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## SPECIAL

Modern seawater acidification: the response of foraminifera to high-CO<sub>2</sub> conditions in the Mediterranean Sea

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he seas around the island of Ischia (Italy) have a lowered pH as a result of volcanic gas vents that emit carbon dioxide from the sea floor at ambient seawater temperatures. These areas of acidified seawater provide natural laboratories in which to study the long-term biological response to rising  $CO_2$  levels. Benthic foraminifera (single-celled protists) are particularly interesting as they have short life histories, are environmentally sensitive and have an excellent fossil record. Here, we examine changes in foraminiferal assemblages along pH gradients at  $CO_2$  vents on the coast of Ischia and show that the foraminiferal distribution, diversity and nature of the fauna change markedly in the living assemblages as pH decreases.

Short-term laboratory experiments indicate that, with rising atmospheric pCO<sub>2</sub> and a lowering of oceanic pH, many organisms with calcareous shells may be unable to survive the next 100 years (Orr et al. 2005: Doney et al. 2009: Moy et al. 2009: Veron et al. 2009) although others appear to be resistant (Findlay et al. 2009; Ries et al. 2009). A wide range of benthic macroorganisms (corals, barnacles, gastropods, echinoderms, seagrasses, calcareous and non-calcareous algae) living in the shallow-water environments around the coast of Ischia (Bay of Naples, Italy) show marked responses to increasing levels of CO<sub>2</sub> in the water (Hall-Spencer et al. 2008; Martin et al. 2008; Rodolfo-Metalpa et al. 2010). Around Castello Aragonese (Fig. 1) shallow-water habitats have zones with increasing CO<sub>2</sub> levels where mean pH values (measured in Total Scale at the sediment-water interface) range from 8.14 to 6.57 (for details, see Hall-Spencer et al. 2008). These waters provide a location at which predicted effects of ocean acidification (Caldeira & Wickett 2005; Royal Society 2005; IPCC 2007; Feely et al. 2004) can be tested by a study of the distribution of the living biota. The seawater adjacent to Ischia is acidified by gas



**Fig. 1.** Location of Ischia and sampling locations on the Castello Aragonese. Samples were collected from sites on traverses undertaken both north and south of the bridge and the foraminiferal assemblages were compared.

comprising 90.1–95.3% CO<sub>2</sub>, 3.2–6.6% N<sub>2</sub>, 0.6–0.8% O<sub>2</sub>, 0.08–0.1% Ar and 0.2–0.8% CH<sub>4</sub> (no sulphur). The salinity of the water (38‰) and total alkalinity (2.5 mequiv. kg<sup>-1</sup>) are uniform between all the sampling stations and the annual temperature range is 13–25 °C (Hall-Spencer *et al.* 2008).

Samples and methods. We have studied the assemblages of foraminifera from samples collected in the shallow waters around Ischia. We used a trowel to collect 1 cm deep 200 ml surface sediment samples at 2 m depth from areas with mean ambient pH levels of 8.14, 7.87, 7.83, 7.09 and 6.57. The sediment samples were collected by diving, and they were stored in buffered formalin, washed on a 63  $\mu$ m sieve, stained with Rose Bengal (to distinguish live from dead individuals), re-washed and dried at <40 °C. Normal micropalaeontological dry sieving and counting techniques were employed. The foraminifera were identified (in the first instance using Cimerman & Langer 1991) and imaged using a JEOL 5600 scanning electron microscope with a digital capture system.

The living assemblages from the sea-floor areas with a 'normal' pH of *c*. 8.14 contain a range of taxa dominated by miliolids, as would be expected in a shallow-water Mediterranean environment with a salinity of 38‰ (Murray 2006). The dominant taxa are *Elphidium* sp. cf. *E. advenum*, *Triloculina tricarinata*, *Pyrgo* sp., *Miliolinella elongata* and *Peneroplis planatus* (Fig. 2). Samples (S2 and N2 in Fig. 1) with lowered pH (7.87–7.83) contain *Miliammina fusca*, *Trochammina inflata*, *Ammonia tepida* and *Reophax* sp. Samples (S3 and N3 in Fig. 1)



Fig. 2. Selected species from Ischia, Italy. 1, Lobatula lobatula; 2, Cibicides refulgens; 3, Elphidium aculeatum; 4, Ammonia tepida; 5, Peneroplis planatus; 6, Laevipeneroplis karreri; 7, Planorbulina mediterranensis; 8, Rosalina bradyi; 9, Parrina bradyi; 10, Miliolinella labiosa; 11, Quinqueloculina seminula; 12, Triloculina tricarinata; 13, Spiroloculina ornata; 14, Sigmoilinita costata; 15, Miliammina fusca; 16, Textularia sp. cf. T. bocki. Scale bar represents 100 µm. It is important to note that, in the case of specimens collected in the areas of normal pH, the preservation is excellent.

from areas with the highest  $CO_2$  levels (mean pH 7.09–6.57) contain (at a pH of 7.6) an impoverished assemblage of agglutinated foraminifera that includes *Trochammina inflata*, *Miliammina fusca*, *Textularia* sp. cf. *T. bocki* and *Ammoglobigerina globigeriniformis*.

**Impact of acidification.** The reduction of the foraminiferal diversity (and abundance) mirrors that recorded for macrobenthic calcifying organisms (Hall-Spencer *et al.* 2008; Martin *et al.* 2008). Figure 3 shows that diversity falls from 24 species to four species from the normal marine samples towards the areas with high  $CO_2$  levels. The foraminiferal assemblage also shifts from one dominated by calcareous forms to one dominated by agglutinated taxa within a distance of <200 m as a result of the acidification of the water by  $CO_2$  with no other measurable gradients; depth, salinity, temperature, granulometry and light levels were similar across all sites (Hall-Spencer *et al.* 2008). The change in the assemblages is illustrated by use of the

triangular diagram developed by Murray for the investigation of foraminiferal ecology (Murray 1991*a*, pp. 232–244; Murray 1991*b*). The samples from Ischia with normal pH are seen to plot in the hypersaline field (dominated by miliolids), but with reducing pH the data points migrate towards the sector with 100% agglutinated (textulariid) taxa (Fig. 4).

**Implications for the fossil record.** Samples with *c*. 100% agglutinated assemblages of foraminifera are well known from the geological record (Scott *et al.* 1983; Jones 1988; Charnock & Jones 1990; Czarniecki 1993) and there has been much debate as to whether these are primary (a response to the environment) or secondary (the result of dissolution during taphonomy or diagenesis) (see Hart 1983, pp. 251–263, fig. 3). In the 1990s Alve and Murray conducted a series of acidification experiments to test the fate of a 'normal' foraminiferal assemblage when subjected to treatment with weak acid (Murray 1989, 2006; Alve & Murray 1994, 1995; Murray & Alve 1994, 1999*a*,*b*, 2000). The assem-



**Fig. 3. (a)** Graph of changing composition of the living assemblage (calcareous foraminifera:agglutinated foraminifera) across the range of pH values: (b) diversity (measured as total number of living species) across the range of pH values.



**Fig. 4.** Triangular diagram plotting miliolid taxa, agglutinated taxa and rotaliid taxa, which shows the changing composition of the living assemblage with the recorded changes in pH. It should be noted that the change in pH required to completely change the nature of the assemblage is relatively significant (from pH 8.2 to pH 7.8) although intermediate values have not yet been investigated.

blages generated (acid-treated assemblages or ATAs) by Alve & Murray (1995) were, not surprisingly, composed of 100% agglutinated taxa lacking calcareous cement. In the case of the samples from Ischia, the change from a calcareous-dominated assemblage (75%) to one with no calcareous taxa appears to be entirely due to changing pH and not post-death acidification during burial and diagenesis. In another example of the dominance of agglutinated taxa in an assemblage (Murray *et al.* 2003) there was a restriction caused by depleted levels of dissolved oxygen in the water column (though not anoxia) and pH was not recorded.

The Palaeocene-Eocene Thermal Maximum (PETM) is one of the most significant events in the Cenozoic and there is a body of evidence to suggest that there was a period of ocean acidification, which, in turn, affected the marine isotope record that is based on benthic foraminifera (Spero et al. 1997; Zachos et al. 2005; Pagani et al. 2006). The data from Ischia certainly confirm that a change in pH could cause a marked change in the benthic assemblage and even 'extinction' from a geological succession (Speijer et al. 1996; Zachos et al. 2005; Kaiho et al. 2006; Giusberti et al. 2009). Nguyen et al. (2009) have recently reported on dissolution experiments (using water with a pH value as low as 6.6) aimed at understanding the events during the PETM. The data from Ischia, however, provide information on the pH conditions under which the living assemblage is affected, rather than providing information on post-mortem dissolution. Presence or absence of foraminifera at the PETM must be a combination of both a biological restriction and a diagenetic impact.

**Summary.** The living (and dead) foraminiferal assemblages showed significant changes resulting from  $CO_2$  acidification of the shallow waters around Ischia. The assemblages in normal conditions (pH 8.2–8.14) were dominated by calcareous forms including abundant miliolids (*Triloculina*, *Pyrgo*, *Miliolinella*, *Quinqueloculina* and *Peneroplis*). The most impoverished assemblages (pH 7.6) were all agglutinated species of *Trochammina*, *Miliammina*, *Textularia* and *Ammoglobigerina*. This work confirms that, in open-water marine conditions, foraminifera are sensitive to the effects of ocean acidification, thereby indicating that rising levels of atmospheric  $pCO_2$  may cause significant changes in these assemblages. It also confirms that changes in pH could have caused the extinctions recorded at events such as the PETM.

**Appendix.** Foraminifera recorded in the samples from Ischia. The original author names and dates are given (for references, see Cimerman & Langer 1991).

Normal pH 8.2-8.14

Ammonia inflata (Seguenza 1862); Brizalina sp.; Cibicides advenum (d'Orbigny 1839); Cibicides refulgens Montfort 1808; Cycloforina tenuicollis (Wiesner 1923); Elphidium aculeatum (d'Orbigny 1846); Elphidium sp. cf. E. advenum (Cushman 1922); Lepidiodeuteramina sp.; Massilina gualtieriana (d'Orbigny 1839); Miliolinella elongata Kruit 1955; Miliolinella labiosa (d'Orbigny 1839); Parrina bradyi (Millett 1898); Peneroplis pertusus (Forskål 1775); Peneroplis planatus (Fichtel & Moll 1798); Planorbulina mediterranensis d'Orbigny 1826; Pyrgo sp.; Quinqueloculina berhelotiana d'Orbigny 1839; Quinqueloculina jugosa Cushman 1944; Quinqueloculina seminula (Linné 1758); Rosalina vilardeboana d'Orbigny 1839; Triloculina tricarinata d'Orbigny 1826; Triloculina sp.

Medium pH 7.8

Ammonia tepida (Cushman 1926); Miliammina fusca (Brady 1870); Reophax sp.; Trochammina inflata (Montagu 1803).

Low pH 7.6

Ammoglobigerina globigeriniformis (Parker & Jones 1865); Miliammina fusca (Brady 1870); Textularia sp. cf. T. bocki Höglund 1947; Trochammina inflata (Montagu 1803).

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