Mediterranean limpets and Mg/Ca ratios

Using LIBS to screen for SST changes and physiological effects

Niklas Hausmann[1,2], **Amy Prendergast**[3*], Demetrios Anglos[1,4]

[1]Institute of Electronic Structure and Laser, Foundation for Research and Technology - Hellas, Heraklion, Greece
[2]BioArCh, University of York, York, United Kingdom
[3]School of Geography, University of Melbourne, Australia
[4]Department of Chemistry, University of Crete, Heraklion, Greece

* Presenting Author

@NiklasHausmann
@AmyLPrendergast

Summary

Question: How can we improve elemental analysis of mollusc shells to acquire climatic information?

Task: 2D-mapping of whole shells to better understand variation of climatic and physiological influences on elemental record. Combine with high-resolution $\delta^{18}O$-values.

Results: Large variability of Mg/Ca ratios between coeval specimens and within growth increments. Intra-specimen correlation of Mg/Ca, $\delta^{18}O$, and Sea Surface Temperature (SST) (R^2>0.8). Physiological effects common but avoidable.

Discussion: Intra-increment variability of Mg/Ca in marine shells is likely very common. LIBS-screening can help to cheaply study limpet records by revealing physiological effects, locating increments grown during annual minimun and maximum temperatures, and determining season of death.

Background and Methods

Element ratios in gastropod shells are more easily acquired than oxygen isotope values, but often provide unreliable results for sea surface temperature (SST) reconstructions and also for determining the mollusc's season of death.

Physiological effects can superimpose environmental controls on the element ratios and thus prevent any interpretation of past environmental conditions.

By mapping whole shells we aimed to better understand how the shell record is skewed and to circumvent physiological effects when they occur (Hausmann et al. 2019).

Fig. 1, *Map of sample locations of modern* Patella caerulea *specimens from: Tunisua, Croatia, Malta, Libya, Greece, Turkey, and Israel.*

We employed Laser Induce Breakdown Spectroscopy (LIBS) to analyse the elemental records of *Patella caerulea* (Mediterranean limpet) shells. These shells were previously analysed by Prendergast and Schöne (2017) to study their growth structure and oxygen isotope composition using high-resolution sampling (30-100 µm). We collected 19 modern shell specimens from 9 locations across the eastern Mediterranean (**Fig. 1**). This wide spread allowed us to assess intra- and interspecimen variability in multiple locations, some of which are of high archaeological interest (Haua Fteah, Knossos).

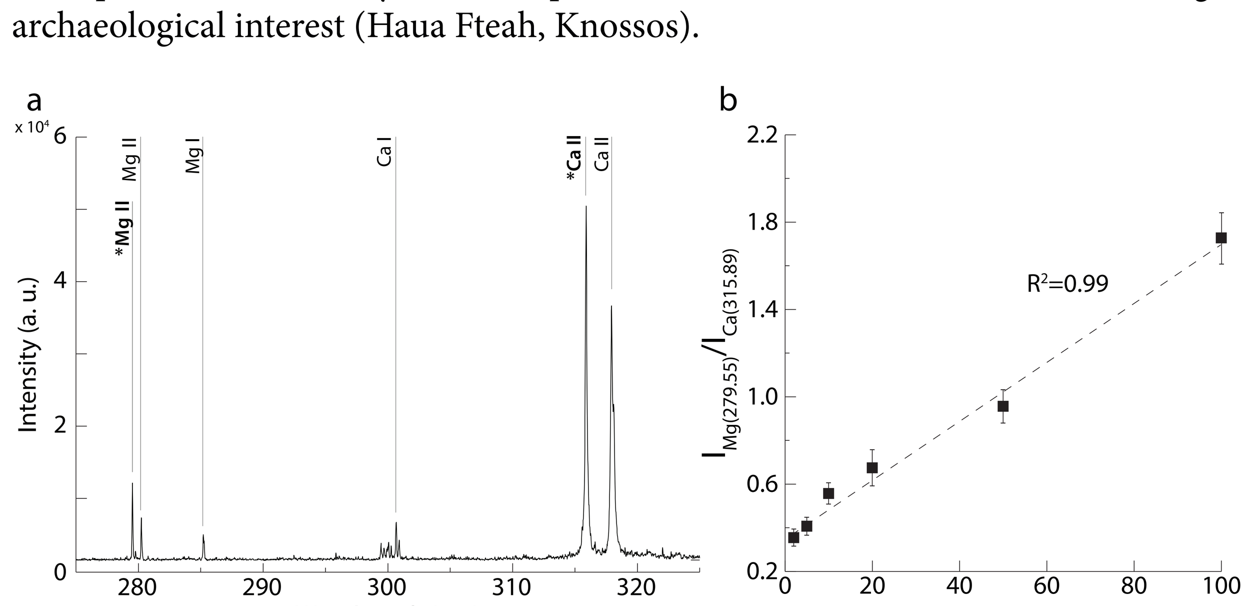


Fig. 2, *a: Single-shot LIBS spectrum of shell sample (measured peaks: Mg II: 279.553 nm and Ca II 315.887 nm in bold and indicated by asterisks). b: Calibration curve of Mg/Ca intensity ratio as a function of Mg/Ca concentration ratio, based on standards with known concentration (Hausmann et al. 2017).*

LIBS is based on the atomic emission spectroscopy of plasma that is generated by focusing a high intensity laser beam onto the sample and creating plasma in a small area (30–50µm). Qualitative and quantitative results of the elemental composition (here Mg and Ca) is gained from the light emitted by the plasma (**Fig.2**).

This approach provides a close to instantaneous measurement (0.01 seconds) that is minimally invasive (0.1 µg). Little sample preparation is necessary: only sectioning, a rough polish (800 grit paper) and cleaning with ethanol. Theses advantages allowed us to quickly generate extensive elemental maps at almost no cost (~€200/year).

Results

2D elemental (Mg/Ca) maps of whole limpet shells (*Patella caerulea*) revealed patterns in the shell sections (**Fig. 3**). We consistently found aragonite layers with distinctly low (purple/dark blue) Mg/Ca ratios than in the calcite layers of *P. caerulea* which were consistently higher in Mg/Ca ratios (green/yellow).

We found repeating patterns of Mg/Ca ratios along the direction of growth and in coeval specimens (Fig. 3, a and b), suggesting an exterior control. We also found in 42% of all specimens a consistent (16%) or inconsistent (26%) increase of Mg/Ca ratios towards the exterior layer (M+3), suggesting a physiological influence that potentially affects the microstructure of this shell layer.

We used the high-resolution oxygen isotope data from Prendergast and Schöne (2017) to compare them with the repeating 'annual' patterns that we found in the Mg/Ca ratios (**Fig. 4**).

Both datasets compared well (R^2>0.8) but when comparing data from different shells no clear correlation was possible (**Fig. 5**).

Partially this has to do with the physiological effects in some shells that increase Mg/Ca ratios towards the exterior (Fig. 3c), but even in specimens lacking these effects, deviations can be found, that prevent a general temperature equation.

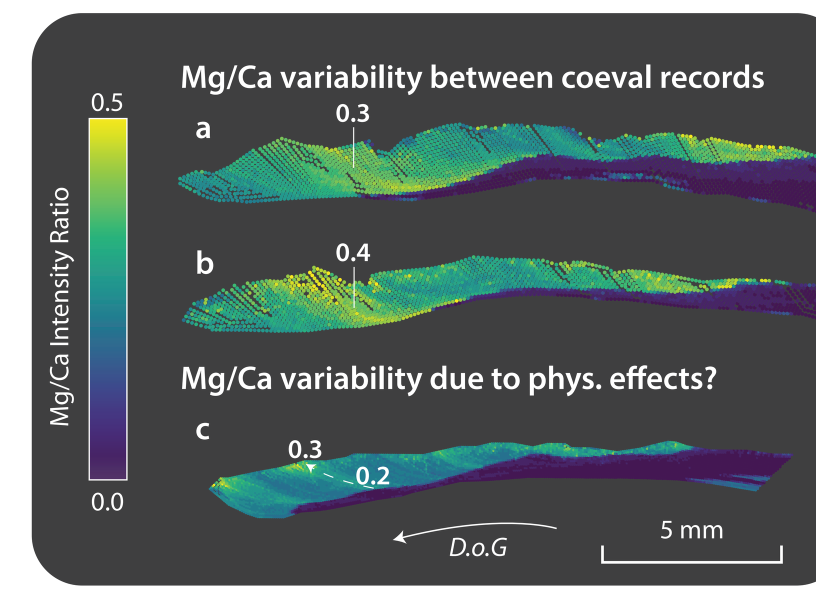


Fig. 3, *Mapped limpet shell sections of Mg/Ca variation (0.3–0.4) between coeval specimens (a,b) and one specimen (c) with increasing Mg/Ca ratio (0.2 to 0.3) towards the exterior of the shell (arrow). D.o.G.: Direction of growth. Variation between records is expected, however below (Fig. 5) we see that these differences are linked to specimen specific ways of how SST-changes are reflected in the Mg/Ca record.*
The increase of the Mg/Ca ratio (c) happens within one increment and indicates a non-environmental influence that is part of a physiological process during biomineralisation. This 'zonation' could be a result of microstructural differences between M+2 and M+3 layers in limpets. Notably, it does not occur in all limpet shells.

Fig. 4, *Comparison of high-resolution $\delta^{18}O$ values (purple) and Mg/Ca ratios (green). Both records compare well and no major deviations or ontogenetic trends are visible in the Mg/Ca record. Note that the values on the y-axis for Mg/Ca are reversed to better compare both datasets.*

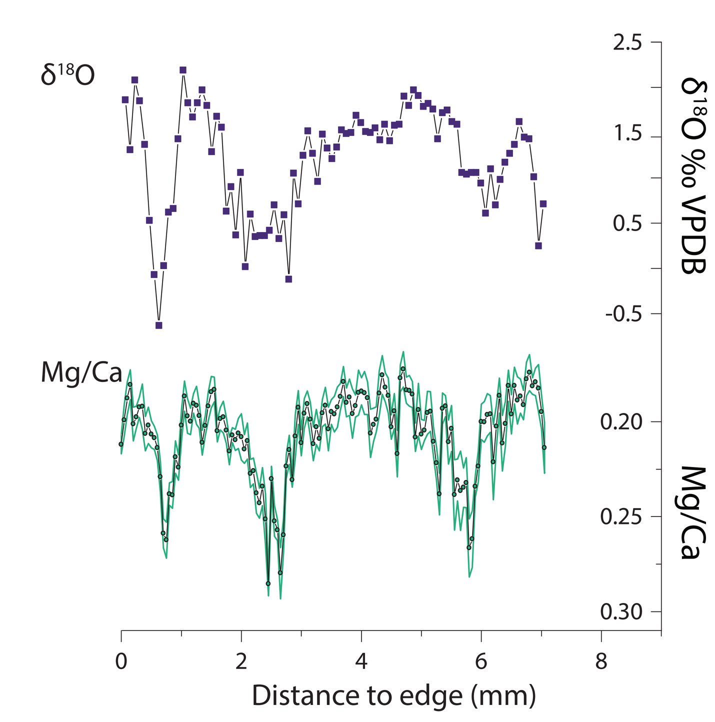
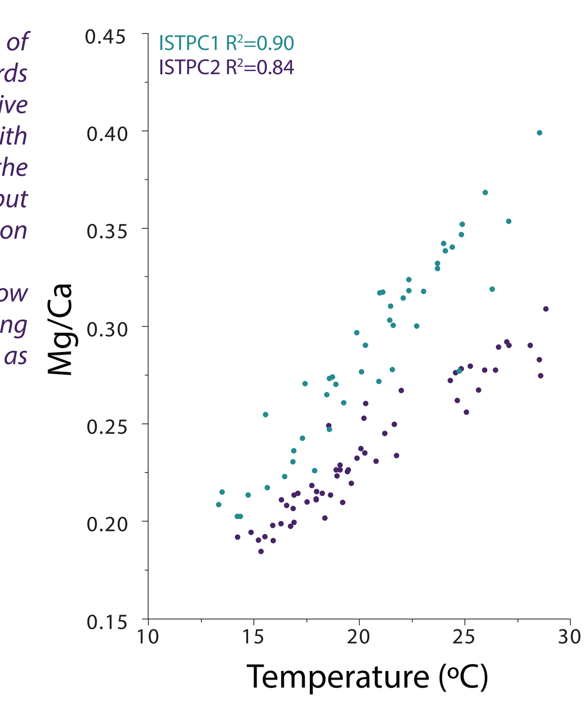


Fig. 5, *Comparison of two coeval shell records and their respective linear correlation with SST. Individually, the correlation is good, but no 'general' equation can be derived.*
See below in Fig. 6 how we solve this by using only a few $\delta^{18}O$ values as anchor points.



Discussion and future Steps

Lacking a temperature equation for *P. caerulea* in general as well as within one locality of the Mediterranean is very problematic. We can determine the season of death but cannot reconstruct absolute temperatures . However, we can use the relative SST change or the 'floating correlations' (Fig. 5) as indicators of which increments were grown during the highest or lowest temperatures and subsequently sample them for oxygen isotope analysis (**Fig. 6**):

Step 1 Screen the specimens using rapid LIBS mapping (e.g. Fig.3) to reveal the locations of annual SST minima and maxima, and whether physiological effects occur.
Step 2 Sample shell carbonate for $\delta^{18}O$-analysis in the locations of interest revealed in Step 1.
Step 3 Calibrate Mg/Ca ratios from line scans (e.g. Fig. 4) using the $\delta^{18}O$-values as 'anchor points'.

Through this approach we gain information about the season of death through LIBS alone and information on annual SST range with as few as two $\delta^{18}O$-values per shell. In the future we hope to use this approach to increase sample numbers and make models of intra-annual SST change more extensive and more robust.

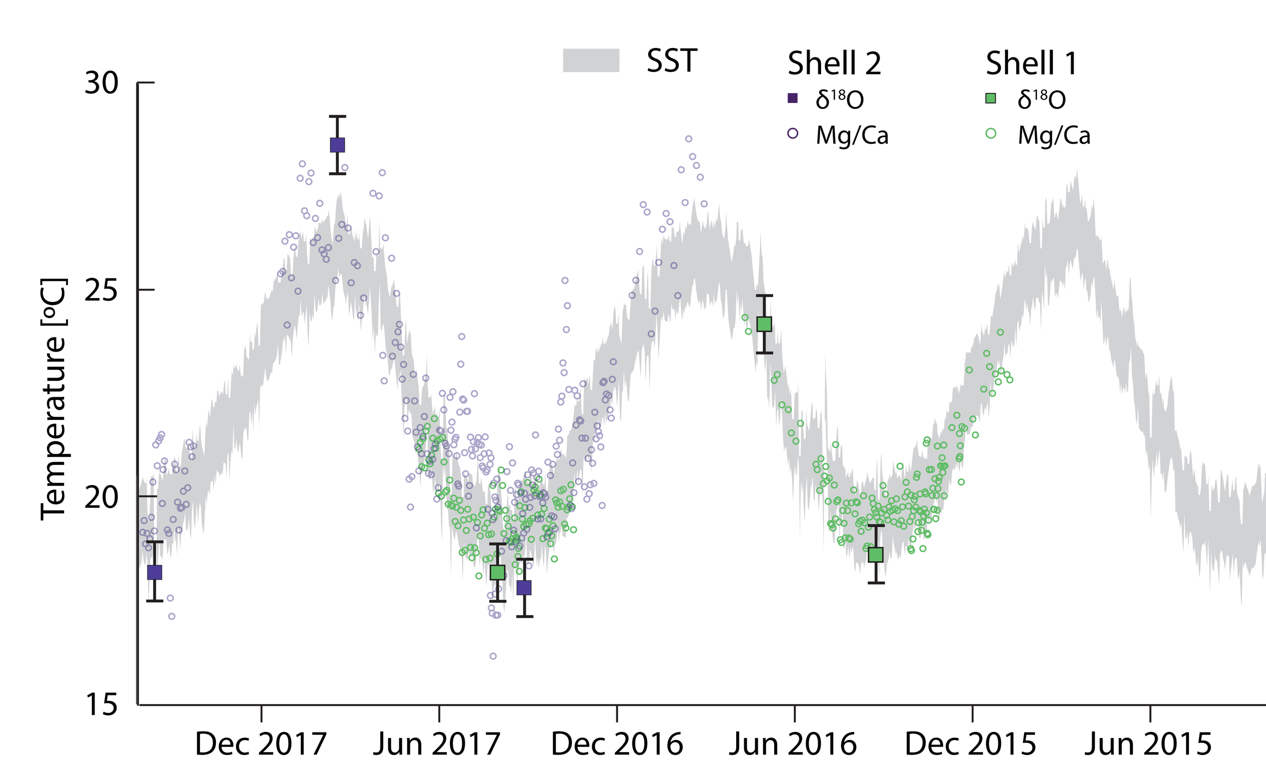


Fig. 6, *Daily Sea Surface Temperature (SST) data from Satellite measurements with SST estimates (circles) based on Mg/Ca on shells of two coeval* P. caerulea *specimens. Previous screenings of Mg/Ca ratios (e.g. Fig. 3) allowed us to locate annual minima and maxima and produce $\delta^{18}O$ values for only these growth increments, to inexpensively assess intra-annual SST ranges.*

References and Acknowledgements

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