High risk of extinction of benthic foraminifera in this century due to ocean acidification

S. Uthicke¹, P. Momigliano¹² & K. E. Fabricius¹

¹Australian Institute of Marine Science, PMB No 3, Townsville, Queensland 4810, Australia, ²Department of Biological Sciences, Macquarie University, Sydney, 2109 NSW, Australia.

Increased atmospheric CO₂ concentrations lead to decreased pH and carbonate availability in the ocean (Ocean Acidification, OA). Carbon dioxide seeps serve as ‘windows into the future’ to study the ability of marine invertebrates to acclimatise to OA. We studied benthic foraminifera in sediments from shallow volcanic CO₂ seeps in Papua New Guinea. Conditions follow a gradient from present day pH/pCO₂ to those expected past 2100. We show that foraminiferal densities and diversity declined steeply with increasing pCO₂. Foraminifera were almost absent at sites with pH < 7.9 (<700 μatm pCO₂). Symbiont-bearing species did not exhibit reduced vulnerability to extinction at pH < 7.9. Non-calcifying taxa declined less steeply along pCO₂ gradients, but were also absent in samples at pH < 7.9. Data suggest the possibility of an OA induced ecological extinction of shallow tropical benthic foraminifera by 2100; similar to extinctions observed in the geological past.
Results
Seawater chemistry contrasted strongly between the 12 control and seep sites, with mean values generally ranging from 8.08 to 7.52 pH units and 369 to 11603 µatm pCO₂, respectively (Supplementary Table 2). One extreme site assumed pH values of ~7.0 and pCO₂ values of >5000 µatm pCO₂. With the exception of that site, calcite and aragonite saturation values at each location remained well above 1.0 (the value at which carbonate theoretically dissolves). Mean total alkalinity was 2326 µmol kg⁻¹ (N = 134, SD = 66). Thus 95% of all alkalinity values were between 2197 and 2456 µmol kg⁻¹, a range we used together with the pH measurements from all stations to calculate pCO₂ concentrations (Fig. 1 A). These pH measurements showed a good correlation to the long term averages for the 12 core sites (Supplementary Table 2, R² = 0.89, p < 0.0001).

Concentrations of inorganic carbon (IC) in the sediments declined significantly with declining pH (linear model, Fig. 1 B, Table 1), from nearly 100% carbonate sediment (IC = 12%) at ambient pH to nearly zero in most samples below pH 7.7. Sediment organic carbon and nitrogen were unrelated to pH (C: R² = 0.01, p = 0.58; N: R² < 0.01, p = 0.99). Data found were on a low level typical for coral reef sediments (organic-C: average = 0.28%, SD = 0.21%; N: average = 0.03%, SD = 0.02%). Because of the low level of organic content, the fact that values do not differ between seeps and control areas and that most foraminifera investigated are epibenthic or epiphytic it is unlikely that dissolution driven by interstitial respiratory processes contributed to the observed patterns.

Water collected over the benthos showed no significant (ANOVA, p = 0.1509) difference in the concentration of arsenic between control (mean = 1.62 µg L⁻¹, SD = 0.08 µg L⁻¹) and high (mean = 1.88 µg L⁻¹, SD = 0.50 µg L⁻¹) CO₂ sites, and values were in the range on typical marine samples (1–2 µg L⁻¹). Sediments had slightly higher arsenic concentrations directly at the main seeps (which emit almost pure CO₂) compared with the control sites (Seeps: mean = 12.8 mg kg⁻¹, SD = 9.8 mg kg⁻¹, N = 4; Controls: 7.1 mg kg⁻¹, SD = 1.8 mg kg⁻¹, N = 5), most likely reflecting the low carbonate content in the seep sediments. Concentrations in sediments were low compared to those at other volcanic vents (e.g., Ambitle Island, PNG: range: 52–33,200 mg kg⁻¹). In addition, diverse and abundant foraminiferal communities have been described under sediment and water column arsenic concentrations
Foraminifera were abundant at control sites (mean densities: 93 individuals g$^{-1}$ sediment, SD = 54 individuals g$^{-1}$ sediment). Densities declined steeply with increasing pCO$_2$ (Table 1, Fig. 1 C), and foraminifera were almost absent at pCO$_2$ conditions predicted for the end of this century under all but the most optimistic emission scenarios (RCP2.6, peak at 440 µatm pCO$_2$; Fig. 1 A, C). The density of mixotrophic and heterotrophic taxa decreased at the same rate, and their slopes of decline did not differ significantly (Table 1).

In other words, both groups followed the same trajectory towards density declines which appeared to increase in relative importance with elevated pCO$_2$. However, agglutinate taxa were too rare to fill the niches vacated by calcifying taxa. The loss for the end of this century under all but the most optimistic emission scenarios (Table 1). The loss of diversity was significantly slower for agglutinate taxa, but only three taxa were recorded.

Additional dive searches (>5 h total) at Upa-Upasina for living large mixotrophic foraminifera on algal, seagrass and coral rubble substrata revealed that several species can be readily found at the control site (Amphistegina spp., Calcarina spp., Heterostegina depressa, Marginopora vertebralis), whereas none could be detected at locations with reduced (<~7.9) pH values.

A permutational multivariate analysis of variance (Permanova) indicated significant differences in foraminiferal assemblage composition between the three sampling locations (Supplementary Table 4). The main difference between locations was that mixotrophic species were dominant at one of the locations (Upa-Upasina) and heterotrophic species dominated at the other two locations (Supplementary Table 5). A distance-based redundancy analysis (dbRDA) showed that the relative contribution of most taxa to the foraminiferal assemblage did not change along the pCO$_2$ gradient (Supplementary Fig. 1). pCO$_2$ explained only a small (5.2%) albeit significant proportion of the variation in relative abundances between samples (Pseudo-$F$ = 2.41, p = 0.0133). This confirmed that most taxa disappeared at a similar rate with increasing pCO$_2$.

Exceptions were three species (two Elphidium spp. and Amphistegina lessonii) which increased slightly in relative importance at intermediate pCO$_2$ (Supplementary Fig. 1).

Closer inspection of the three samples that had intermediate foraminiferal densities at <7.9–8.0 pH (Fig. 1) showed that most of their foraminifera were Amphistegina spp. and Elphidium spp., i.e., taxa that appeared to increase in relative importance with elevated pCO$_2$. However, many of these specimens had a corroded or pitted appearance (Fig. 2). This appearance was not detected in any of the control samples in Milne Bay at present-day pCO$_2$ conditions, or in sediment core slices >1000 yr of age from the Great Barrier Reef (Fig. 2). The pitted appearance is similar to that observed in another Amphistegina species under experimental CO$_2$ increase.

### Discussion

Foraminiferal density and diversity at the control sites in PNG were high and similar to those observed on the Great Barrier Reef and other sites in PNG. Our data show no sign of acclimation by benthic foraminifera to high pCO$_2$, although a proportion of genotypes will have lived near the seeps for many generations (their reproduction is partly non-dispersive asexual, as well as through gametes). Our analyses along the pH/pCO$_2$ gradients around the seeps in PNG suggested that all common tropical foraminiferal species will likely be ecologically extinct at the CO$_2$ conditions predicted for the year 2100, except under the most optimistic scenario. However, that scenario is unlikely, requiring immediate drastic emission cuts and negative net emission (i.e. carbon sequestration) in the second half of this century.

Several mass extinctions of deep sea benthic foraminifera occurred in the geological past, most of which were linked to increased pCO$_2$ and/or temperature, but some geological studies from shallow reef environments also observed increased foraminiferal dominance when corals became rare. None of these previous extinctions were as severe as the ecological or even taxonomic extinction in shallow carbonate areas we predict. Previous natural pCO$_2$ increases occurred one to two orders of magnitude slower and were associated with less reduced calcite or aragonite saturation states than the anthropogenic increases presently observed.

Contrary to our initial hypothesis, no shift towards mixotrophic taxa was observed. We assumed that photosymbiont-bearing taxa might be less vulnerable to OA because carbon fixation may increase pH in their diffusive boundary layer; this can lead to pH differences of >0.8 units between day and night. A recent study in foraminifera, however, showed that these increases cannot compensate for decreased pH in ambient water. We hypothesized that agglutinate taxa which do not rely on calcification might replace calcifying species. In our study, diversity and density of agglutinate taxa declined less steeply than calcifying taxa. However, agglutinate taxa were too rare to fill the niches vacated by calcifying taxa.

Carbonate sediments at seeps disappeared at calcite or aragonite saturation states >1.0 (Supplementary Table 2). Previous experiments have shown test dissolution and corrosion in temperate heterotrophic foraminifera at >900 µatm pCO$_2$; although one species.
still calcified at 1900 \( p\text{CO}_2 \) \( \mu \text{atm} \). However, our electron microscopy and sediment inorganic carbon data showed corrosion of foraminifera at 7.9–8.0 pH, and absence of biogenic carbonate accumulation, suggesting that carbonate dissolution began at \( p\text{CO}_2 \) levels of 320\( \mu \text{atm} \). Dissolution of foraminifera and sediment was previously observed under elevated \( p\text{CO}_2 \) and can start at aragonite saturations states above 333. Indeed, most biogenic carbonates dissolved at much higher saturation values than predicted from abiotic carbonates. Thus, in addition to the described direct ecological impacts, the loss of benthic foraminifera, together with dissolution and loss of biogenesis of carbonate by other organisms under near-future \( p\text{CO}_2 \) conditions may also have far-reaching ecological flow-on effects.

**Methods**

50 sediment samples were collected during two expeditions to the seep sites in August 2010 and April 2011. Samples were collected at various distances from the three seeps (Esa’Ala, Upa-Upasina, Dobu Island) and their control sites. Samples of the top 1 cm sediment were sourced from 4 to 15 m depth. At each site, three haphazard samples were taken, and pooled into one sample.

For the estimation of foraminiferal abundances, sediments were rinsed over a 63 \( \mu m \) sieve. After drying (\( >24 \) h, 60°C), all foraminifera were collected from subsamples until 200 specimens per sample were obtained. This yield could not be achieved for some of the samples collected near the seep sites. In that case, all available sediment was searched. We collected only intact specimens which showed no sign of degradation and little damage (‘optimally preserved’, sensu\(^1\)) under the dissection microscope at 25× magnification. These specimens are regarded as a good representation of the present-day (time averaged over the last few years) biocoenosis. Taxa were determined under a dissection microscope, following \textit{Uthicke et al.}, 2010\(^2\). The dry weights of both sediment and foraminifera were determined to calculate foraminiferal densities. A subsample of foraminifera was observed via Scanning Electron Microscopy (SEM) in order to identify signs of corrosion. Organic matter was removed by incubating the tests in 2% NaClO for 1 h at 60°C. Foraminiferal tests were then dehydrated in 100% ethanol, mounted on SEM stubs and sputter-coated with gold and visualized on a JEOL JSM-5410LV scanning electron microscope.

A subsample of the sediment prior to rinsing and sieving was dried and ground for total carbon and nitrogen determination. Samples were analyzed on a Truspec C/N Analyser (Leco). After acidification (2 M hydrochloric acid), organic carbon in the sediments was measured using a TOC-V Analyser (Shimadzu, equipped with a SSM-5000A Solid Sample Module).

Arsenic concentrations in sediments were determined on acid digested (HNO\(_3/\)HClO\(_4\)) subsamples, by Hydride Generation Atomic Absorption Spectrometry using a Thermo SOLAAR M Atomic absorption spectrometer. To assess surface sediment temperatures, six temperature loggers (TidbiT v2, Hobo Data loggers) were deployed on the substrata of all seep and control sites for 363 days (April 2011 to April 2012; one logger from Upa-Upasina failed and data are reported for 5 loggers only). Seawater carbonate chemistry data were based on those presented in \( ^5 \), complemented by additional samples from April 2011 following the same procedures and based on total alkalinity and dissolved inorganic carbon (DIC) measurements.

Arsenic and 10 common heavy metal pollutants were analysed from triplicate (unless marked otherwise in Supplementary Table 3) samples from each of the seep and control sites on an ICP-MS using an octapole reaction system to limit matrix interferences. Samples for these analyses were collected in two opposite seasons, in May and December 2012.
Linear models were used to test for relationships between average pH and the dependent sediment chemistry variables, and foraminiferal total density and diversity, and density and diversity of individual groups. Models were fitted after log transformation (natural logarithm) of the dependent variable. The initial models included depth and pH, but depth was removed from the final models because it did not explain a significant amount of variation for any of the parameters (Supplementary Table 6). Slopes of two groups were compared by adding 'groups', pH and their interaction term into the model and accepting slopes as homogenous when the interaction term was non-significant.

Distance-based redundancy analysis (dbRDA) was used to investigate patterns in assemblage composition across samples. Initial permutation tests showed that depth had no effect of assemblage patterns (Pseudo-F = 1.62, p = 0.0958), but location (EsA/A, Upa-Upasina and Dobu Island) affected distributions (Pseudo-F = 6.57, p < 0.0001). These location effects were removed (partialled out) because they were not of interest in the context of our study. The mean pH value for each sample was converted to pCO2 and used as environmental variable. DBRDA was conducted on a Bray-Curtis distance matrix of fourth-root transformed relative abundance data. Single-factor permutational multivariate analysis of variance (PERMANOVA) was used to test the significance of differences in assemblage composition between sample locations. PERMANOVA post-hoc tests presented here were based on 10,000 permutations, using type III sums of squares and permutation of residuals under a reduced model. Subsequent to PERMANOVA, similarity percentage (SIMPER) was used to investigate which taxa contributed most to between-group differences. DBRDA and linear model analyses were performed in R. PERMANOVA and SIMPER analyses were conducted in Primer 6.0.

1. Feely, R. A., Doney, S. C. & Cooley, S. R. Ocean acidification: Present conditions and future changes in a high-CO2 world. Oceanography 22, 39–47 (2009).
2. Feely, R. A. et al. Impact of anthropogenic CO2 on the CaCO3 system in the oceans. Science 305, 362–366 (2004).
3. Ries, J. B. Skeletal mineralogy in a high-CO2 world. J. Exp. Mar. Biol. Ecol. 403, 54–64 (2011).
4. Uthicke, S. & Fabricius, K. Productivity gains do not compensate for reduced calcification under near-future ocean acidification in the photosynthetic benthic foraminifera Marginopora vertebrella. Global Change Biology 18, 2781–2791 (2012).
5. Fabricius, K. E. et al. Losers and winners in coral reefs acclimatized to elevated carbon dioxide concentrations. Nature Climate Change 1, 165–169 (2011).
6. Hall-Spencer, J. M. et al. Volcanic carbon dioxide vents show ecosystem effects of ocean acidification. Nature 454, 96–99 (2008).
7. Dias, B. B., Hart, M. B., Smart, C. W. & Hall-Spencer, J. M. Modern seawater acidification: the response of foraminifera to high-CO2 conditions in the Mediterranean Sea. Journal of Geological Society 167, 843 (2010).
8. Martin, S. et al. Effects of naturally acidified seawater on seagrass calcareous epibionts. Biology Letters 4, 689 (2008).
9. Porzio, L., Buia, M. C. & Hall-Spencer, J. M. Effects of ocean acidification on macroalgal communities. J. Exp. Mar. Biol. Ecol. 400, 278–287 (2011).
10. Johnson, V. R. et al. Responses of marine benthic microalgae to elevated CO2. Marine Biology online first, 1–12, doi:10.1007/s00227-011-1840-2 (2011).
11. Blackmon, P. D. & Todd, R. Mineralogy of some foraminifera as related to their classification and ecology. Journal of Paleontology, 1–15 (1959).
12. Fujita, K., Hikami, M., Suzuki, A., Kuroyanagi, A. & Kawahata, H. Effects of ocean acidification on calcification of symbiont-bearing reef foraminifera. Biogeosciences Discussions. 8, 1809–1829 (2011).
13. Vogel, N. & Uthicke, S. Calcification and photobiology in symbiont-bearing benthic foraminifera and responses to a high CO2 environment. J. Exp. Mar. Biol. Ecol. 424, 15–24 (2012).
14. Kuroyanagi, A., Kawahata, H., Suzuki, A., Fujita, K. & Irie, T. Impacts of ocean acidification on large benthic foraminifers: Results from laboratory experiments. Marine Micropaleontology 7, 190–195 (2009).
15. Reymond, C. E., Lloyd, A., Kline, D. I., Dove, S. G. & Pandolﬁ, J. M. Decline in growth of foraminifer Marginopora rossi under eutrophication and ocean acidification scenarios. Global Change Biology 19, 291–302, doi:10.1111/gcb.12035 (2013).
16. Moss, R. H. et al. The next generation of scenarios for climate change research and assessment. Nature 467, 747–756 (2010).
17. De Beer, D. & Larkum, A. Photosynthesis and calcification in the calcifying alga Halimeda disoida studied with microsensors. Plant, Cell & Environment 24, 1209–1217 (2001).
18. Kocher-Rink, S. & Kuehl, M. Microsensor studies of photosynthesis and respiration in larger symbiotic foraminifera. I. The physico-chemical microenvironment of Marginopora vertebralis, Amphistegina lobifera and Amphisorus hampichii. Mar. Biol. 137, 473–486 (2000).
19. Price, R. E. & Pichler, T. Distribution, speciation and bioavailability of arsenic in a shallow-water submarine hydrothermal system, Tutm Bay, Ambitile Island, PNG. Chemical geology 224, 122–135 (2005).
20. McManus, C. E., Farnham, P. M. et al. The IPCC greenhouse gas concentrations and their extensions from 1765 to 2300. Climatic Change 109, 213–241 (2011).
21. Uthicke, S., Patel, F. & Ditchburn, R. Elevated land runoff after European settlement perturbs persistent foraminiferal assemblages on the Great Barrier Reef. Ecology 93, 111–121 (2012).
22. McIntyre-Wrenning, A., Bernhard, J. M., McCorkle, D. C. & Hallock, P. Non-lethal effects of ocean acidification on the symbiont-bearing benthic foraminifer Amphistegina gibbosa. MEPS 472, 45–60 (2013).
23. Uthicke, S., Thompson, A. & Schaffelke, B. Effectiveness of benthic foraminiferal and coral assemblages as water quality indicators on inshore reefs of the Great Barrier Reef, Australia. Coral Reefs 29, 209–225 (2010).
24. Langer, M. R. & Lips, J. H. Foraminiferal distribution and diversity, Madang reef and lagoon, Papua New Guinea. Coral Reefs 22, 143–154 (2003).
25. Arora, V. K. et al. Carbon emission limits required to satisfy future representative concentration pathways of greenhouse gases. Geophys. Res. Lett. 38, L05805 (2011).
26. Zachos, J. C. et al. Rapid acidification of the ocean during the Paleocene-Eocene thermal maximum. Science 308, 1611–1615 (2005).
27. Hönisch, B. et al. The Geological Record of Ocean Acidification. Science 335, 1058–1063 (2012).
28. Scheiner, C. & Speijer, R. L. Paleocene-early Eocene Tethyan carbonate platform evolution: A response to long- and short-term paleoclimate change. Earth-Science Reviews 90, 71–102 (2008).
29. Glas, M. S., Fabricius, K. E., de Beer, D. & Uthicke, S. The O2, pH and CaCO3 microenvironment of benthic foraminifera in a high CO2 World. PLoS ONE 7, e50010 (2012).
30. Haynert, K., Schönfeld, J., Ries, J. B., Ichorn, U. & Polovodova, I. Biommetry and dissolution features of the benthic foraminifer Ammonia aomoriensis at high pCO2, Mar Ecol Prog Ser 432, 53–67 (2011).
31. Dessard, D., Nehrke, G., Reichart, G. J. & Bijma, J. Impact of seawater pCO2 on calcification and Mg/Ca and Sr/Ca ratios in benthic foraminifera calcite: results from culturing experiments with Ammonia tepida. Biogeosciences 81, 81–93 (2011).
32. Yamamoto, S. et al. Threshold of carbonate saturation state determined by CO2 control experiment. Biogeosciences 9, 1441–1450 (2012).
33. Yordanova, E. K. & Hohenegger, J. Taphonomy of larger Foraminifera: relationships between living individuals and empty tests on flat reef slopes (Sesoko Island, Japan). Facies 46, 169–204 (2002).

Acknowledgements
We thank the Councillors of the Dobu RLLG and the Traditional Owners of the seep sites for allowing us to survey their reefs. We are grateful for comments by R. Albright on an earlier draft. The research was funded by the Australian Institute of Marine Science.

Authors contributions
S.U. conducted and designed experiments, analysed data and wrote the MS. P.M. Analysed data and foraminiferal samples, and contributed to the writing. K.F. conceived the overall seep project and contributed to writing and analysis.

Additional information
Supplementary information accompanies this paper at http://www.nature.com/scientificreports

Competing financial interests: The authors declare no competing financial interests.

License: This work is licensed under a Creative Commons Attribution-NonCommercial-NoDerivs 3.0 Unported License. To view a copy of this license, visit http://creativecommons.org/licenses/by-nc-nd/3.0/

How to cite this article: Uthicke, S., Momigliano, P. & Fabricius, K.E. High risk of extinction of benthic foraminifera in this century due to ocean acidification. Sci. Rep. 3, 1769, DOI:10.1038/srep01769 (2013).