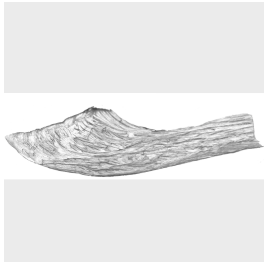
Mar 20, 2019 Version 1

Growth Incremental analysis of thins sections from marine molluscs V.1



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Protocol status: Working
We use this protocol and it's working

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Keywords: Ostrea edulis, Incremental growth-line analysis, Seasonality, ecological baseline, laser induced breakdown spectroscopy, geochemical data, structural context for the geochemical data, marine ecologies through time, marine ecology, growth incremental analysis of thins section, elemental map, geochemical pattern, marine mollusc, elemental maps with the result, sustainable marine resource, isotopic analysis, marine mollusc shell, overall robustness of palaeoecological interpretation, growth pattern, palaeoecological interpretation, annual growth pattern, annual growth increment, prehistoric seasonality practice, repeating growth pattern

Abstract

Annual growth patterns in marine mollusc shells are valuable indicators of the condition of marine ecologies through time. In archaeological contexts, the mollusc's time of death (i.e. the last season of growth) is an indicator of human exploitation patterns throughout the year, enabling the reconstruction of when and how often gathering occured as well as when sites were occupied. Both pieces of information, growth rate and season of death, are vital for understanding exploitation pressure in the past, and building baselines for modern environmental policies that secure sustainable marine resources. Previously, these parameters have been determined by incremental growth-line or isotopic analyses, which are time consuming and often expensive techniques, thus restricting sample size and the overall robustness of palaeoecological interpretations.
Here, we apply Laser Induced Breakdown Spectroscopy (LIBS) to produce elemental maps (Mg/Ca) with the potential to trace and display growth patterns quickly, and at a reduced cost. We further compare the elemental maps with the results of incremental growth-line analysis to provide a structural context for the geochemical data, and demonstrate the utility of an integrated methodological approach.
Our study was undertaken on 12 European oysters (Ostrea edulis) from the Late Mesolithic shell midden at Conors Island, Co. Sligo in the Republic of Ireland. Our LIBS analysis enabled us to accurately and quickly determine repeating growth patterns, which were in agreement with the annual growth increments visible through the microscopic analysis. Based on this comparative dataset, including structural and geochemical patterns, the Late Mesolithic site of Conors Island had been occupied throughout the year. Moreover, our analyses highlight the applicability of LIBS to determine prehistoric seasonality practices as well as biological age and growth at an improved rate and reduced cost than was previously achievable.

Materials

STEP MATERIALS

- Ethanol
- Release Agent **Buehler Catalog #**20-8185-016
- Epoxy Resin **Buehler Catalog #**No 20-8140-12B
- Epoxy Hardener **Buehler Catalog #**20-8142-016
- Polycrystalline suspension **Buehler Catalog #**40-6016
- Adhesive **Catalog #**n/a

Protocol materials

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Producing thin sections

1

Equipment

ISOMET 1000 Precision Saw	NAME
Saw	TYPE
Buehler	BRAND
11-2180	SKU
Using a Buehler Diamond Wafering Blade (Series 15LC Diamond No. 11-4276)	SPECIFICATIONS

Section oysters following Milner (2001) and using a precision saw with a diamond blade. First section is at the hinge and in direction of growth, the second (optional) section is done to remove unnecessarily long portions of the shell section.

Milner, N., 2001. At the Cutting Edge: Using Thin Sectioning to Determine Season of Death of the European Oyster, Ostrea edulis. Journal of archaeological science, 28(8), pp.861–873.

2 Clean cylindric plastic moulds with
Ethanol
leave to dry for 00:05:00. Then coat the inside with
Release Agent **Buehler Catalog #**20-8185-016
and dry again for 00:30:00

3

Epoxy Resin **Buehler Catalog #**No 20-8140-12B

Place sectioned shells face down in 1- and 2-inch plastic moulds depending on the size of the cut hinge.
Then add resin (2 parts by volume of Epoxy Resin mixed with 1 part by volume Epoxy Hardener.

Epoxy Hardener **Buehler Catalog #**20-8142-016

Leave to cure in an environment of constant humidity and temperature overnight (or 12:00:00)

4

Equipment

Motorpol 2000	NAME
Polisher	TYPE
Buehler	BRAND
49-7268	SKU

Remove resin block from plastic moulds once hardened, and grind hinge-surface using successively finer metallographic grit papers (P600, P1200 and P2500 grades respectively) on a Buehler Motorpol 2000.

Polycrystalline suspension **Buehler Catalog #**40-6016

Clean the surfaces of the resin blocks with water and polish further using a polishing cloth and polycrystalline suspension of 3µm.
Leave to dry overnight (or 12:00:00)

5

Adhesive **Catalog #**n/a

Attach the polished facets of the resin blocks to a glass slide using Loctite 322 Adhesive, exposed to an ultraviolet light source and leave to harden for 24:00:00 .

6

Equipment

ISOMET 1000 Precision Saw	NAME
Saw	TYPE
Buehler	BRAND
11-2180	SKU
Using a Buehler Diamond Wafering Blade (Series 15LC Diamond No. 11-4276)	SPECIFICATIONS

Once the sample had bonded to the slide, cut the the bodies of the resin blocks from the slide using a Buehler ISOMET 1000 Precision Saw and leave a slice of the embedded hinge of approximately 200 to 500 µm in thickness on the glass slide.

7

Repeat Step 4, iIn order to lightly ground the shell and the resin to produce the final thin section of ~50 µm.