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The bivalve *Glycymeris pilosa* as a multidecadal environmental archive for the Adriatic and Mediterranean Seas

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ABSTRACT

We evaluated the potential of *Glycymeris pilosa* as an environmental indicator for the Mediterranean region by applying sclerochronological techniques on a sample set collected from Pašman Channel in the middle Adriatic Sea. Maximal longevity of analyzed shells was 69 years. Growth increments in acetate peels of the hinge region had clear boundaries, and there was a strongly synchronous signal in growth-increment width among individuals. The final, replicated chronology spanned 1969 to 2013. Shell growth negatively correlated with local summer sea temperatures and positively with November precipitation. High correlation between shell growth and circulation patterns in the northern Ionian was also observed, with slower growth occurring during cyclonic regimes. Given its broad distribution in the region and the ability to crossdate, generate annually-resolved chronologies, and of a length that substantially overlaps with observational records, *G. pilosa* has considerable potential to test hypotheses relating to environmental variability and biological response in the Mediterranean.

1. Introduction

Populations of marine organisms face increasing and unprecedented pressure from overfishing, global climate change, eutrophication, coastal degradation, and toxic contamination with implications for ecosystem functioning and food security (García and Rosenberg, 2010; Tacon and Metian, 2013). Of particular concern is understanding historical ranges of physical and biological variability to identify whether climate or ecological processes are exceeding pre-industrial boundaries and to help disentangle human from natural impacts. Indeed, society's desired transition from the management of individual fishery stocks to more holistic ecosystem-based strategies requires knowledge of biotic and abiotic interactions as well as the establishment of ecosystem indicators and their target and threshold values (Levin et al., 2009). These issues cannot be adequately addressed without multidecadal to multicentennial records, yet such time series are lacking, especially in marine systems given logistical and financial demands of acquiring such datasets (Poloczanska et al., 2013).

In the absence of observational records, growth increments in hard parts of marine organisms, provide a "shortcut" to generate continuous, multidecadal time series of growth and marine environmental variability, especially with the application of tree ring (dendrochronology) techniques, which ensure adequate replications and that all data points are placed exactly in time (Matta et al., 2010; Black et al., 2013). Tree rings have long been recognized as archives of environmental variability and change in terrestrial systems. In the same way, hard structures of marine organisms, including bivalve shells, fish otoliths and scales, as well as corals, contain continuous information of the environment experienced over the organism's lifespan (Morrongiello et al., 2012). Over the past decade this field of sclerochronology has been rapidly developing and includes investigating morphological (i.e. increment width) as well as geochemical composition of these structures (Schöne and Gillikin, 2013). A major focus has been directed to extremely long lived bivalves such as *Arctica islandica* (>500 years, e.g. Butler et al., 2013a; Schöne, 2013), *Glycymeris glycymeris* (~200 years, Reynolds et al., 2013), and Pacific geoduck, *Panopea generosa* (Strom et al., 2004; Black et al.,

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2009) which can pre-date instrumental records by orders of magnitude (Schöne and Gillikin, 2013). Bivalve chronologies are comparable in quality to those of tree-ring chronologies and thus can be used to establish climate-biology relationships and also hind-cast environmental variability prior to the start of observational records (Black et al., 2009).

Chronology work is expanding into a number of the world's oceans including the North Atlantic (Wanamaker et al., 2012; Butler et al., 2013a; Schöne, 2013), Southern Ocean (Gillanders et al., 2012), Indian Ocean (Coulson et al., 2014), and North Pacific (Black, 2009). Yet there have been very few sclerochronological studies in the Mediterranean despite a number of possible marine archives including vermetid gastropods (*Dendropoma petraeum*), non-tropical zooxanthellate corals (*Cladocora caespitosa*) and cold-water corals (*Desmophyllum dianthus, Lophelia pertusa* and *Madrepora oculata*) (Montagna et al., 2008). In perhaps the only Mediterranean example from corals, Peirano et al. (2009) constructed a multidecadal growth chronology from a living *Cladocora caespitosa*. Though developed from only one colony, the chronology was negatively related to instrumental records of water and air temperature. In addition to these species, there is also potential for bivalves, as has been documented for *Glycymeris bimaculata*. However, that chronology spans only 16 years (Bušelić et al., 2015) and underscores a need for identifying other suitable species that could be used to develop longer histories. *Glycymeris glycymeris* is one such possibility and has been used to generate multi-decadal and even multi-centennial chronologies in the North Atlantic (Brocas et al., 2013; Reynolds et al., 2013; Royer et al., 2013). There are records of *G. glycymeris* distribution in the Adriatic Sea (e.g. Legac and Hrs-Brenko, 1999; Peharda et al., 2010), and in some other parts of the Mediterranean, but those populations are almost certainly the very closely related *Glycymeris pilosa* (Purroy et al., personal communication), which is distributed from the Western Sahara, Mauritania, Madeira and Canaries Archipelagoes and in the Mediterranean Sea (Goud and Gulden, 2009; Huber, 2010; Nolf and Swinnen, 2013), and has been confirmed in shallow sites along the entire Croatian coast (Peharda, personal communication) as well as the coasts of Greece and Israel (Sivan et al., 2006; Lécuyer et al., 2012).

The objective of this study is to evaluate the potential of *G. pilosa* as an environmental indicator for the Mediterranean region using a sample set collected from Pašman Channel in the middle Adriatic Sea. The site is shallow and nearshore, and we were thus also interested in quantifying the contributions of terrestrial influence, particularly from precipitation, relative to marine influences. More specifically, the principal goals were to (i) determine whether crossdating is possible in *G. pilosa*, ii) that a well-replicated an annually resolved chronology can be developed from growth-increment widths, (iii) identify environmental variables that correlate with the resulting chronology and (iv) estimate longevity of *G. pilosa* samples. Based on previous research, we hypothesize that environmental variability influences growth and will allow for crossdating, and that those conditions associated with productivity, including high levels of precipitation and cool summer temperatures, will be associated with positive anomalies in the chronology. As such, this study offers a unique opportunity to evaluate climate-growth responses in a Mediterranean species, but also one in nearshore habitat subject to considerable human influence. Most importantly, this bivalve species could represent an important and broadly-distributed environmental archive for investigating long-term climate-biology relationships and possibly climate reconstructions in the Mediterranean region.

2. Material and methods

2.1. Collection and sample preparation

Glycymeris pilosa was live collected by SCUBA and skin diving from Pašman Channel, middle Adriatic Sea (43°56′53″N, 15°23′15″ E, Fig. 1) at 1.5–3 m depth. This is an inshore habitat, protected by numerous islands and known for underground freshwater springs. The area is popular for bivalve harvesting, primarily for Noah's ark shell *Arca noae*. This Arcidae bivalve is more abundant in locations characterized by lower salinities that can be caused by terrestrial run-off and underwater springs (Morton and Peharda, 2008). In May 2011, *G. pilosa* samples were collected by a commercial diver, while in May 2014 and April 2015 sampling was conducted in the framework of the ARAMACC project (Butler et al., 2013b). Bivalves were transported to the laboratory, soft tissue was removed, shells were washed, and air dried. Fifty of the largest shells were archived for sclerochronological study.

Analyzed shells ranged in size from 68.3 to 85.0 mm ($N = 50$, $x = 73.8 \pm 3.9$ mm) in length, 42.0–61.2 mm ($x = 52.0 \pm 3.7$ mm) in width, and 62.81–181.70 g (119.32 ± 23.76 g) in weight. The area around the hinge of one valve was carefully excised using Struers Labotom 3 saw and embedded in epoxy resin. Each sample was then cut along the axis of maximum growth, and the exposed surface ground on a series of wettened silicon carbide papers (220–2000 grit) and polished using a soft cloth impregnated with diamond paste (3 μm). Shell sections were etched in 0.1 M HCl for 2–3 min and/or in 0.3 M HCl for 30 s and acetate peels were prepared (see Richardson, 2001). Composite images were constructed from digitally photographed acetate peels using Leica 125 microscope 64× magnification equipped with Leica DFC295 camera and Image-Pro Plus 7.0 software.

2.2. Crossdating and chronology construction

Only samples in which growth increment boundaries were well defined and easily discerned were chosen for chronology development. Acetate peel images of these specimens were visually crossdated using the list year method (Yamaguchi, 1991) to ensure that each increment was assigned to the correct year of formation. This technique is based on assumption that some aspect of environment limits growth, and as it varies over time, it induces a synchronous growth pattern in individuals sampled from the same area (Fritts, 1976; Yamaguchi, 1991). The correct calendar year of each increment was assigned by crossdating backward from the known year of sampling (Fig. 2).

After visual crossdating, growth increment widths were measured continuously from the margin toward the origin using the caliper function in the Image Pro Premier 9.1 program. Three sets of measurements were made per specimen, always following the main growth axis. Qualitative verification of crossdating was conducted using the program COFECHA (Holmes, 1983; Grissino-Mayer, 2001), which has been successfully applied to other bivalve species (e.g. Black et al., 2008; Holland et al., 2014). COFECHA was used to fit each measurement time series with a cubic smoothing spline of 15-year 50% frequency cutoff, which closely tracked low-frequency variability. Each measurement time series was then divided by the values predicted by the spline, isolating high-frequency variability and standardizing each time series to a mean of one. Each standardized time series was then correlated with the average of all other standardized time series; any individuals with unusually low ($p < 0.01$) correlations were visually re-inspected for possible dating errors. The overall average comparison between each individual and the average of all others was reported as the series intercorrelation. COFECHA was also used for