

**Bioerosion in
foraminifera from
heavy metal-polluted
coastal area**

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Bioerosion by microbial euendoliths in benthic foraminifera from heavy metal-polluted coastal environments of Portovesme (South-Western Sardinia, Italy)

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Abstract

A monitoring survey of the coastal area facing the industrial area of Portoscuso-Portovesme (South-Western Sardinia, Italy) revealed intense bioerosional processes. Benthic foraminifera collected at the same depth (about 2 m) but at different distances from the pollution source show extensive microbial infestation, anomalous Mg/Ca molar ratios and high levels of heavy metals in the shell associated with a decrease in foraminifera richness, population density and biodiversity with the presence of morphologically abnormal specimens. We found that carbonate dissolution induced by euendoliths is selective, depending on the Mg content and morpho-structural types of foraminiferal taxa. This study provides evidences for a connection between heavy metal dispersion, decrease in pH of the sea-water and bioerosional processes on foraminifera.

1 Introduction

Boring microflora, constituted by cyanobacteria, algae and fungi may colonize carbonate substrates of both dead and living tissues of carbonate organisms (Tribollet, 2008). Boring is evident in carbonate organisms from the Mesoproterozoic (Zhang and Golubic, 1987) to the present day as a consequence of environmental conditions (Hallock, 2005). Microbial euendoliths have been described in coral reefs, molluscs, thalli of red algae (e.g. Golubic et al., 1975; Perkins and Tsentas, 1976; Budd and Perkins, 1980; Glaub, 1994, 2004; Chazottes et al., 1995, 2002; Le Campion-Alsumard et al., 1995; Vogel et al., 2000; Tribollet and Payri, 2001; Ghirardelli, 2002; Golubic and Schneider, 2003; Tribollet and Golubic, 2005; Tribollet, 2008; Tribollet et al., 2009), in coastal eroded limestones (Schneider and Torunski, 1983; Radtke et al., 1996), in carbonate grains (Tudhope and Risk, 1985; Al-Thukair and Golubic, 1991; Al-Thukair, 1999), as well in oil polluted marine environments (Campbell, 1983; Al-Thukair, 2002; Al-Thukair et al., 2007) and heavy-metal polluted lagoons (Succi et al., 2010).

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Microborings are major agents of bioerosion dissolving large quantities of calcium carbonate with a potential in buffering seawater pH, which leads to new questions on the effects of environmental factors such as eutrophication and atmospheric CO₂ increase on ocean composition evolution (Tribollet, 2008). On the other hand, present day geochemical models predict that the saturation state of surface ocean water with respect to carbonate minerals should decline during the twenty-first century. As a result calcareous organisms may have difficulties to calcify, leading to production of weaker skeletons and greater vulnerability to bioerosion. Organisms utilizing the more soluble form of CaCO₃ (aragonite or High-Mg calcite) would be more adversely affected by elevated *p*CO₂ than those utilizing the less soluble low-Mg calcite (Morse, 1983; Morse et al., 2007).

Investigations on microborings in foraminifera are documented in a small body of literature (Perkins and Halsey, 1971; Alexanderson, 1972; Golubic et al., 1984; Peebles and Lewis, 1988; Shroba, 1993; Freiwald, 1995; Perry, 1998; Nielsen et al., 2003; Crevison and Hallock, 2007) and open a debate that needs to be developed. Benthic foraminifera secreting calcium carbonate shells are considered to be very sensitive to the environmental conditions of the host sea water (e.g. Boyle, 1981; Wefer et al., 1999; Lea, 2004; Munsel et al., 2010). Species distribution and population densities of foraminifera are in fact used for pollution monitoring (Alve, 1991, 1995; Yanko et al., 1994, 1999; Debenay et al., 2001, 2005; Coccioni et al., 2003, 2005; Arminot du Châtelet et al., 2004; Cherchi et al., 2009; Frontalini et al., 2009; Romano et al., 2009; Frontalini and Coccioni, 2011; Denoyelle et al., 2012; Foster et al., 2012). Hallock (2000 a, b) suggested that climatic change including increasing atmospheric CO₂ and stratospheric ozone depletion may be the cause of borings developing on carbonate organisms. Indeed, effects of elevated *p*CO₂ on coral reef bio-erosion have been observed and explained through a mechanism of carbonate dissolution in a more acidic ocean (Tribollet et al., 2009). The activity of boring microflora can result in differential bioerosion depending on both *p*CO₂ partial pressure (Tribollet et al., 2009) and Mg/Ca ratio of the foraminifera tests (Peebles and Lewis, 1988; Bentov and Erez, 2005).

The aim of our study is to investigate the effects of microbioerosion on benthic foraminifera and trace element dynamics in a known coastal industrial site polluted by heavy metals and industrial release of CO₂ (Schintu and Degetto, 1999). Field studies in the coastal area neighbouring the Industrial Complex of Portoscuso-Portovesme (Southern Sardinia, Italy, Fig. 1) show that the presence of high levels of heavy metals results in an increase in bioerosional processes on foraminifera by boring microflora. The stressed environmental conditions in this area have been highlighted in Cherchi et al. (2009) through an analysis of the biotic indices of foraminifera tests (Dominance, Shannon–Weaver, Simpson, Evenness, Menhinick, Margalef, Equitability, Fisher- α , Berger-Parker and Q-mode Cluster Analyses – Ward Method) in this polluted environment.

2 Environmental setting

The study area is characterized by a relatively confined shallow shelf, which slopes gently to deeper water environments (Fig. 1). Bottom sediments comprise sandy sub-marine beach (Pleistocene-Holocene in age), while Oligo-Miocene calc-alkaline volcanics crop out north of the sampling area. This south-western coast of Sardinia has been under major anthropogenic pressure since the 1960s, when the Portoscuso-Portovesme district began to develop and expand its activities. The Portovesme Harbour, built in 1870 to ship ores (zinc blend, galena and pyrite) from the historical mines of the Sulcis-Iglesiente district, received major inputs from industrial discharge until a few decades ago. Chemical analysis of marine sediments from 4 cores collected in the harbour of Portovesme facing the Industrial Complex revealed in the upper 2 cm high concentrations of Hg (up to 50 mg kg⁻¹), Cd (up to 120 mg kg⁻¹), Pb (up to 20 mg kg⁻¹) and Zn (up to 70 mg kg⁻¹) were found. Metal concentration decreases with an increase in distance between the sampling station and the industrial effluent discharge point (Schintu and Degetto, 1999). This large industrial development had a considerable environmental impact, such that this industrial district has been

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declared an environmental hazard (D.P.C.M. – Prime Ministerial decree, 23.04.1993) because of the several “danger centres” recognized (Agenzia Regionale per la Protezione dell’ambiente della Sardegna – ARPAS, 2007).

Emissions into the atmosphere and all of the surrounding environments have been estimated annually at 65 000 tons of SO_x, 4000 tons of dust, 10 tons of Pb and 100 tons of Fe (Gazzetta Ufficiale Italiana, 1993). The large electric power stations and the numerous chimneys produce also significant amounts of CO₂ (Schintu and Degetto, 1999; Bettini and Zanin, 2002). The Industrial Complex, developed on an alluvial plain near the sea, includes factories producing aluminium from bauxite and a lead-zinc smelter producing Pb, Zn, Cd, H₂SO₄ and Hg from Pb and Zn mixed sulphides. A large dump of red muds (seawater-neutralized bauxite refinery residues) is located near the coast.

3 Material and methods

Sixteen surface sediment samples, coming from an area about 12 km², were collected in the inner shelf facing the Industrial Complex of Portoscuso-Portovesme where seawater is less than 2 m deep. Four stations (PP – Porto Paglietto, LC – La Caletta, PV – Portovesme, PA – Punta S’Aliga, Fig. 1) were sampled in summer. Sampled sediments came from the upper 1–2 cm below the water column. The water temperature ranged between 27.7 °C and 30.6 °C. Temperature, pH, and Eh were measured for both the seawater and pore water of the sediments in every sampling station. Blank samples were also collected for comparison purposes at the same bathymetry and temperature from an unpolluted coastal area (NP – Portopauleddu, Fig. 1). Benthic foraminifera assemblages and their biodiversity indices, as well the geochemical characterization of the investigated area, was the aim of our earlier paper, in order to assess the foraminiferal response to heavy metal pollution (Cherchi et al., 2009).

A cluster of four subsamples of constant volume (50 cm³) from every station (PP, LC, PV, PA) was collected to study foraminifera assemblages used as environmental

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bioindicators. In laboratory, 16 subsamples were stained with Rose Bengal (Walton, 1952) to differentiate living from dead specimens. Samples were then washed through a set of nested sieves (63, 125, 180, 250 μm). Foraminifera from 5 cc of the > 125 μm size fraction (a – medium grained) and 5 cc of the < 125 μm size fraction (b – fine-grained) were picked, counted and classified.

Considering the very low number (1–4 %) of living individuals in the polluted environments (mainly at the PV and PA stations), total benthic foraminifera assemblages (dead and living) were used for statistical purposes, providing a larger database (Scott and Medioli, 1980; Samir and El-Din, 2001; Fatela and Taborda, 2002; Armynot du Châtelet et al., 2004; Debenay and Guiral, 2006; Frezza and Carboni, 2009). Total assemblages are almost identical statistically and only diverge when the living/total becomes large (Murray, 1976, 1991) and biotic indices do not show significant differences between living and dead populations (Yanko et al., 1994).

The bioerosional process on foraminiferal tests was analyzed via ESEM (QUANTA 200, FEI, Hillsboro, Oregon, and partly with EVOLS15, ZEISS). The epoxy resin-casts of foraminifera were made at Erlangen University (Germany). Samples were gold coated before imaging, for analysis of both external surface and on epoxy-resin cast (Golubic et al., 1970, 1983; Wisshak et al., 2008). From ESEM images, microbial filaments and boreholes on the external surfaces have been compared with positive epoxy-resin casts, and measured. To estimate quantitatively the impact of the microbial borings on the foraminifera and the selective bioerosion on high-Mg and low-Mg foraminiferal tests, 300 individuals were picked randomly from the fraction > 63 μm of each sample and were observed with a scanning electron microscope. A total of 1200 foraminifera was examined. The bioerosional features on several morpho-structural taxa have been quantitatively calculated. The percentage of calcareous dissolution in foraminifera, distinguishing between high-Mg and low-Mg tests, has been compared with heavy metal values both in foraminiferal tests and the sediments, the pH of pore waters and richness of specimens.

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Seawater was collected at the water–sediment interface of the 4 polluted stations and at the reference pollution-free station. After sampling seawater was rapidly filtered in-situ through a 0.4 μm pore-size polycarbonate filter with an all-plastic filtration assembly. Samples for cation analysis were acidified with HNO_3 suprapure acid to pH around 3. At each sampling site, temperature, pH, and Eh were measured for both seawater and sediment porewater. The Eh was measured by platinum electrode and the value was corrected against Zobell's solution (Nordstrom, 1977). The electrode used for pH potentiometric determination was calibrated against three NIST-traceable buffer solutions (pH = 4.01, 7.00, 9.00 at 298 K). Reproducibility of pH calibrations, carried out before and after measurements of a single solution, was better than 0.005 pH unit. However because of problems inherent to the use of glass electrodes calibrated using NIST buffers in strong electrolyte solutions (see Dickson and Goyet, 1994, for a review), this measurement was only used to verify the solution electrochemical difference between the different sampled stations.

Anions were determined by ion chromatography (IC Dionex dX-120) and cations by ICP-AES (ARL-3520B) or ICP-MS (Perkin Elmer dRC-e). Because of the complex seawater matrix the standard addition method was employed for the trace element determination (Cd, Pb and Zn) (Danzer and Currie, 1998; Cidu, 1999). The method validation was verified by applying the same conditions to the CASS-3 coastal seawater reference material. Samples and CASS-3 were diluted five times with a 1 % ultrapure HNO_3 solution before sample spiking. Accuracy and precision were estimated at 10 % or better using the standard reference solution and random duplicate samples. The limit of quantification (10σ value of blank solution response over time) was respectively 0.01 $\mu\text{g l}^{-1}$ for Cd, 0.6 $\mu\text{g l}^{-1}$ for Pb and 0.1 for Zn. The ionic balance was always in the order of $\pm 8\%$.

For digestion of samples with porcelanaceous foraminiferal tests, two portions of each sample were washed with ultrapure water (MilliQ[®]) by mechanical agitation in order to remove detrital grains. The samples were dried, accurately weighed and digested by slow heating in a temperature bath at 30 °C in a Teflon beaker with 3 ml of

ultrapure HNO₃ (67 %). Solutions were diluted to 10 ml with ultrapure water in volumetric flasks, and transferred to new Hd-polyethylene bottles for storage (Jarvis, 1992). Metals were determined by ICP-AES and ICP-MS.

Field Emission Gun SEM (FEG-SEM) is a scanning electron microscope with a high-energy beam of electrons in a raster scan pattern producing information about surface topography and composition. The Quanta 200 FEG Environmental Scanning Electron Microscope (ESEM) uses a field-emission gun (FEG) electron source in an exceptionally high chamber pressure environment. It combines the advantages of nanometer resolution to high signal to noise ratio in both regular high vacuum and environmental (wet) modes. EdX Analysis stands for Energy dispersive X-ray analysis; it is sometimes referred to as EdS or EdAX. EdX analysis allows identification of the elemental composition of the specimen, or an area of interest thereof. The EdX analysis system works as an integrated feature of a scanning electron microscope (SEM), and can not operate on its own without the latter.

4 Results and discussion

4.1 Bioerosion features

The presence of microboring traces and cavities on sampled benthic foraminifera has been recognized using ESEM images of both surface tests and epoxy resin-casts. Identified taxa, reported in Table 1, provide evidence of traces and cavities produced by phototrophic (cyanobacteria, chlorophyta) and heterotrophic (fungi) organisms. In Figs. 2 to 6, we illustrate evidence of several endolithic traces of microbial borings with morphological differences. Details of borings will be described according to the different observational techniques: wall surfaces and epoxy resin-casts.

Test surfaces of the porcelanaceous miliolids present the heaviest bioerosion features under the form of microbial clusters. Endolithic traces have morphological differences related to biodiversity and to mode of life of the boring microbial organisms.

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Infestation is characterized by thin individual tunnels, occasionally bifurcated, bag-shaped cavities and branching patterns composed by rhizoidal and short galleries radiating laterally from the central area. This branching pattern shows similarities with *Fascichnus* isp. (Fig. 7a, b). From ESEM images of the traces on external surfaces of foraminiferal tests, our observations show that the taxa affected by higher bioerosion belong to the high-Mg porcelanaceous group (Suborder Miliolina) and, among these, to the genus *Quinqueloculina* which exhibits heavy microbial bioerosion. Figure 8 shows that in *Quinqueloculina* sp. the Mg content at the bottom of a boring is one order of magnitude higher compared to the unaltered shell surface, suggesting chemical reorganisation of the carbonate mineral composition through a dissolution-precipitation process.

In several specimens the infestation is concentrated along the sutures of the chamber walls providing more easily a nutrient source (Fig. 2b). A heavy infestation by diversified microborings in the test of *Triloculina* has been recorded (Fig. 2c). Radiating traces are developed parallel to the whole surface of foraminifers, in both porcelanaceous and hyaline types (Figs. 3a, g, h and 7b), as previously observed by Glaub (2004) for “*Fasciculus* isp. 2”. Lined rod-shaped bacteria are sometimes visible on the later chambers of *Adelosina* among their weakly developed striae. These cryptoendoliths bacteria (*sensu* Golubic et al., 1981) can go into pre-existing holes, characterized by a larger diameter, clearly bored previously by another larger boring organism (Fig. 2d). Hyaline perforated tests (e.g. *Elphidium crispum*) frequently exhibit well-organized systems of microborings belonging to *Ichnoreticulina elegans* surrounding and directed to pores (Fig. 3e). Bacteria infestation can produce teratological modifications as observed in *E. crispum* (Fig. 3f) where bacteria colonies induce lateral enlargement of primary pores with enlargement of contiguous pores (cf. Freiwald, 1995, in *Cibicides lobatulus*). Superficial traces made by endolithic boring communities can completely cover the walls (Fig. 3a–c).

Several specimens of *E. crispum* show a boring pattern made of curved tunnels of 1–2 µm in diameter. Tunnels, after long runs, go towards the primary pores showing

a comparable behaviour to *I. elegans* and they are tentatively attributed to *Scolecia filosa*. Infestation producing the primary pore connection may be related to possible CO₂ bioavailability for phototrophic organisms from the respiratory activity of the heterotrophic host.

5 Epoxy resin-casts reveal rich microbial communities, fairly diversified, both in porcelanaceous and hyaline tests (Table 1). Analysis of resin-casts reveals euendoliths belonging to phototrophic (chlorophyta, cyanobacteria) and heterotrophic (fungi) organisms boring both porcelanaceous and hyaline foraminifera. The wider and thicker walls of miliolids provide a protected niche for the growth of euendoliths, as shown in Fig. 4.
10 In porcelanaceous tests, dense pavements of well-developed colonies of cyanobacteria and fungal sporangial cavities are very common. The presence of organic films inside the skeleton structure of these foraminifera provides a source of food.

The ichnological interpretation of the euendoliths inside the imperforate porcelanaceous tests is easier than for those inside the perforate hyaline hosts. The very complex
15 internal architecture of the taxa belonging to the families Elphidiidae and Rotaliidae sometimes makes it difficult to distinguish euendoliths from foraminifera ultrastructures (Figs. 5 and 6). Even if the ichnological attribution of the euendoliths in the hyaline perforate taxa is generally uncertain, their presence in the fine skeletal structure indicates that microbial colonies can use foraminiferal canal-systems to penetrate inside the test,
20 and subsequently to develop boring activity in the foraminiferal skeleton.

Our study shows that microbioerosion affects porcelanaceous (High–Mg) imperforate miliolids (*Adelosina*, *Pseudotriloculina*, *Quinqueloculina* and *Triloculina*) and peneropliids as well as hyaline (Low–Mg) perforate foraminifera (*Ammonia*, *Elphidium*, *Lobatula*, *Rosalina*) (Fig. 9). Porcelanaceous miliolids (*Quinqueloculina* and *Triloculina* spp.)
25 exhibit higher percentages of bioerosional features in PA station (28.72 %) and in PV station (21.75 %) while hyaline *Lobatula lobatula* and *Elphidium crispum* reveal higher values of microbial infestation in PA (16.89 %) and LC stations (15.02 %), respectively. In particular, *Elphidium* tests show well-developed colonies of *Ichnoreticulina elegans* surrounding and directed to primary pores. The clorophyceans in our samples seem to

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be more frequent in less polluted sediments (LC). Figure 10a, b show that the number of infested tests is 2 times higher in porcelanaceous compared to hyalines foraminifera and that the proportion of infested tests is 2–3 times higher in the lowest pH conditions. Bioerosion in hyaline specimens increases from 19.9% (PP) to 59.7% (PA), whereas in porcelanaceous forms it increases from 35.1% (PP) to 80.3% (PA). Our observations demonstrate that taxa affected by higher bioerosion belong to the high-Mg porcelanaceous group (Suborder Miliolina), especially *Quinqueloculina*. Infestation is often concentrated along the chamber wall sutures where nutrient material is high (Fig. 2b). Diversified microborings on an infested test of *Triloculina* are illustrated in Fig. 2c.

4.2 Environmental conditions and bioerosion

Seawater composition from four sampled stations is given in Table 2. As previously reported porcelanaceous foraminiferal tests, known to be high-Mg foraminifera, are more infested by bioerosion than hyaline forms. We found that Mg/Ca molar ratio of porcelanaceous infested tests is between 0.09 and 0.13, while in foraminiferal tests collected from an unpolluted site (NP) without apparent sign of bioerosion have a Mg/Ca value of 0.01 (Fig. 10c). Since Mg/Ca molar ratio of benthic foraminifera is normally assumed to be between 0.0005 and 0.01 mol mol⁻¹ (Lea, 1999; Toler et al., 2001; Toyofuku and Kitazato, 2005), we propose that conditions of environmental stress may influence the amount of Mg incorporated during the foraminifera's growth. Our results are in agreement with the experimental investigation on Mg intra-shell reactions of Bentov and Erez (2005) and which are attributed to a kinetic mechanism played by the carbonate ions in the calcite growth by Lopez et al. (2009). Porcelanaceous foraminiferal tests from the anthropogenically polluted lagoon of Santa Gilla (Frontalini et al., 2009) have a similar Mg/Ca ratio (0.11 mol mol⁻¹) which is greater than that collected at station NP, confirming the influence of the stressed environment on the Mg/Ca ratio of these biominerals.

Figure 10d shows that the amount of Cd, Zn and Pb incorporated in the bioinfested shells in PP, LC, PV and PA is higher compared to the pollution-free NP station. We

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found that the amount of heavy metals incorporated in the infested tests is higher when the proportion of infested tests is higher.

The process of bioerosion in calcareous skeletal structures can be regarded differently in high-Mg porcelanaceous imperforate tests and low-Mg bilamellar hyaline perforate tests and can also be related to the decrease of seawater pH (Fig. 10b). The porcelanaceous wall consists of a thick layer of high-Mg calcite needles with relatively large interstices filled with organic matter. Needles are randomly arranged and coated inside by an inner organic lining and outside by the outer organic layer (Towe and Cifelli, 1967; Hemleben et al., 1986; Debenay et al., 2000a). The common infestation of euendoliths in porcelanaceous tests rather than in hyaline tests can be also related to the thick calcite layer of the porcelanaceous wall, allowing an adequate erosional-space for their growth. The perforate foraminifera are characterized by structures of greater complexity (chamber-partitions, shell cavities, canal systems) compared with those of the porcelanaceous group. In the bilamellar hyaline perforate group, the wall comprises carbonate layers separated by an organic median layer (Towe and Cifelli, 1967; Hansen and Reiss, 1971; Hottinger, 1978, 2000; Hansen, 1999; Debenay et al., 2000b). Our data show that increasing heavy metal concentrations in the sediments corresponds to an increase of microbial infestation, reaching a peak at the more polluted sites (PV and PA; Fig. 10e).

The observed high level of microboring infestation is related to the anomalous mechanism of bio-mineral construction in the presence of a high level of heavy metals. In these heavily polluted environments, euendolithic bioerosion develops in small shells of the biocarbonate substrates such as calcareous foraminifera. Microbial organisms need the development of a trophic strategy related to the decrease of micrograzers (mainly benthic foraminifera). According to Mojtahid et al. (2011) some foraminifera species, such as *Haynesina germanica*, *Ammonia beccarii* and the single-chambered species *Psammophaga* sp., consume and digest large quantities of bacteria. The estimated grazing rates of these three intertidal species were comparable: $\sim 3.2 \text{ ng C ind}^{-1} \text{ h}^{-1}$ ($\sim 16\,000 \text{ bacterial cells ind}^{-1} \text{ h}^{-1}$) for low bacterial concentration

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Our results are in agreement with earlier laboratory experiments (Morse, 1983; Morse et al., 2007) where it was predicted that dissolution of various benthic marine organisms may increase under natural higher $p\text{CO}_2$ partial pressure (Ries et al., 2009).

5 Summary



In our study, samples were collected at the same depth (about 2 m) from shallow sediments affected by serious industrial contamination in the coastal environments of Portoscuso-Portovesme. In this area, foraminiferal shells have a high content of heavy metals and are actively infested by microborings. As far as we know, these infested foraminiferal communities provide a state-of-the-art dataset for the understanding of bioerosional processes in foraminiferal hosts in polluted environments.

The results show: (i) microbioerosion is higher in high-Mg foraminiferal tests compared to the low-Mg tests, (ii) the role of foraminiferal skeletal architecture in the boring process, (iii) the abundance of euendoliths is favoured by the effect of greater concentrations of heavy metals, especially Zn, as an inorganic nutrient.

We interpret that in the shallow water and low hydrodynamic conditions of the Portoscuso Portovesme lagoon, the heavy metals leached from mine tailings and industrial discharge are not immediately dispersed in the sea water and, thus, foraminifera can concentrate heavy metals in their shells. This takes place in a complex biomineralization process. In agreement with geochemical models and previous literature, we found that high-Mg carbonate shells dissolve faster and are deeply infested by microborings. While geochemical models already predict that CO_2 increase is affecting biogenic carbonate reservoirs, this study demonstrates that the heavy metal dispersion process can contribute to global CO_2 change via a complex ecological process.

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Table 1. Distribution on microborings at different sampling stations (PP, LC, PV and PA, see Fig. 1), taken from porcelanaceous (Po) and hyaline (Hy) foraminiferal substrates and their relative abundance (XX very common, X common, = rare).

Borings	Foraminifera substrata Trace-makers	PP		LC		PV		PA	
		Po	Hy	Po	Hy	Po	Hy	Po	Hy
<i>Eurigonum nodosum</i> Schmidt	<i>Mastigocoleus testarum</i> Lagerhein (cyanophyte)	X		X		X	X	XX	X
<i>Fascichnus</i> cf. <i>dactylus</i> (Radtke)	<i>Hyella caespitosa</i> Bornet and Flahault (cyanophyte)			X		XX	X	XX	X
<i>Scolecia filosa</i> Radtke	<i>Plectonema terebrans</i> Bornet and Flahault (cyanophyte)	X	X		X	XX	X	X	X
<i>Scolecia meandria</i> Radtke	“vermicular borings” (Zeff and Perkins, 1979) (cyanophyte)							X	
? <i>Planobola</i> <i>maurogota</i> Schmidt	cyanobacterium? alga?					X	X	X	
? <i>Cavernula pediculata</i> Radtke	<i>Gomontia polyrhiza</i> (Lagerhein) Bornet and Flahault (chlorophyte)				X	X			X
<i>Ichnoreticulina elegans</i> (Radtke)	<i>Ostreobium quekettii</i> Bornet and Flahault (chlorophyte)		XX		XX	X	X		X
<i>Rhopalia catenata</i> Radtke	<i>Phaeophila dendroides</i> Cronan (chlorophyte)		=		X		X		
<i>Orthogonum fusiferum</i> Radtke	<i>Ostracoblabe implexa</i> Bornet and Flahault (fungus)				X			X	
<i>Orthogonum lineare</i> Glaub	?fungus				=	XX	X	X	X
<i>Orthogonum</i> isp. (Form 1 in Wisshak et al., 2005)						X			
<i>Saccomorpha clava</i> Radtke	<i>Dodgella priscus</i> Zebrowski (fungus)	=			X	=	XX	X	XX
	Coccoid bacteria						XX	X	X

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Table 2. Seawater composition at the four sampling stations.

		PP	LC	PV	PA
pH		8.1	8.2	8.1	7.9
Eh	V	0.35	0.36	0.34	0.37
T	°C	27.7	27.9	29.6	30.6
Ca	mol l ⁻¹	0.011	0.012	0.011	0.011
Mg	mol l ⁻¹	0.058	0.059	0.057	0.058
K	mol l ⁻¹	0.012	0.012	0.012	0.012
Na	mol l ⁻¹	0.480	0.482	0.472	0.483
S04	mol l ⁻¹	0.037	0.040	0.043	0.037
Cl	mol l ⁻¹	0.604	0.592	0.564	0.564
HCO ₃	mol l ⁻¹	0.002	0.002	0.002	0.002
Br	mol l ⁻¹	0.001	0.001	0.001	0.001
Ba	mol l ⁻¹	0.053	0.052	0.058	0.076
Cd	mol l ⁻¹	0.002	0.002	0.002	0.004
Li	mol l ⁻¹	19.882	20.602	19.882	20.314
Mo	mol l ⁻¹	0.125	0.136	0.125	0.125
Pb	mol l ⁻¹	0.010	0.014	0.016	0.016
Sr	mol l ⁻¹	91.589	93.460	93.232	94.122
Zn	mol l ⁻¹	0.061	0.076	0.061	0.076

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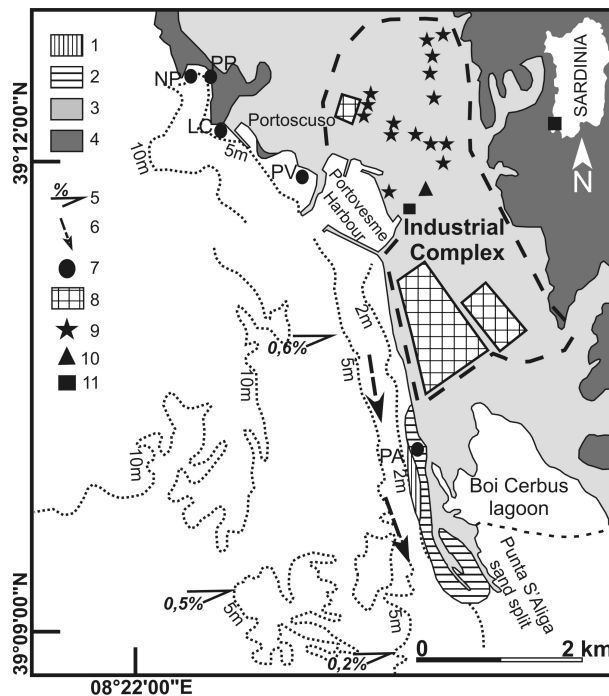


Fig. 1. Study area, location of sampling stations (Porto Pauleddu – NP, Porto Paglietto – PP, La Caletta – LC, Portovesme – PV, Punta S'Aliga – PA) and Industrial Complex of Portoscuso (SW Sardinia). 1. Submarine beach (grain size mean 0.13–0.18 mm). 2. Submarine beach (grain size mean 0.18–2.00 mm). 3. Alluvial deposits (Pleistocene–Holocene). 4. Volcanics (Oligo-Miocene). 5. Seabottom slope (%) from shoreline to 5 m isobath. 6. Longshore drift. 7. Sampling stations. 8. Red mud dumps. 9. Chimneys. 10. Lead-zinc smelter. 11. Electric power plant (1, 2, 5, 6 from Di Gregorio et al., 1996; modified).

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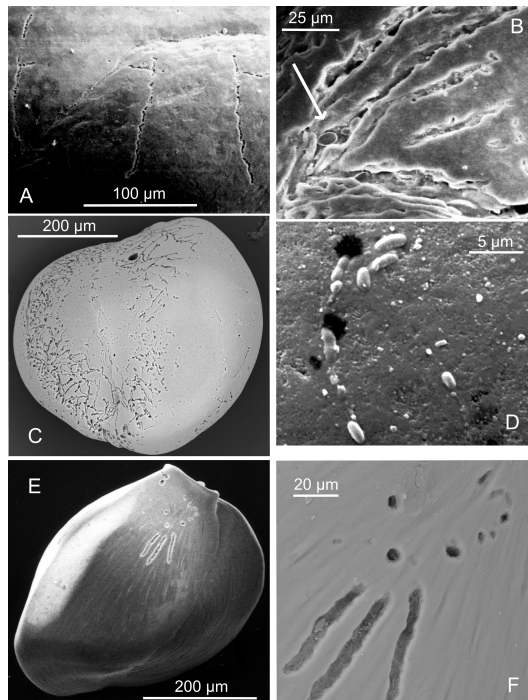


Fig. 2. SEM images of surface bioerosion of porcelanaceous imperforate foraminifera. **(A)** Tubular wavy tunnels parallel to substrate in *Pseudotriloculina rotunda* (d'Orbigny) exhibiting a rectangular ramification belonging to *Orthogonum* (? Form 1 in Wisshak et al., 2005). **(B)** Invasive infestation of microborings concentrated along the sutures of the chamber walls producing large galleries frequently occupied by pennate diatoms (arrow) in *Quinqueloculina* sp. **(C)** Dense boring pattern made up of fungal filaments developed in *Triloculina* cf. *marioni* Schlumberger. **(D)** Colony of rod-shaped cryptoendolithic bacteria made into previous holes of larger diameter in the wall surface of *Adelosina* sp. **(E)** Tubular borings, 4–5 μm in diameter, running parallel to surface of the test and penetrating into the wall of *Quinqueloculina ungeriana* d'Orbigny. **(F)** Enlargement of **(E)**.

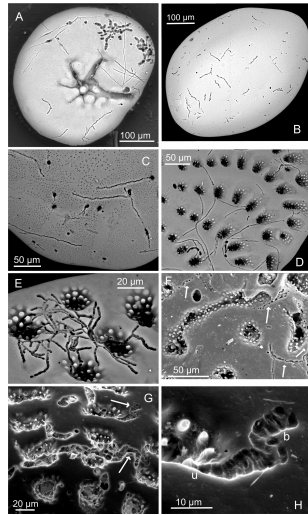


Fig. 3. SEM images of surface bioerosion of hyaline perforate foraminifera. **(A)** *Rosalina bradyi* Cushman (umbilical side) exhibiting undeterminable filaments and, at right, two bunches of radiating tunnels (cf. *Fascichnus*). **(B)** *Polymorphina* sp. (oblique side view) showing filaments of undeterminable borings in the whole test. **(C)** Detail of *Ammonia tepida* (Cushman) spiral side intensively bored by long filaments of fungal hyphae with reproductive organs (cf. *Saccomorpha clava*). **(D)** *Elphidium crispum* (Linnaeus) showing in dorsal view a boring pattern of curved thin tunnels (1–2 μm in diameter) belonging to ?*Scolecia filosa*, going towards the primary pores of the foraminifer. **(E)** Densely ramified boring pattern of *Ichnoreticulina elegans*, characterized by dichotomous branching goes into the primary pores of *E. crispum*. **(F)** Invasive colonies of undeterminable microborings (back? – cyanobacteria?) around and into primary pores of *E. crispum* (arrows) producing a lateral enlargement of these until two-three contiguous pores join (cf. Freiwald, 1995, Fig. 4). **(G)** Boring system developed among contiguous pores of *E. crispum*, characterized by large tunnels (arrows). **(H)** Enlargement of G showing large microbial overprint going into the pore and its transition from uniseriate (u) to biseriate (b) cell arrangements, belonging to *Fascichnus*.

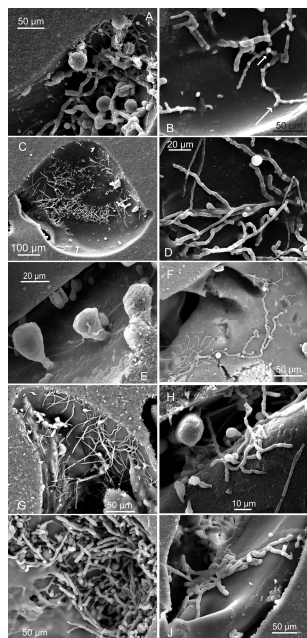


Fig. 4. SEM images of porcelanaceous foraminifera resin-casts. **(A)** Chamber-wall of miliolid densely bored by *Eurigonum nodosum* and globular *Saccomorpha clava*. **(B)** *E. nodosum* with diagnostic lateral heterocysts (arrows) inside the thick chamber wall of miliolid. **(C)** Dense assemblage of euendolithic borings in miliolid test. **(D)** Enlargement of C showing *E. nodosum* with diagnostic heterocysts. **(E)** Pavement of *Saccomorpha clava* into miliolid chamber wall. **(F)** Colony of *Scolecia meandria* in miliolid chamber wall. **(G)** Microbial pavement in miliolid chamber wall exhibiting *Scolecia filosa*, *E. nodosum* and fungal sack-shaped cavities. **(H)** Diversified ichnotraces belonging to *S. clava*, *Planobola macrogota* and *Ichnoreticulina elegans* in miliolid test. **(I)** Dense colonies of *E. nodosum* in miliolid test. **(J)** Colonies of *E. nodosum* inside the thick chamber wall of miliolid.

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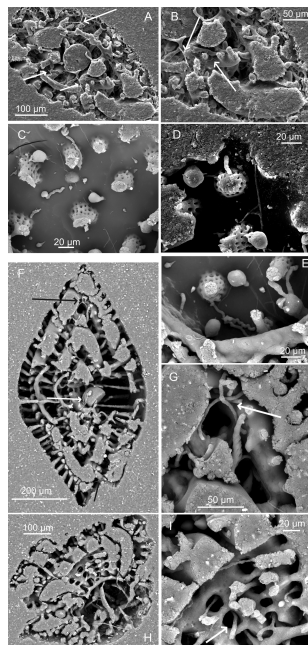


Fig. 5. SEM images of hyaline foraminifera resin-casts. **(A)** Euendoliths (arrows) inside the skeletal structure of *Elphidium crispum* (Linnaeus). **(B)** Enlargement of A showing undeterminable filaments. **(C)** and **(D)** *Saccomorpha clava* and undeterminable cyanobacteria boring skeletal structure of *E. crispum*. **(E)** Enlargement of **(D)** showing spherical cavities of *?Planulina macrogota* and sack-shaped cavity of *?Cavernula*. Morse, J. W.: The kinetics of calcium carbonate dissolution and precipitation, in: Carbonates: Mineralogy and Chemistry, edited by: Reeder, R. J., Mineralogical Society of America, Washington, D. C., 11, 227-264, 1983. **(F)** *E. crispum* (vertical section) showing undeterminable filamentous (arrows) in the complex skeletal pattern. **(G)** Enlargement of **(F)** showing filaments inside the skeletal of the foraminifera. **(H)** *E. crispum* (oblique section). **(I)** Enlargement of H showing tunnels (arrows) of borings in skeletal structures.

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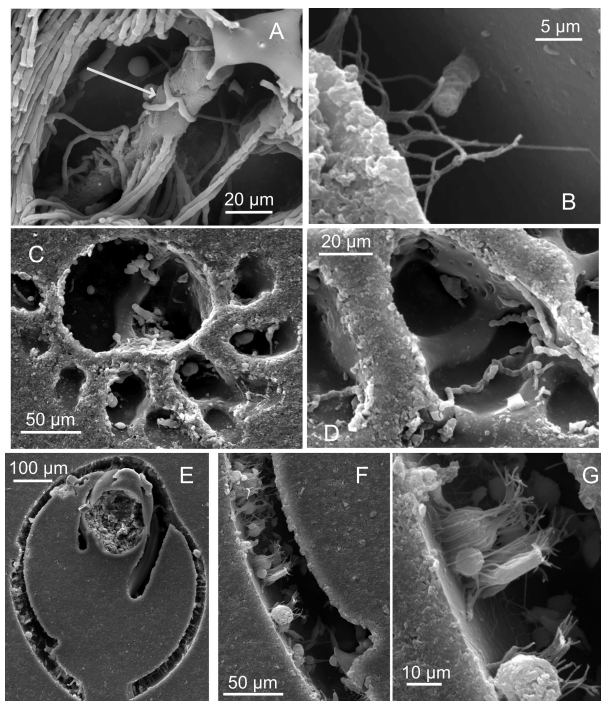


Fig. 6. SEM images of hyaline foraminifera resin-casts. **(A)** Tripartite gallery of undeterminable boring (?*Orthoanum* sp.) and globular-shape cavities in skeletal structure of *Ammonia beccarii* (Linnaeus). **(B)** Enlargement of **(A)** showing spherical cavities of ?*Planobola macrogota* and sack-shaped cavities of ?*Cavernula* surrounded by foraminiferal tubules. **(C)** Euendoliths inside the peripheral wall chambers of *Ammonia tepida* (Cushman). **(D)** Colony of *Eurigonum nodosum* in *Ammonia* sp. **(E)** Pavement of spherical cavities in the finely perforate test of *Polymorphina* sp. **(F)** and **(G)** Enlargement of **(D)** showing spherical cavities of ?*Planobola macrogota* and sack-shaped cavities of ?*Cavernula* surrounded by foraminiferal tubules.

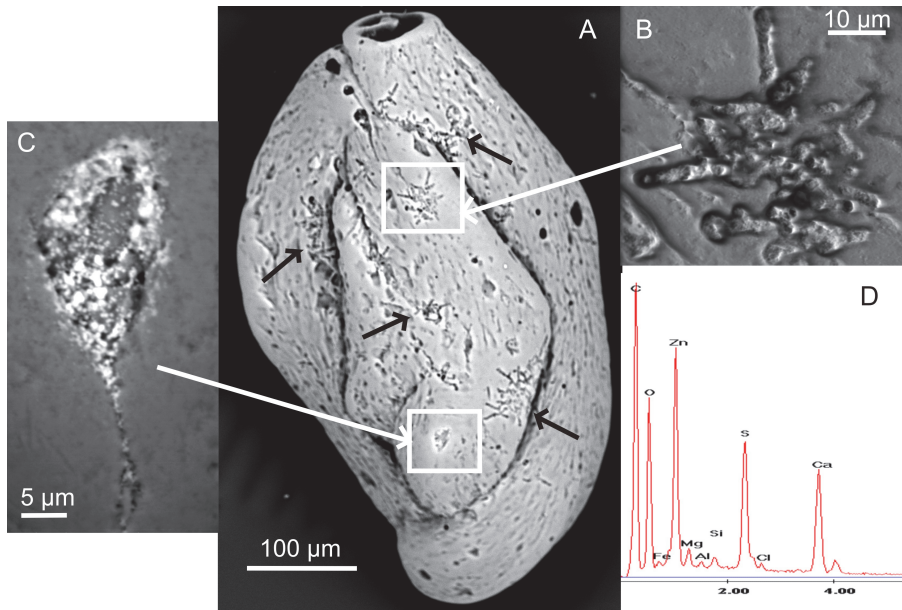


Fig. 7. SEM images of porcelanaceous foraminifera. **(A)** *Quinqueloculina* cf. *berthelotiana* d'Orbigny intensively bored by euendoliths (black arrows) among which are colonies of *Fascichnus* cf. *dactylus*. **(B)** Enlargement of **(A)** exhibiting a well-developed colony of *F.* cf. *dactylus* characterized by tunnels radiating from a central area. **(C)** Enlargement of **(A)** showing dome-shaped cavity of ?*Cavernula pediculata* with rhizoidal appendages, Zn biomineralized. **(D)** EDX analysis of dome-shaped cavity of ?*Cavernula pediculata*.

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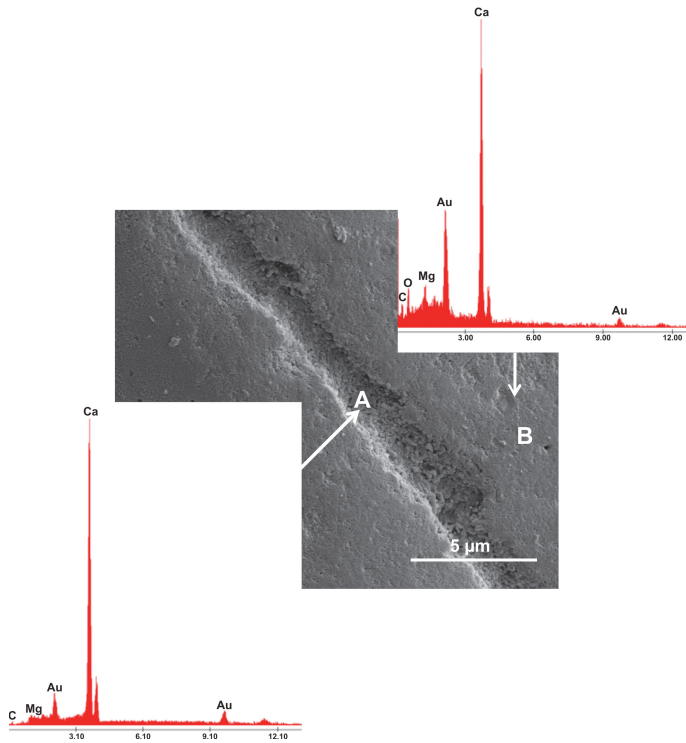


Fig. 8. SEM FEG images of porcelanaceous foraminifera (*Quinqueloculina* sp.). EDX analysis shows that Mg peak at the bottom of boring (A) is significantly lower than on unaltered shell surface (B).

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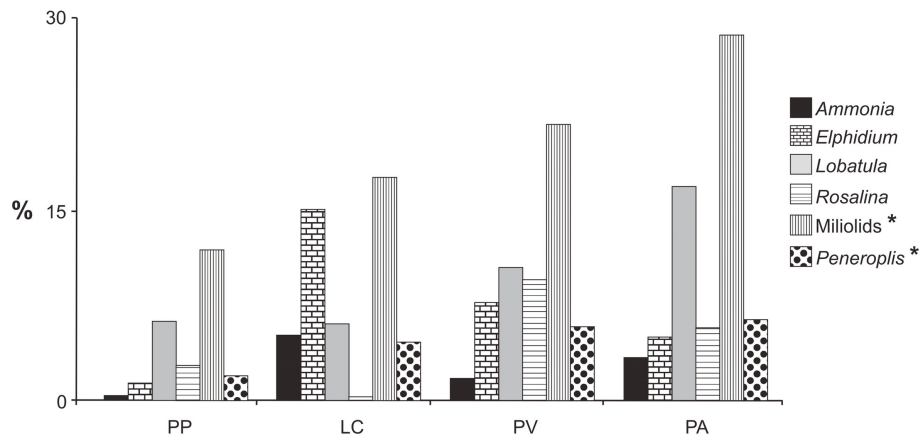


Fig. 9. Occurrence of bioerosion percentages in selected taxa belonging to porcelanaceous (high-Mg) imperforate miliolids and peneroplids (*) and hyaline (low-Mg) perforate foraminifera (*Ammonia*, *Elphidium*, *Lobatula*, *Rosalina*) from sampling stations (PP, LC, PV, PA).

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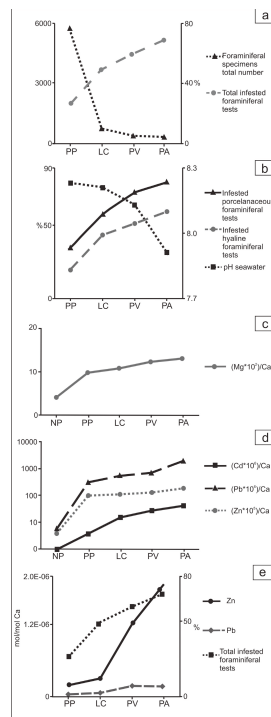


Fig. 10. Bioerosional percentages in microbially infested foraminiferal tests from Portoscuso coastal environment. **(a)** Correlation between infested foraminifera percentages and total number of foraminiferal tests. **(b)** Correlation between porcelanaceous high-Mg and hyaline low-Mg tests and sea water pH. **(c)** Mg/Ca ratio in foraminiferal tests increases in more highly polluted sites. **(d)** Cd/Ca, Pb/Ca and Zn/Ca ratio in foraminiferal tests increase in more highly polluted sites. **(e)** Increase of heavy metal values in sediments corresponds to increase in microbial infestation.