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Nota científica/Scientific note

# FIRST RECORD OF CYANOBACTERIA MICROBORING ACTIVITY IN PAMPEAN SHALLOW LAKES OF ARGENTINA

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ABSTRACT – Bioerosion led by cyanobacteria occurs in terrestrial, freshwater and marine environments and depends on several ecological and climatic conditions. Therefore, the traces left by these boring organisms can give additional information about the paleoenvironment. In the case of freshwater environments of Argentina, the dissolution of mollusk shells has mainly been attributed to chemical and biological processes. The effect of microbioerosion processes in taphonomic lost is still lacking. With the aim of identifying microbioerosion processes, mollusk shells were studied. The specimens collected were specially picked when a biofilm covered the shell or dissolution marks were present. The shell surface of some specimens were scrapped removing the biofilm or the shells were dissolved with HCl, and the biological remains were observed under light microscope. Other specimens were treated with 2.5% sodium hypochlorite to remove organic matter, and then coated with gold and analyzed using a scanning electronic microscope. Surface pitting and circular to ovoid penetrations were observed. The endolithic cyanobacteria found could be identified as belonging to the family Pseudanabaenaceae. This is the first record of microbioerosion borings in shells of *Heleobia parchappii* in a pampean shallow lake, and the presence of the trace produced producing cyanobacteria. The record of this endolithic bioerosion traces enhances the discussion of taphonomic processes affecting mollusk shells in freshwater settings, and may allow adjusting the composition of fossil assemblages and in consequence paleoecological interpretations.

**Keywords:** microbioerosion, freshwater environments, mollusks, Pampean Region, taphonomy.

## INTRODUCTION

The taphonomic processes that occur in the first centimeters of sediment below the sediment-water interface (*i.e.* the benthic taphonomically active zone, TAZ, Davies *et al*., 1989) constitute a barrier for the preservation of recently dead organisms. These early diagenetic processes tend to degrade calcareous skeletons by mechanical fragmentation, bioerosion, maceration, and carbonate dissolution (Freiwald, 1998). One of the major processes is bioerosion, which degrades carbonate skeletal material and rocky limestone coasts in all marine and some freshwater environments (Wisshak *et al*., 2005). At the same time, it makes these materials more susceptible to mechanical abrasion, fragmentation, dissolution, and further bioerosion (Young & Nelson, 1988; Mloszewska, 2014).

Bioerosion led by cyanobacteria (*i.e.* microbioerosion) occurs in terrestrial, freshwater, and marine environments, and depends on several ecological and climatic conditions (Schneider & Le Campion-Alsumad, 1999). As cyanobacteria are organisms that hardly leave fossil record, the traces left by these organisms provide additional information about the environment. In fact, actuo-paleontological studies, which relate traces with the biological entities, are important because provide by analogy paleoenvironmental context to these references (*e.g.* Vogel *et al*., 1987; Radtke, 1991; Glaub, 1994; Radtke & Golubic, 2005). In fact, there is a number of boring cyanobacteria described for freshwater environments (Tribollet *et al*., 2008; Lawfield *et al.*, 2014). However, an ichnotaxonomical assignment of traces, and the taxonomic identification of the trace-producing cyanobacteria in these ecosystems are still lacking.

In freshwater environments dissolution has been pointed as the main process affecting shell surface (Cummins, 1994; Kotzian & Simões, 2006; Nielsen et al., 2008). In the particular case of Argentina, Tietze & De Francesco (2014) concluded that the main process affecting shell surface was dissolution, due to the granular texture and loss of original color and luster observed in shells. This is mainly attributed to biological dissolution through de decomposition of organic matter that release carbon dioxide, organic acids, methane and alcohols, which favor dissolution of carbonate remains and/or microbioerosion.

This is the first contribution in the region regarding the identification of fossil lentic paleoenvironments through cyanobacteria ichnotaxa. The aim of this work is to

communicate the first record of a trace left by a cyanobacteria in a shallow lake of the Pampean Region of Argentina, and to analyze its implication for the fossil record , improving the paleoecological interpretations.

## STUDY AREA

The Pampas are a vast grassy plain that covers central Argentina. From a geological and geomorphological point of view, the Pampean Region is a heterogeneous environment. The geomorphology of the Pampean Region produce a slow drainage of surface water, allowing the formation of a great number of lentic systems (Fernández Cirelli & Miretzky, 2004). These lakes are very shallow, with low permanence of water and highly variable salinity, and naturally eutrophic. They are permanent or transitory waterbodies that are located in a basin of well defined outline, and lack permanent thermal and chemical stratification, with sediments different from the surrounding land (Grosman, 2008). Lake basins were originated during the Late Pleistocene by the action of the prevailing westerly winds (westerlies). With the onset of moister Holocene conditions, these depressions became areas of groundwater discharge and surface-water accumulation, being filled gradually with sediments that contain abundant mollusk shells (Stutz *et al*., 2010; De Francesco *et al*., 2013).

La Brava Lake, a representative Pampean waterbody, was selected for this study (Figure 1). It is one of the few permanent freshwater shallow lakes of the area (Irurzun *et al*., 2014), located in the northern area of the Tandilia range, 42 km far from Mar del Plata city (37º52'S,57º59'W). This shallow lake has an area of 4 km$^2$, a maximum depth of 4.75 m and is classified as bicarbonate-sodium oligohaline (Romanelli *et al*., 2010). Like others waterbodies of the region, it is immersed in crop-livestock agroecosystems (Romanelli *et al*., 2011).

## MATERIAL AND METHODS

During summer and winter of 2015 shells of the mollusks *Heleobia parchappii* (d'Orbigny, 1835) and *Biomphalaria peregrina* (1835) were collected in the littoral zone of La Brava Lake. The specimens collected were specially picked when the biofilm covered the shell externally or dissolution marks were observed; however, a few shells without any cover were also collected. This was performed in order to analyze

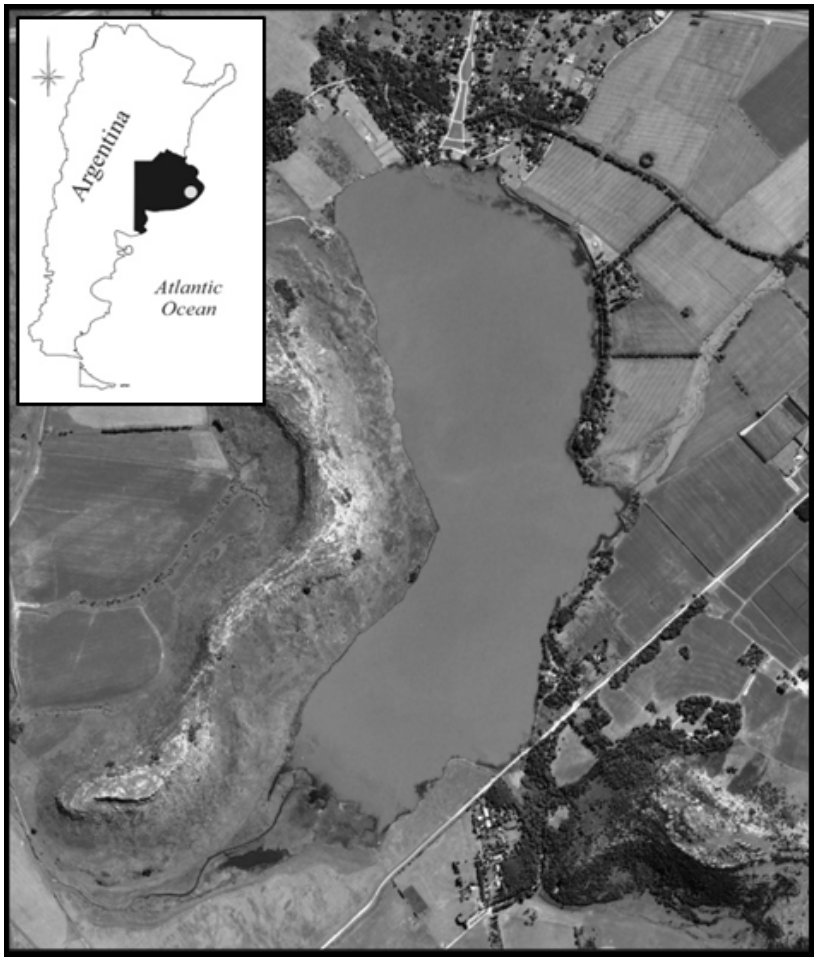


**Figure 1.** Map of the study site: La Brava lake.

the different shell surfaces under this three conditions, and to know if microbioerosion was associated to the presence of biofilm. Samples were stored in alcohol 70° and carried to laboratory. Shell surface of some specimens were scrapped, and the removed biofilm was observed under light microscope. In some cases, shells were dissolved using dilute HCl (5 %) in order to determine the presence of euendolithic species, and observe them under light microscope. Other specimens were treated with sodium hypochlorite 2.5% to remove organic matter, and then coated with gold and analyzed using scanning electronic microscope (**SEM**) at the Laboratory of Microscopy of the National University of Mar del Plata.

## RESULTS AND DISCUSSION

This note corresponds to the first record of microbioerosion borings in shells of *Heleobia parchappii*, and the presence of trace producing cyanobacteria. Taphonomic decay of studied shells was noted, with the external periostracum and prismatic aragonite layers deteriorated (Figure 2A). The presence of a biofilm covering the shells do not determinate the presence of euendolithic cyanobacterias, *i.e.*, shells presenting biofilm were observed under SEM and its surfaces were intact.

Surface pitting and circular to ovoid penetrations were observed at SEM in some shells presenting signs of dissolution at naked eye (Figures 2B–D). The circular and subcircular penetrations are interpreted as the aperture of deep endolithic perforations on the shell. These shells also presented loss of proteinaceous parts (periostracum), and most of them presented signs of fragmentation. These taphonomic features are very common in shells deposited in dead assemblages in freshwater systems of the Pampean Region (Tietze & De Francesco, 2014, 2017) suggesting that the shells presenting microbioerosion do not seem to be older than the other ones collected. However, time-averaging studies in pampean lakes are still missing. The longitudinal tunnels, which end in circular to ovoid apertures, were present throughout the shell, ending in the internal and external layers. Euendolithic filamentous cyanobacteria were found when shells were observed at light microscope and

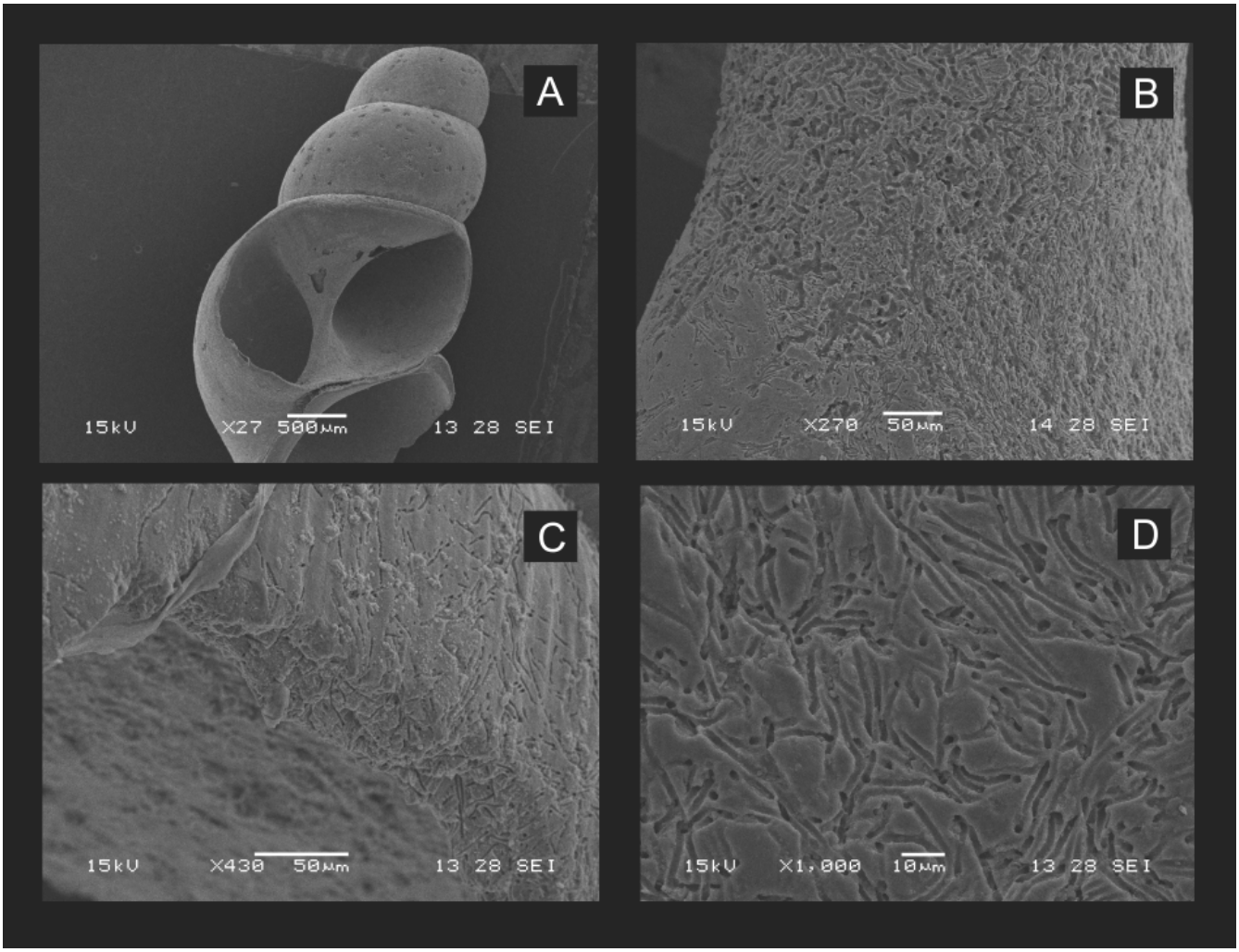


**Figure 2.** Photographs at scanning electronic microscope of *Heleobia parchappii* shells. **A**, shell surface; **B–D**, tunnels and ovoid to circular pitting caused by euendolithic cyanobacteria.

SEM. Cyanobacteria cells are arranged in uniseriate straight trichomes enveloped by a fine and mucilaginous sheath (mean filamentous wide: 2 µm) (Figures 3A–B). Trichome cells are longer than wide; long: 2.15–4.8 µm (mean: 3.13 µm); wide: 0.93–1.5 µm (mean 1.1 µm), and present constrictions at the cross walls. Apical cells are longer than trichome ones (mean long: 4 µm) with rounded ends. Cells sizes of trichome were confirmed at SEM long: 2.14–5.4 µm (mean: 3.77 µm); wide: 1.46–1.9 µm (mean: 1.71 µm) (Figures 3C–D).

Previous studies of mollusk preservation performed in the region highlight the importance of destroying processes in the freshwater environments (Cristini & De Francesco, 2012; Tietze & De Francesco, 2012, 2014, 2017). One of the taphonomic attributes most commonly found in shells is the 'fine-scale surface alteration' (defined by Best & Kidwell, 2000), an attribute that combines the alteration produced by dissolution, abrasion and microbioerosion, at low magnification. This feature is generally attributed to dissolution due to the subsaturation of calcium carbonates found in freshwater environments. However, pampean shallow lakes are highly productive environments (*i.e.* naturally euthrophic lakes) where shells are prone to be affected by dissolution associated to decomposition of organic matter and/or microbioerosion processes. Therefore, it is important to discern among the alterations produced by different processes. In accordance, this contribution highlights the importance of considering microbioerosion as a process affecting shell preservation in shallow lakes of the region.

The endolithic cyanobacteria found in *Heleobia parchappii* shells could be identified as belonging to the family Pseudanabaenaceae (Komárek & Anagnostidis, 2005). However, future molecular studies are needed to identify at the genera or species level the microorganisms that generate the traces. The identification of the cyanobacteria will allow to know the ecological requirements of these microorganisms, enhancing the paleoecological interpretations done in shallow lakes of the region, because physico-chemical parameters (temperature and salinity, as examples) affect the presence and distribution of certain euendoliths (Gektidis *et al*., 2007). Besides, microendoliths occur over a wide range of salinities, from hypersaline to freshwater environments, implying the feasibility of applying microboring trace fossils as paleosalinity indicators if different species are found (Wisshak, 2012).

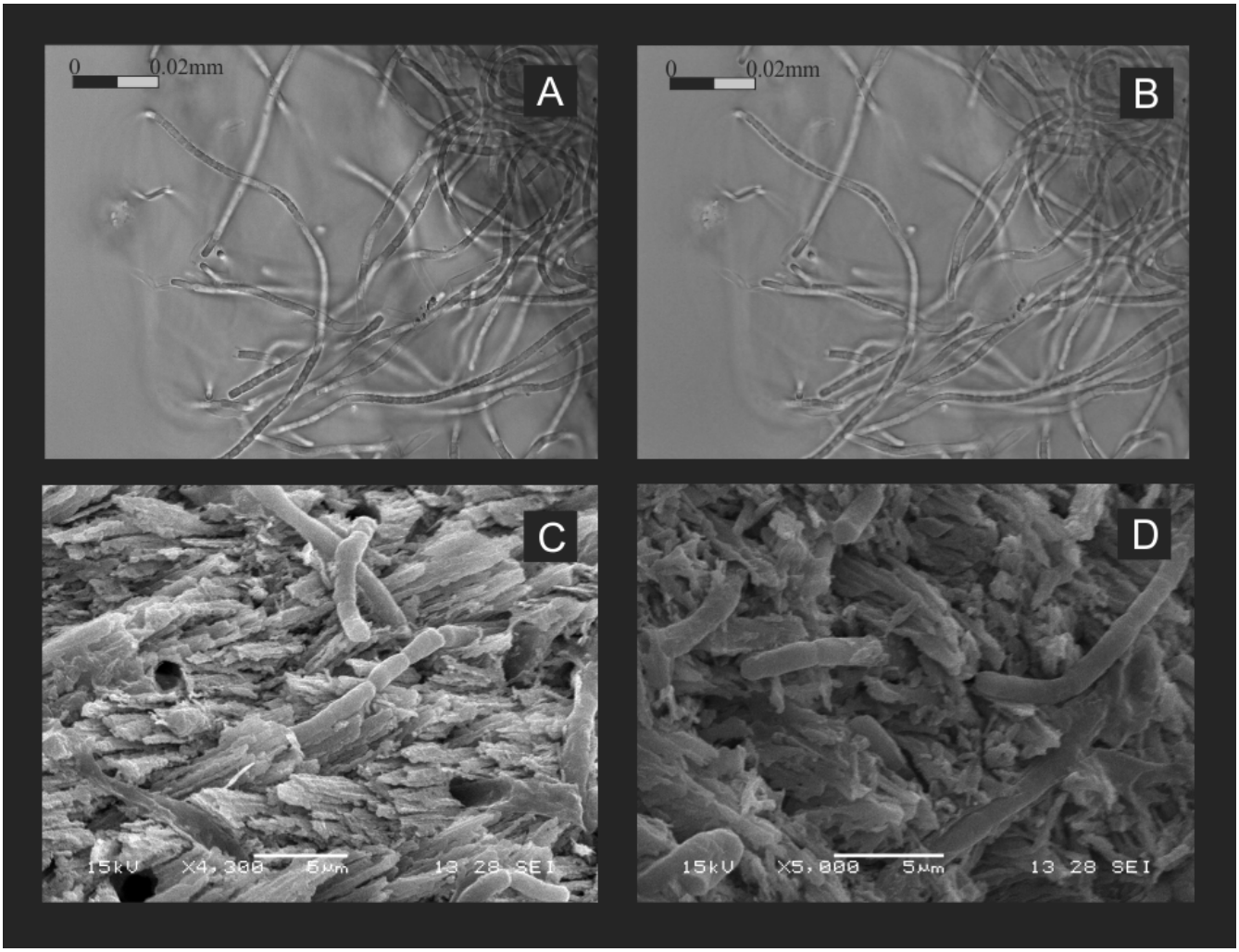


**Figure 3.** Photograph at light (**A–B**) and scanning electronic (**C–D**) microscopes of cyanobacteria filaments found in *Heleobia parchappii* shell surface.

## CONCLUSION

This work describes a new finding of microbioerosion in shells of freshwater mollusks of Pampean shallow lakes, characterized by surface pitting and circular to ovoid penetrations. The tunnels were present throughout the shell, ending in the external and internal surfaces. Genetic analysis would be necessary to identify correctly the endolithic cyanobacteria species. The record of this endolithic bioerosion trace enhances the discussion of taphonomic processes affecting mollusk shells in freshwater settings, and may allow to adjust the composition of fossil assemblages. In the future it would be interesting to perform a study comparing the microbioerosion among different lakes (different salinity levels as an example) and find out if the same or different species of euendolithic species are present in the different environments, allowing their differentiation in the fossil record.

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