 8. Discrimination between smooth grenadier otolith samples based on the concentrations of 18 elements.

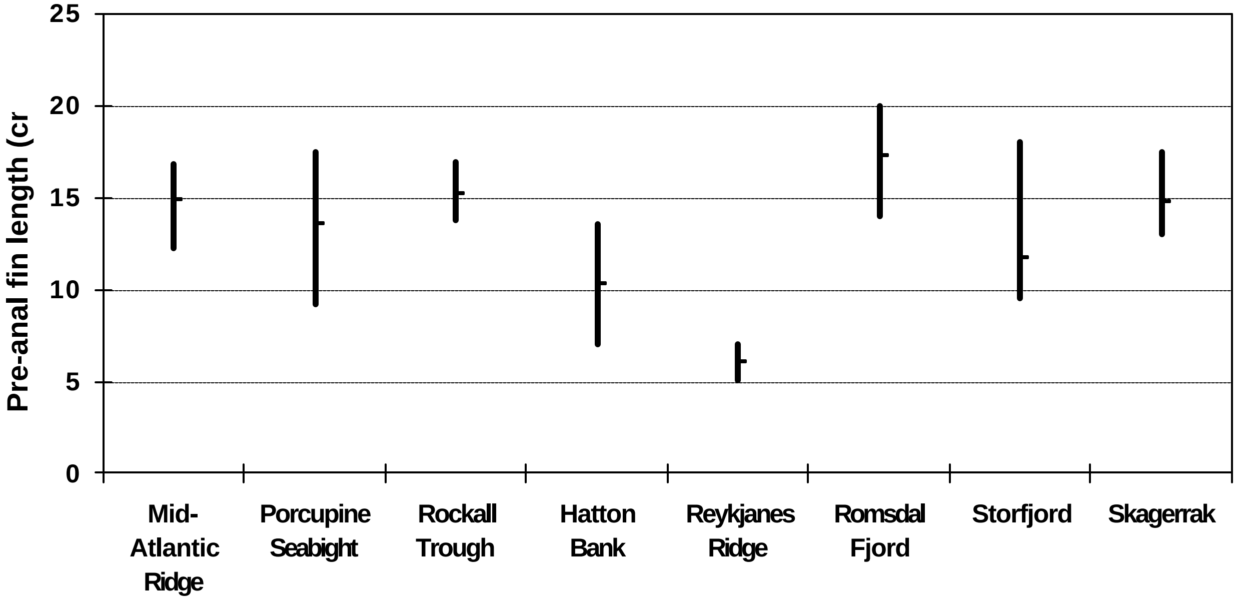


FIGURE 9. Length ranges and mean pre-anal fin lengths of roundnose grenadier samples by area.

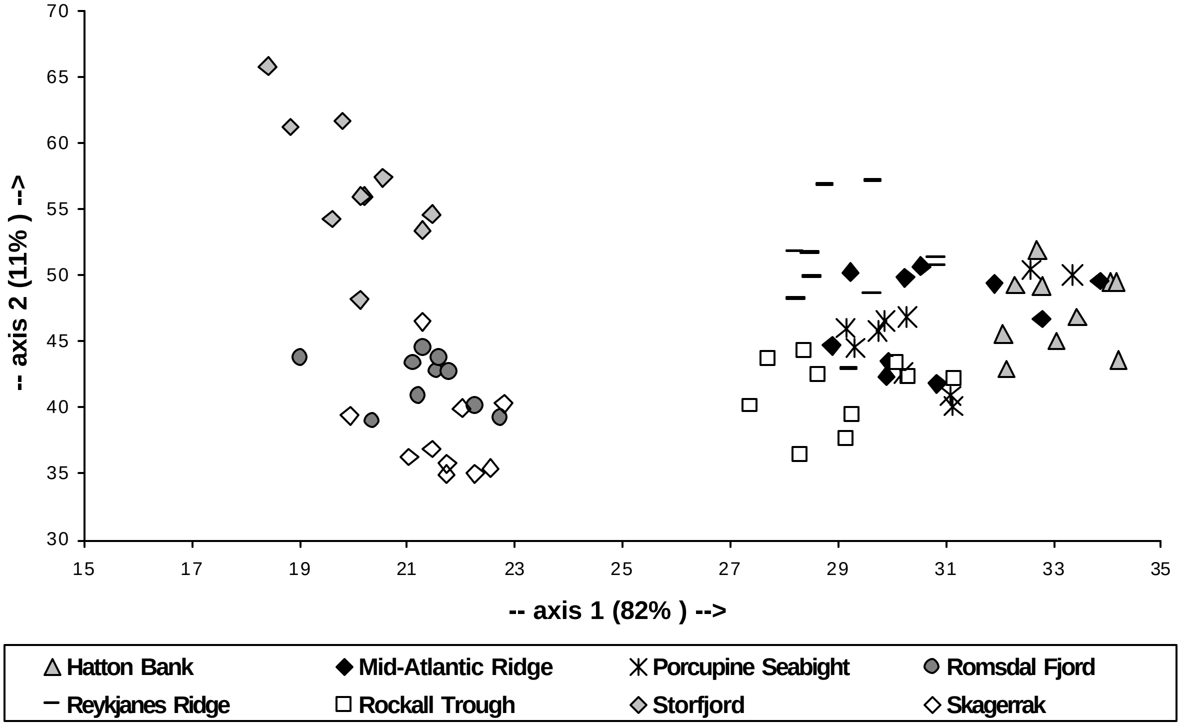
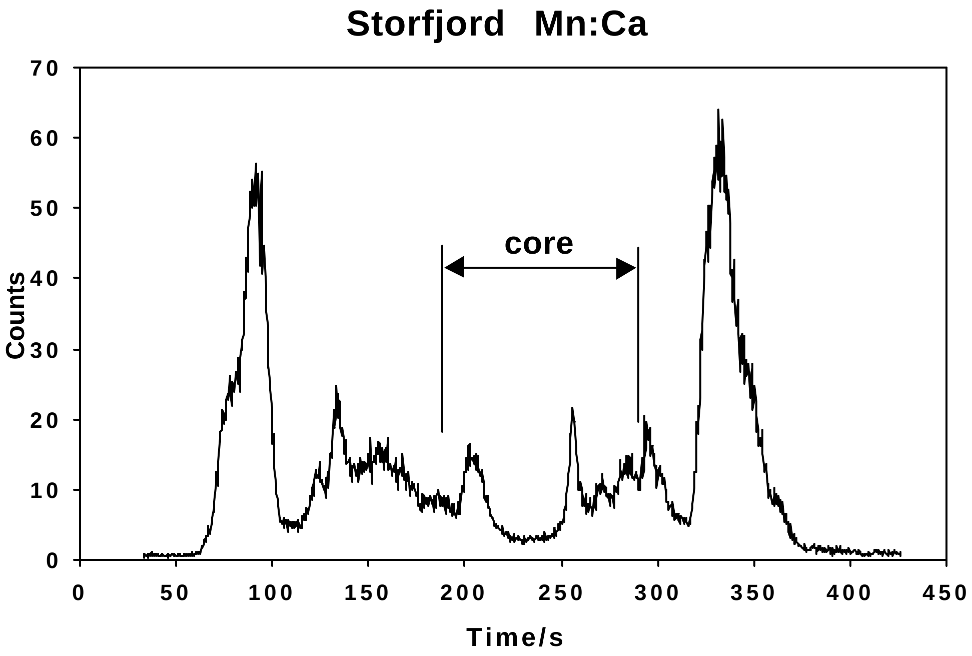


FIGURE 10. Discrimination between roundnose grenadier otolith samples based on the concentrations of 11 elements.



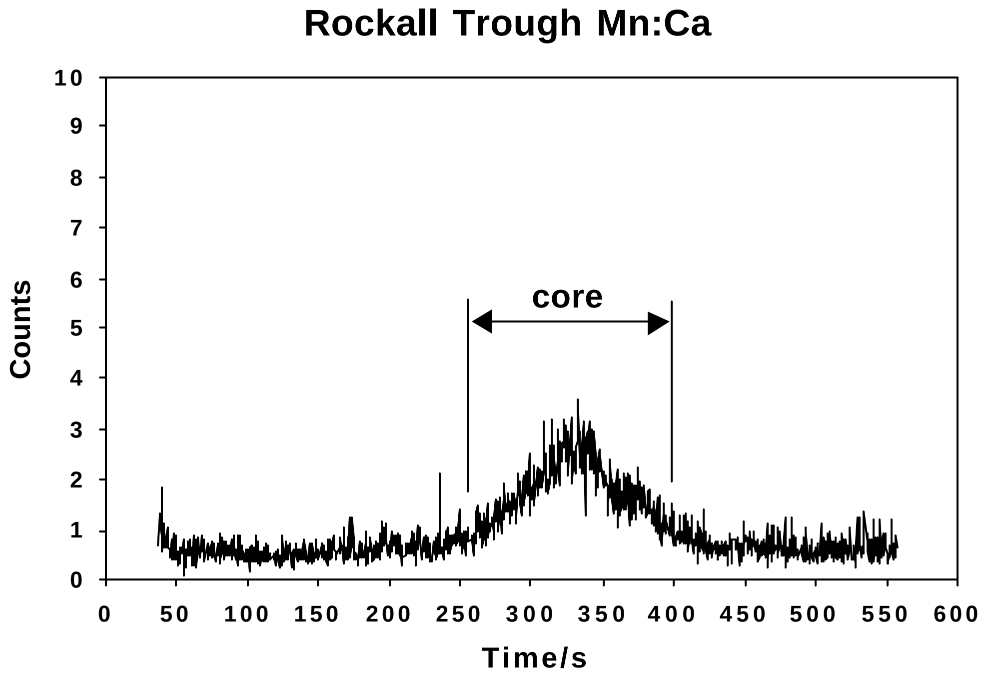


FIGURE 11. Variation in manganese concentration across roundnose grenadier sectioned otoliths.