PHD COURSE IN LIFE AND ENVIRONMENTAL SCIENCES

Report Form for PhD student annual evaluation (XXXVII and XXXVIII cycles)

Name of PhD student: Teo Marrocco

Title of PhD research: Bioerosion: which effect on habitat complexity and biodiversity?

Name of PhD supervisor: Barbara Calcinai
Research lab name: Zoology laboratory

Cycle:
[ ] XXXVI
[X] XXXVII

PhD Curriculum:
[X] Marine biology and ecology
[ ] Biomolecular Sciences
[ ] Civil and environmental protection

DISVA instrumentation labs/infrastructure eventually involved in the project:
[X] Actea Mobile Laboratory
[ ] Advanced Instrumentation lab
[ ] Aquarium
[ ] MassSpec lab
[ ] MaSBiC
[ ] Simulation/informatics lab
[ ] Other. Please, indicate: .....

ABSTRACT
Spatial heterogeneity is a fundamental driver of ecosystem biodiversity and stability. Organisms capable of modifying the benthic seascape can be divided in autogenic (when the physical presence of the organisms create the habitat complexity) and allogenic (when the activity of the organisms create the habitat complexity) engineers [1].
While the autogenic group, mainly represented by sponges, corals and macroalgae, is able to create epibenthic three-dimensional structures and thus increase microhabitats (e.g. animal forests and canopies), the allogenic group can erode or burrow and deeply modify the substrate, creating a three-dimensional complexity of holes and crevices that can be used by infaunal species and provide new habitats for a variety of organisms such as fish, gastropods, crustaceans and cnidarians, especially in their early life stages.

Both groups affect the production of ecosystem services, but while there is a large body of scientific literature on autogenic engineers, knowledge on the habitat created by allogenic organisms is still scarce [2].

Part 1. Scientific case of the PhD Research

- BACKGROUND

Mediterranean coasts, and in particular the rocky coasts of the Conero promontory in the Adriatic Sea, are strongly influenced by erosive organisms belonging to different taxa. Porifera and Bivalvia are the endolithic taxa eroding the most carbonate rocks, but polychaetes, echinoderms, microalgae and fungi also participate in the erosion process [2]. Among them, the mollusc bivalves *L. lithophaga* and *P. dactylus* are ecosystem engineers, defined as allogenic. They are capable of altering the rock in which they live, eroding it and creating the structural complexity that organisms such as fish, crustaceans and gastropods rely on to find shelter, food or simply to rest. *L. lithophaga* and *P. dactylus* are included in the Bern and Barcelona Convention while *L. lithophaga* is also included in the Habitats Directive and are defined as "Animal species of Community interest requiring strict protection"; moreover, the European Regulation 1967/2006 (art.8) prohibits their capture, transport, and sale in Italy [3]. However, illegal fishing continues to affect not only the conservation status of the eroding bivalves, but also undermines the integrity of their habitats and, by extension, all associated species [1] [4].

- SCIENTIFIC AIMS

My PhD thesis aims to evaluate bioerosion processes by *L. lithophaga* and *P. dactylus* along the Conero coast as potential enhancers of local biodiversity. To better understand the role of holes in shaping benthic assemblages, a study with experimental substrates was designed to address and tailor future conservation management [3] [4]. Before starting the research project, a review was conducted to have a clear understanding of bioerosion among marine ecoregions of the world and consider when and how the allogenic effects are reported.
My PhD project is structured in 4 main sections:

1 A review of current knowledge on the effects of bioerosion, with the aim of identifying taxa mainly involved in the processes and gathering information on the geographical areas of most interest for bioerosion studies. By the time the objective is achieved, there will be a higher consciousness of the general effect of bioerosion by geographic area and the main gaps of knowledge that need to be explored with further research.

2 Holes and biodiversity. Installation of artificial substrates to recreate the habitat eroded by *L. lithophaga* and *P. dactylus*, with the aim of studying the associated biodiversity. Data from the artificial panel will demonstrate the importance of boring bivalves in influencing local biodiversity and will quantify the species living in this habitat.

3 Borers and holes interactions. Application of the 'time-lapse' technique [3] to analyse the community present in the natural habitat of *L. lithophaga* and *P. dactylus*, with the aim of identifying possible facilitation mechanisms created by the presence of these bioeroders. The aim of the research is to provide data on how macroborers not only influence the number of species found in the habitat, but also how they preferentially use the habitat characterised by the presence of bivalve burrows.

4 Borers assessment. Application of standard monitoring protocols with photographic transects, to analyse the distribution of *L. lithophaga* and *P. dactylus* populations along the coast of Passetto (AN). The purpose of the habitat analysis is to assess the density of the two species, distinguishing between live and active individuals and empty holes in the substrate because of past or current illegal fishing activity. This data may provide also an estimation of the illegal fishing activities in the area.

- WORKPLAN AND RESEARCH ACTIVITIES

WP 1. Objective.

Review of current knowledge on the effects of bioerosion with the aim of identifying taxa mainly involved in bioerosion processes and gather information on geographic areas mostly interested by the bioerosion

studies.

Methods.

A systematic review with Scopus and Web of science databases using the following keywords was conducted: bioero* OR boring OR *borer OR endolithic* OR epilithic* OR excavating OR grazing OR grazer* OR borehole AND cryptic OR fragmentation OR rubble OR facilitation OR recruit* OR biodiversity OR dissolution AND ocean OR ecosystem-engineer OR vacant

Expected/Obtained Results.

The purpose of the review is to reveal that information on the effects of bioerosion is geographically limited and very scarce, with the actual scientific bibliography focusing on only a few aspects of bioerosion and not considering its facilitation capacity.
Before starting the review, we expected to find a large number of scientific publications from tropical regions due to the high level of research activity on coral reefs as a result of climate change and ocean acidification.
A total of 1554 articles were extracted from the search, 753 from Scopus and 801 from Web of Science. An initial screening to remove duplicates reduced the total number of selected publications to 921, which was further reduced to 110 after exclusion due to titles, abstracts or entire articles not meeting our criteria. Finally, after adding the papers from the reference analysis (45) and the expert opinion approach (23), the total number of publications included in the present study was 178.
Excluding review papers (22) and excluding those falling outside the coast and shelf water shallower than 200 meters of depth (11), 152 papers were counted, covering 36 marine provinces. The majority of studies were conducted in tropical regions (105), representing more than 69% of the papers considered, with temperate and polar regions representing only 40 and 7 papers respectively. Overall, most studies were conducted in the tropical Atlantic (32.8%), followed by the central Indo-Pacific (15.8%) and the temperate northern Atlantic (15.1%). Conversely, the Tropical Eastern Pacific, Southern Ocean and Arctic realms were the least studied, with only 4, 4 and 3 documents respectively.

For each marine province, information on bioerosion effects was recorded and included in one or more of the 9 defined categories to analyse the available data on bioerosion processes per geographical area. Although the effect on the control of algal communities was the most recorded of all the areas, being present in 9 out of 12, the effect on damage to coral structures was the most studied, with a total of 58 documents spread over 7 areas. All other effects studied are scattered among the realms, with carbonate cycle (8), microhabitat formation (7) and substrate shaping (5) present in only a few realms.

WP 2. Objective.

Installation of artificial substrates to recreate the habitat eroded by *L. lithophaga* and *P. dactylus*, with the aim of studying the associated biodiversity.

Methods.

With regard to the research activity based on artificial substrates, the first phase consisted of exploratory dives to identify the most suitable area for the installation of the material. The two types of artificial substrate, i. e. perforated bricks, are comparable to the natural substrate burrowed by the bivalves, while the unperforated substrates (solid bricks) are used as control (FIGURE 1). The bricks were then installed in a row, at least one metre apart, to avoid the influence of one type on the other.

Six months after installation, the fauna living in the holes of the bricks was sampled with syringes. The removal of the first substrates has just started and will continue according to the supposed schedule, while the analysis of the associated fauna of the first substrates is in progress.

Expected/Obtained Results.

We expect to collect different taxa inhabiting the two groups of artificial substrates. The meiofauna living in the accumulated sediments will be analysed and compared with the surrounding sediments. fishes, Decapoda, Crustacea, Porifera, Cnidaria are described in the literature as taxa that could rely on the *L. lithophaga* microhabitat to fulfil different ecological needs; the holes are expected to attract these species and organisms typical of soft sediments that are likely to fill the holes of the bricks. On the other hand, the solid bricks are expected to recruit a smaller number of organisms typical of the surrounding habitat.

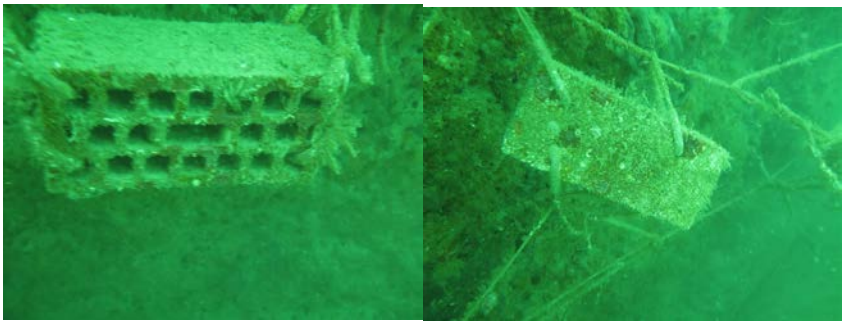

Figure 1 Example of the two types of artificial substrates: perforated bricks to recreate the natural substrate (left); solid brick used as control (right)

WP 3. Objective

Application of the 'time-lapse' technique to analyse the community present in the natural habitat of *L. lithophaga and P. dactylus*, with the aim of identifying possible facilitation mechanisms created by the presence of bioeroders.

Methods.

The time-lapse (TS) technique allowed the study of the fauna associated with the habitat of the boring bivalves. The TS technique was carried out with a modified GO PRO, implemented with a motherboard that allows the adjustment of the image acquisition times; the camera was placed in a steel diving suit resistant to the pressure of 10 ATM, all connected to an external battery that lasted 7 days. The collection of images produced a series of photos (about 2600) taken every 3 minutes for 7 days (FIGURE 2).

Expected/Obtained Results.

The analysis of the images involved first standardising the behaviour of the identified fauna and then distinguishing between habitats with holes and habitats without holes, all within the same time-lapse captured field. In this way, it was possible to obtain comparable and statistically analysable results.

Finally, the statistical analysis carried out using the PRIMER-E7 software allowed us to confirm:

i) the importance of macroborers in creating substrate complexity and thus influencing its biodiversity (using SIMPER analysis WITH 96% dissimilarity between habitats with holes and habitats without).

ii) the dissimilarity in the behaviour of fish, crustaceans, and gastropods between habitats with holes and those without (PERMANOVA analysis with $p<0.001$), confirming that structural complexity is reflected in high community structure.

This leads to the conclusion that macroborers not only influence the number of species found in the habitat, but also how they preferentially use the habitat characterised by the presence of bivalve burrows.

These data highlight the role of these ecosystem engineers in supporting high biodiversity and underline the importance of conserving their habitat and preserve *L. lithophaga* and *P. dactylus* populations.

Figure 2 Example of the pictures obtained through the time-lapse technique.

WP 4. Objective

Application of standard monitoring protocols with photographic transects, to analyse the distribution of *L. lithophaga* and *P. dactylus* populations along the coast of Passetto (AN).

Methods.

Transects research activity was carried out in February, April, and August 2023. The aim of the surveys was to estimate the distribution of *L. lithophaga* and *P. dactylus*. Each transect consisted of placing a 20-metre tape to record the geomorphology of the seabed and taking photographs every metre of the transect, recording the depth and angle of slope of the substrate. More, data of the density of *L. lithophaga* and *P. dactylus* will be extrapolated from the pictures using ImageJ analysis as well as the ratio between living and dead organisms.

Expected/Obtained Results.

The information collected made it possible to create geomorphological profiles of the seabed (FIGURE 3) analysed and to assess the density of these two species, distinguishing between live and active individuals and empty holes in the substrate resulting from past or current illegal fishing activity. The habitat analysis showed that the density of holes could vary from 1-50 units/m^2 up to 280 units/m^2, almost completely covering entire areas and making the need for fishing prevention measures to reduce the destruction of the three-dimensionality of the seabed. In fact, it has been estimated that it takes 20 years for 1 square metre of substrate to recover.

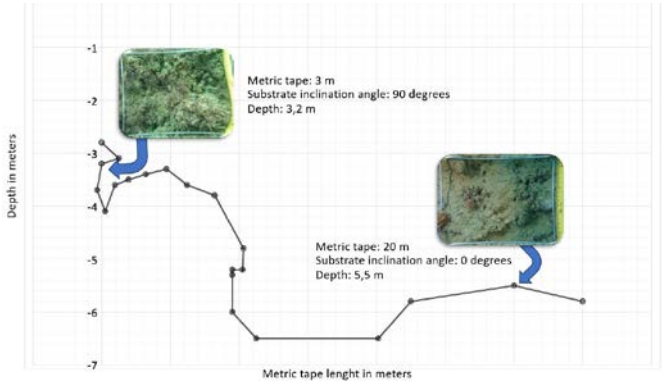

Figure3 Example of a geomorphological profile constructed using data on depth, slope and distance from shore, with associated photographs of each point on the seabed analysed.

REFERENCES

1. Gratwicke, B., & Speight, M. R. (2005). The relationship between fish species richness, abundance and habitat complexity in a range of shallow tropical marine habitats. *Journal of fish biology*, *66*(3), 650-667.
2. Pinn, E. H., Thompson, R. C., & Hawkins, S. J. (2008). Piddocks (Mollusca: Bivalvia: Pholadidae) increase topographical complexity and species diversity in the intertidal. *Marine Ecology Progress Series*, *355*, 173-182.
3. European Commission (2019). The Habitats Directive [WWW Document]. URL https://ec.europa.eu/environment/nature/legislation/habitatsdirective/index_en.htm (accessed 4.27.20).
4. Fanelli, G., Piraino, S., Belmonte, G., Geraci, S., & Boero, F. (1994). Human predation along Apulian rocky coasts (SE Italy): desertification caused by *Lithophaga lithophaga* (Mollusca) fisheries. *Marine Ecology Progress Series*, 1-8.
5. Enrichetti, F., Dominguez-Carrió, C., Toma, M., Bavestrello, G., Betti, F., Canese, S., & Bo, M. (2019). Megabenthic communities of the Ligurian deep continental shelf and shelf break (NW Mediterranean Sea). *PLoS One*, *14*(10), e0223949.

Part 2. PhD student information on the overall year activity (courses/seminars/schools, mobility periods, participation to conferences)

List of attended courses/seminars/schools:

1. Design of research: European project-A.A. 2021/2022- Online itinerant course UNIVPM

   Prof. Nicola Paone

2. Technology transfer and innovation A.A. 2021/2022-Online itinerant course UNIVPM

   Prof. Donato Iacobucci

3. Introduzione all'ambiente LaTex per la redazione di documenti scientifici A.A. 2022/2023- Online computer lessons UNIVPM

   Prof. Francesco Spinozzi

4. Assessing oxidative stress in biological systems- A.A. 2022/2023- Online frontal lessons UNIVPM

   Prof. Elisabetta Damiani

5. Workshop on Multivariate Analysis in Ecology, using PERMANOVA+ for PRIMER. 15-19 May 2023, Napoli.

   Prof. Marti J. Anderson

List of conferences/workshops attended and of contributions eventually presented:

1. SIBM 51° congress- June 14-17, 2022. Online congress participation only- (https://www.sibm.it/)

2. 82° Congresso Unione Zoologica Italiana-Palermo- September 11-16, 2023. Poster presentation.

(Book of abstract available at: https://www.uzionlus.it/82-congresso-uzi-2023/BOOK-ABSTRACT-congresso-uzi.pdf)

3. 5Th International Workshop On Taxonomy Of Atlanto-Mediterranean Deep-Sea & Cave Sponges-Poster presentation- Rapallo-September 11-16, 2023. Poster Presentation.

(Book of abstract available at: https://www.workshopdeepcavesponges.com/wp-content/uploads/2023/09/BOOK_OF_ABSTRACTS-5th-INTERNATIONAL-WORKSHOP-TAXONOMY-SPONGE_MB_KL-alta-definizione.pdf)

Part 3. PhD student information on publications

List of publications on international journals

J1. Roveta, C., **Marrocco, T**., Pica, D., Pulido Mantas, T., Rindi, F., Musco, L., & Puce, S. (2022). The effect of substrate and depth on hydroid assemblages: a comparison between two islands of the Tuscan Archipelago (Tyrrhenian Sea). Marine Biodiversity, 52(1), 9.

J2. Roveta, C., **Marrocco, T.,** Calcinai, B., Pulido Mantas, T., Pica, D., Valisano, L., & Puce, S. (2022). Unravelling the sponge diversity of the Tuscan Archipelago National Park (Tyrrhenian Sea, Italy). The European Zoological Journal, 89(1), 317-330.

J3. Roveta, C., Coppari, M., Calcinai, B., Di Camillo, C. G., **Marrocco, T.,** Pulido Mantas, T., & Cerrano, C. (2023). What's the key for success? Translocation, growth and thermal stress mitigation in the Mediterranean coral *Cladocora caespitosa* (Linnaeus, 1767). Frontiers in Marine Science, 10, 1199048.

J4. Pulido Mantas, T., Roveta, C., Calcinai, B., di Camillo, C. G., Gambardella, C., Gregorin, C., Coppari, M., **Marrocco, T.,** Puce S., Riccardi, A., Cerrano, C. (2023). Photogrammetry, from the Land to the Sea and Beyond: A Unifying Approach to Study Terrestrial and Marine Environments. Journal of Marine Science and Engineering, 11(4), 759.

J5. Di Camillo, C., Roveta, C., Mantas, T. P., Gravili, C., Cerrano, C., Calcinai, B., Coppari, M., Gregorin, C., **Marrocco, T.,** Riccardi, A., Puce, S. (2023). Guests or pests? Eirenid hydroids living on the soft tissue of bivalves. Reviews in Aquaculture.

List of publications on conference proceedings

C1. **Marrocco Teo,** Bertolino Marco, Canese Simonepietro Mazzoli Claudio, Montagna Paolo, Puce Stefania, Roveta Camilla, Vultaggio Carlo & Calcinai Barbara Sponges associated with Antarctic stylasterids (Cnidaria, Hydrozoa) from the deep Ross Sea: what's new? Proceedings of the 5th International Workshop on Taxonomy of Atlanto-Mediterranean Deep-sea and cave Sponges. Rapallo, Genova, (IT). 11-16 September 2023. pp. 34

C2. **Teo Marrocco**, Barbara Calcinai, Martina Coppari, Stefania Puce, Torcuato Pulido Mantas, Matteo Ricotti, Camilla Roveta, Carlo Cerrano. Insights Of A Squatting Community: Boring Bivalves' Home Occupation. Proceeding of the 82° Congresso Unione Zoologica Italiana. Palermo (IT) 19-22 September 2023. pp. 110.

C3 Martina Coppari, Chiara Colonna, Camilla Roveta, **Teo Marrocco,** Torcuato Pulido Mantas, Carlo Cerrano Unveiling The Behavioral Ecology Of *Antipathella Subpinnata* (Antipatharia, Myriopathidae) Associated Fauna Through Time-Lapse Technique. Proceeding of the 82° Congresso Unione Zoologica Italiana. Palermo (IT) 19-22 September 2023. pp. 94.

C4 Coppari M, **Marrocco T.**, Pulido Mantas T., Roveta C., De Benedictis D., Lettini A., Schisa B., Calcinai B., Di Camillo C.G., Puce, S., Cerrano C. The contribution of the time-lapse technique to behavioural ecology studies. 7th European Conference on Scientific Diving, 14-18 May 2023, Roscoff (FR), 448910.

C5. Pulido Mantas T., Calcinai B., Coppari M., Di Camillo C.G., **Marrocco T**., Puce S., Roveta C., Cerrano C. Unveiling hidden seascapes: SfM-photogrammetry, a powerful tool for marine caves monitoring. 7th European Conference on Scientific Diving, 14-18 May 2023, Roscoff (FR), 388543.

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