# Ocean Acidification-Category 1: Calibration and application of the boron isotope seawater-pH indicator in deep-water corals

Cold-water corals occur in all ocean basins and from shallow depths to the abyss, however, little is known about their long-term sensitivity to changing environmental conditions and ocean acidification. While the impact of ocean acidification has been studied extensively in shallow water corals [*Langdon et al.*, 2003; *Langdon and Atkinson*, 2005] the effects on deep-sea corals remain unexplored. Given the invasion of anthropogenic $CO_2$ in high latitude waters and the generally reduced carbonate ion saturation at greater water depth, deep-sea corals, and in particular the aragonitic, bioherm-forming scleractinians, may be particularly vulnerable to ocean acidification [*Guinotte et al.*, 2006]. This vulnerability may have important ecological consequences, as deep-sea coral mounds provide important habitat for a diversity of organisms. For instance, *Lophelia pertusa* reefs in the NE Atlantic have been shown to support over 1300 species [*ACES*, 2003], and *Oculina varicosa* reefs have been observed with over 350 species of invertebrates [*Koenig et al.*, 2000; *Reed*, 1992]. Of direct importance to humans, many commercial species of fish inhabit these reefs for feeding, protection and procreation [*Mortensen*, 2000; *Rogers*, 1999]. Efforts are needed to study the effects of changing seawater chemistry on these ecosystems, and in particular reconstructions of past pH-variations experienced by these corals may help to implement long-term management plans for deep-sea coral reefs.

A new and robust method is needed for reconstructing deep-water pH in this environment. The boron isotope proxy for seawater pH has been used successfully in foraminifers and tropical shallow water corals but not yet in cold-water corals. Here we propose

(1) to calibrate the boron isotope/paleo-pH proxy through analysis of live-collected cold-water corals and specimens cultured over a pH range in the laboratory

(2) to apply the B isotope proxy to reconstructing the magnitude and rate of anthropogenic $CO_2$ invasion into intermediate depths of the North Atlantic and Southern Ocean

(3) to reconstruct last glacial and deglacial pH from radiometrically dated Southern Ocean cold-water corals and evaluate their pH sensitivity.

We have assembled a range of modern corals for this study from recent oceanographic research cruises in the North Atlantic, Southern Ocean, and the Mediterranean. The calibration will include gorgonians, stylasterids, scleractinians and laboratory cultured specimens of the bioherm-forming scleractinian *Lophelia*. Additional cruises planned during the course of this project will collect *in situ* pH and live corals to complement this calibration sample set. The samples come from depths of 20-3460 m, covering a pH-range of 7.7–8.1, a temperature-range of -0.9 to 13.8ºC, and salinity 32.9-38.8.

Live collected corals from the North Atlantic and Southern Ocean will provide an ultra-high resolution temporal record of anthropogenic $CO_2$ invasion at intermediate depths, which can then be compared to model estimates and to changes in coral species composition through time. Dated corals from the same locations will be used to document pH changes over the last glacial and deglaciation. An age survey of the scleractinian

*Desmophyllum dianthus* in the Drake Passage has shown temporal changes in spatial abundance patterns, which may be related to changing seawater carbonate chemistry.

The boron isotopic composition of these samples will be analyzed by negative thermal ionization mass spectrometry (NTIMS) at LDEO and by multi collector-inductively coupled plasma mass spectrometry (MC-ICP-MS) at Bristol University. These paired analyses will be a key part of ongoing interlaboratory calibrations to ensure all measurements are robust. The data will be complemented by B/Ca ratios on the same material. The project will form the basis of the PhD thesis of Sarah Hale at LDEO and it will include collaborations with leading national (WHOI, Rutgers University) and international (Bristol University) geochemistry laboratories.

## INTRODUCTION

### Deep-sea corals in paleoceanography

Compared to shallow dwelling corals, deep-sea corals tend to live in near constant conditions of temperature, salinity and ocean currents, and photosynthesis does not occur due to the lack of sunlight. Scleractinian corals secrete aragonite, a more soluble form of calcium carbonate, whereas the skeletons of octocorals are made of calcite and organic structures; stylasterids can be made of either aragonite or calcite [*Cairns and Macintyre*, 1992]. Guinotte et al. [2006] noticed that reef-forming deep-sea corals primarily occur in locations that have above average surface water productivity but their abundance also decreases with the depth of the aragonite saturation horizon (ASH). At >2000m the ASH in the North Atlantic is very deep and many deep-sea coral bioherms are found there. In particular bioherms of the scleractinian *Lophelia pertusa* cover large but shallow areas off the coasts of Norway and Sweden. The ASH in the North Pacific is much shallower (50-600m) than in the Atlantic and scleractinians in the North Pacific do not appear to flourish or form bioherms. The area is dominated by octocorals and stylasterids instead. These differing distributions may be explained by sensitivity of deep-sea coral calcification to seawater saturation state. However, little experimental work has been done on deep-sea corals. Laboratory experiments are only published for shallow water corals, which, in some species, revealed substantial reductions in calcification rates upon seawater acidification [*Langdon et al.*, 2003; *Langdon and Atkinson*, 2005]. To assess the effect of ocean acidification on deep-sea corals, boron isotope reconstructions offer a powerful tool to estimate past variations in intermediate and deep seawater-pH. Because of their aragonitic skeleton scleractinians such as *L. pertusa* are likely to be most vulnerable but calcite saturation of the deep sea will also decrease and affect stylasterids and octocorals. For reconstructions of intermediate and deep-water chemistry, geochemical analyses will therefore best include all groups of calcifying deep-sea corals, to take advantage of their full depth range and cosmopolitan distribution. When and where scleractinian corals have appeared or disappeared in the geological record could thus be interpreted in the light of changing seawater acidity reconstructed from stylasterids and octocorals.

In comparison to reconstructions using benthic foraminifers from deep-sea sediments, the great advantage of deep-sea corals lies in their highly resolved records of past chemical conditions. Some species live for thousands of years [*Roark et al.*, 2009] and others for hundreds [*Adkins et al.*, 2004]. An additional advantage is that aragonitic scleractinian corals can be dated using U-series dating techniques [*Cheng et al.*, 2000; *Robinson et al.*, 2007] and all species incorporate carbon, allowing $^{14}C/^{12}C$ to be analyzed. In many groups optically defined growth bands provide the potential for paleoreconstructions of sub-decadal to annual resolution [*Sherwood et al.*, 2008].

Because the physicochemical conditions in the deep sea are fairly uniform and these corals bear no photosymbionts, their skeletons are useful for investigating biologically mediated vital effects. Large geochemical heterogeneities have been observed in the microstructures of scleractinian cold water corals, suggesting substantial vital effects during the calcification process of these organisms [*Adkins et al.*, 2003; *Blamart et al.*, 2007; *Gagnon et al.*, 2007]. For instance, Adkins et al. [2003] microsampled 6 deep-sea coral species and found variations in $\delta^{13}C$ as large as 10‰ and in $\delta^{18}O$ of up to 5‰ within single specimens. Cohen et al. [2006] studied Sr/Ca and Mg/Ca by SIMS in *L. pertusa* and observed seasonal variations larger than those observed in zooxanthellate tropical corals. Cohen et al. [2006] observed a correlation with aragonite precipitation efficiency and suggest variations in saturation state of the calcifying fluid must occur in these deep-water corals despite the lack of zooxanthellae. More relevant for this proposed study, Blamart et al. [2007] and Rollion-Bard et al. [2010] used ion microprobe analyses to study the boron and oxygen isotopic composition of *L. pertusa*. They found that both isotope ratios are depleted within the early mineralization zone, and enriched in the outer fibrous skeleton. The large magnitude of these variations (~6‰ in $\delta^{18}O$ and ~10‰ in $\delta^{11}B$) could not be explained by environmental conditions and if not an analytical artifact, has to be explained by vital effects. In particular the early mineralization zone appears to be subject to strong biological isotope fractionation, whereas later mineralization may be controlled by growth kinetics [*Rollion-Bard et al.*, 2010].

Similar to the geochemical heterogeneities observed in aragonitic scleractinians, the calcitic skeletons of the octocorals *Primnoa resedaeformis* [*Heikoop et al.*, 2002] and *Keratoisis ornata* [*López Correa et al.*, in prep.], as well as the aragonitic stylasterid *Errina dabneyi* [*Wisshak et al.*, 2009] also show vital effects. The juvenile skeletal portions in these groups are relatively enriched in trace elements (e.g. Li/Ca, B/Ca and Mg/Ca) and significantly depleted from the expected equilibrium values for $\delta^{18}O$ and $\delta^{13}C$. The magnitude of stable isotope disequilibrium is smaller than in scleractinians, with only ~1.2 ‰ in $\delta^{18}O$ and ~1.8‰ in $\delta^{13}C$ [e.g. *López Correa et al.*, in prep.; *Wisshak et al.*, 2009]. These vital effects are particularly strong in the first ~30 years of the octocorals' lifespan, as observed in *Keratoisis ornata* from Canada (López Correa et al., *in prep.*) and New Zealand [*Noé and Dullo*, 2006]. In contrast, adult skeletons are precipitated close to isotopic equilibrium and elemental compositions like Mg/Ca can be used as temperature proxies. Similar trace element and stable isotopic compositions across the solid axial calcareous skeleton of a live collected specimen of the precious gorgonian coral *Corallium rubrum* from the Mediterranean Sea reveal the internal portion (medullar region) to be affected by vital effects, whereas the external parts of this coral (annular region) show a more homogeneous geochemical pattern [*Montagna et al.*, 2008].

In summary, all available evidence suggests that vital effects play an important role in the geochemical records precipitated by cold-water scleractinians. Less is known about the skeletal composition of gorgonians and stylasterids, but in particular their outer skeletons may be more homogenous compared to scleractinians. Because of the heterogeneities observed in scleractinians, careful calibration and study of this valuable archive is however required if we are to successful reconstruct past pH conditions of seawater.

## Boron isotope systematics in marine carbonates

The systematics of the $\delta^{11}B$ paleo-*p*H proxy have been described in detail by *Hemming and Hanson* [1992]. Briefly, there are two dominant species of dissolved boron in seawater, boric acid [$B(OH)_3$] and borate [$B(OH)_4^-$], and their relative concentration changes with pH. At low pH (<7) virtually all boron is present in the form of boric acid, whereas at high pH (>10) boron exists predominately in the form of borate (Fig. 1a). The stable isotope $^{11}B$ is enriched in $B(OH)_3$ compared to $B(OH)_4^-$, with a constant isotopic fractionation between the two boron species [*Kakihana et al.*, 1977; *Klochko et al.*, 2006]. Consequently, as the relative concentration of the dissolved species changes with pH, so does their isotopic composition (Fig. 1b).

Diverse biogenic and inorganic marine carbonates all showed that modern carbonates record $\delta^{11}B$ similar to the $\delta^{11}B$ of $B(OH)_4^-$ at modern seawater pH [*Hemming and Hanson*, 1992]. Based on this empirical evidence these authors suggested that it is the charged borate species that is sorbed and incorporated into marine carbonates (Fig. 1b). This interpretation has subsequently been supported by a number of empirical calibration studies on planktic foraminifers [*Sanyal et al.*, 1996; *Sanyal et al.*, 2001], corals [*Hönisch et al.*, 2004; *Reynaud et al.*, 2004] and inorganic calcium carbonates [*Hemming et al.*, 1995; *Sanyal et al.*, 2000] over a wide range of seawater pH (Fig. 2). Although $\delta^{11}B$ of these biogenic calcium carbonates show offsets of up to 5‰, the offset from the aqueous reservoir is ~-20‰, indicating that the first order effect is the incorporation of $B(OH)_4^-$ in carbonate.

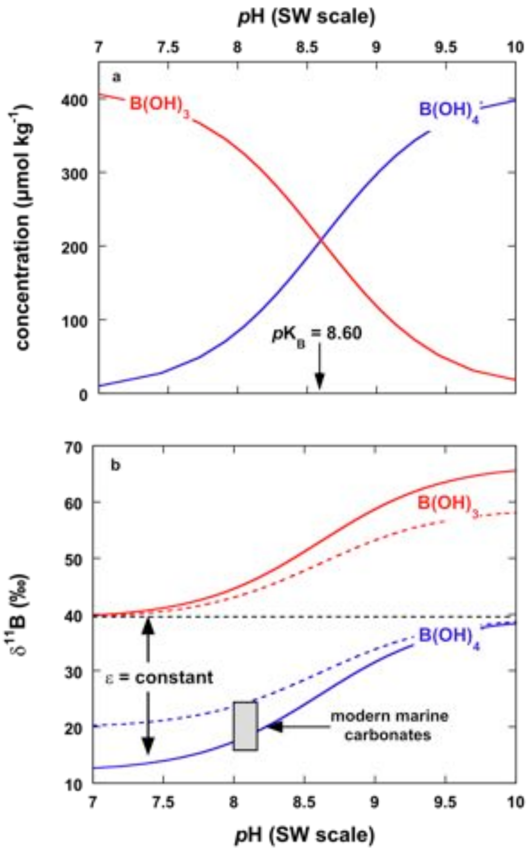

**Fig. 1a.** pH-dependent concentration and **b** isotopic composition of the two dominant boron species in seawater. The isotope fractionation between the two species is constant at ~27‰ (solid lines, Klochko et al. 2006). Also shown is the theoretical fractionation after Kakihana et al. (~20‰, broken lines, 1977), which matches the shape and inflection point of empirical carbonate calibrations (see also Fig. 3). $\delta^{11}B$ of modern marine carbonates (grey box) falls close to the borate curve and suggests the preferential incorporation of borate over boric acid (Hemming & Hanson 1992).

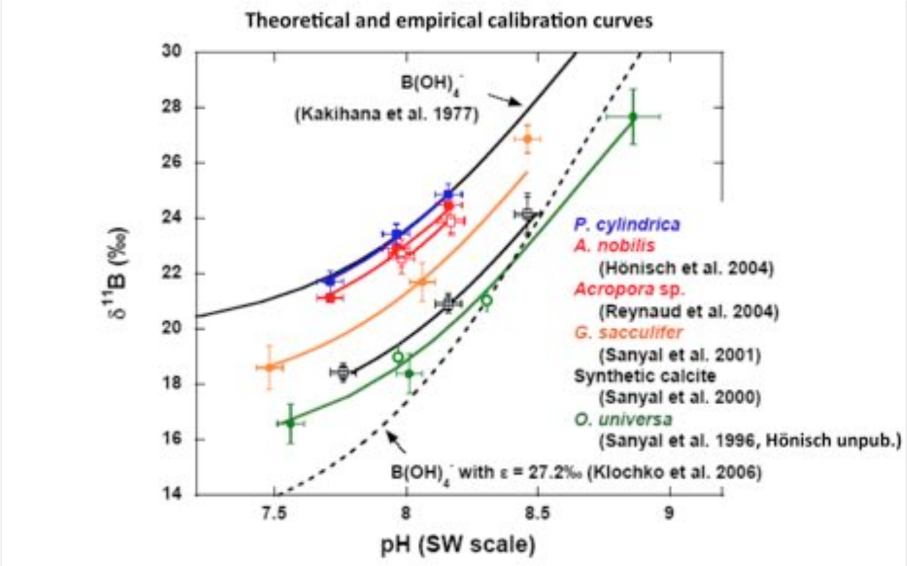

**Figure 2.** $\delta^{11}B$ of laboratory grown carbonates closely follows the theoretical borate curve of Kakihana et al. (1977, black solid line). Also shown is the experimental borate curve after Klochko et al. (2006, dashed line) and new culture data for *O. universa* (open symbols), which were measured by NTIMS (Triton) at LDEO and which confirm the earlier published calibration.

## Previous studies of boron isotopes in deep-sea corals

The only published study of boron isotopes in deep-sea corals used ion-microprobe measurements in *L. pertusa* and found a large range in $\delta^{11}B$ of >10‰ and very high isotope ratios of 28-39‰, i.e. similar to the isotopic composition of seawater (39.5‰). Blamart et al. [2007] suggested this large $\delta^{11}B$ range could not be explained by environmental pH variations and that without a better understanding of the responsible processes, application of the boron isotope proxy to reconstructing seawater pH with this species would be problematic. However, ion microprobe analyses of the shallow water coral *Porites* also show similarly large boron and oxygen isotope variations [~12 and 10‰, respectively, *Rollion-Bard et al.*, 2003], whereas NTIMS analyses of cultured *Porites cylindrica, Acropora nobilis*, and *Acropora* sp. show clear $\delta^{11}B$/pH relationships [*Hönisch et al.*, 2004; *Reynaud et al.*, 2004] that are consistent in slope and inflection point with empirical calibrations of other marine carbonates (Figure 2). Similarly, NTIMS and positive TIMS analyses of 200 to 300-year long *Porites* coral records show $\delta^{11}B$-variations <3‰ [*Pelejero et al.*, 2005; *Wei et al.*, 2009], and bulk oxygen isotope analyses of *Porites* records typically do not exceed 1‰ long-term variation [e.g. *Felis et al.*, 2004; *Linsley et al.*, 2008]. This comparison indicates that both the absolute values and micro-scale variations observed by ion-microprobe may not apply to bulk skeleton analyses. Whilst we will document within-species variability as part of this project through replicate analyses, we anticipate that the sample sizes required for NTIMS and MC-ICP-MS analyses will smooth out any microscale variations.

All published empirical calibration curves for the boron isotope/pH relationship in marine carbonates have been established by Negative Thermal Ionization Mass Spectrometry (NTIMS). They are all parallel to each other and to the theoretical curve of $\delta^{11}B_{borate}$ vs. seawater-pH [*Kakihana et al.*, 1977], but they show a shallower slope than suggested by the more recent empirically determined boron isotope fractionation in seawater [*Klochko et al.*, 2006] (Fig. 2). New developments in the field of MC-ICP-MS have shown promising results for boron isotope analysis [*Foster*, 2008]. Coretop measurements of planktic and benthic foraminifers by MC-ICP-MS suggest that they may follow the steeper boron isotope/pH relationship suggested by Klochko and co-workers' [2006] larger fractionation factor (Fig. 3a). Despite seeming to be at odds with one another there has not yet been a rigorous intercalibration of the NTIMS and MC-ICP-MS methods. Here we briefly summarize the apparent differences and similarities between the data collected from each method, and outline how we will resolve this issue during the early part of the proposed research. Figure 3b shows NTIMS analyses of planktic foraminifers relative to $\delta^{11}B_{borate}$ estimated from the larger fractionation factor. These data also show the same slope as $\delta^{11}B_{borate}$ after Klochko et al. [2006]. There are small differences between the absolute values measured by MC-ICP-MS and NTIMS but the overall patterns appear to be the same when only coretop analyses are considered (Fig. 3a+b). However, these coretop analyses only cover a small range of seawater-pH in the modern ocean (~0.1 pH units for planktic foraminifer species, and ~0.2 pH units for benthic foraminifers). Previous calibration studies have taken advantage of laboratory culture

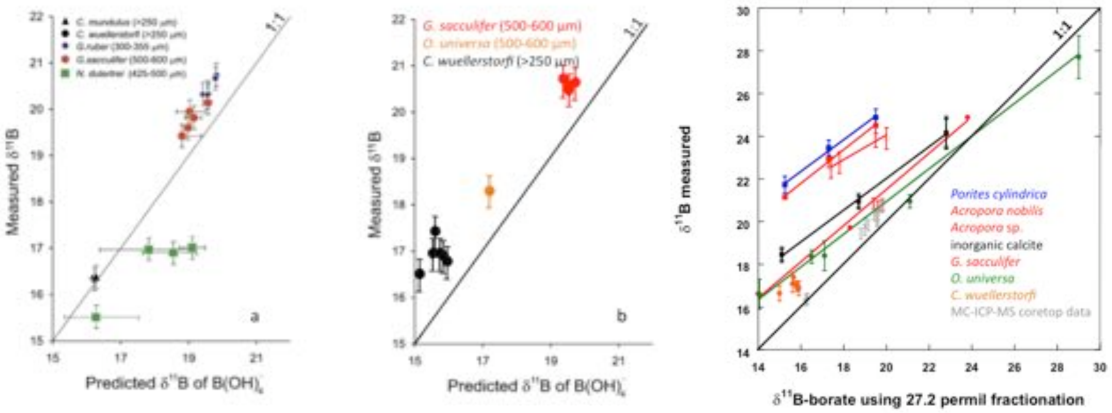

**Figure 3 a+b**. Boron isotope analyses of planktic and benthic foraminifers from coretop samples relative to the isotopic composition of borate predicted by an aqueous boron isotope fractionation of 27.2‰ between boric acid and borate after Klochko et al. [2006]. The 1:1 line depicts the expected isotopic composition of marine carbonates if only borate is incorporated without further fractionation from the seawater isotopic composition. Data in **a** were measured by MC-ICP-MS by Foster [2008], data in **b** are measured by NTIMS by Hönisch et al. [2009]. Although there are absolute differences between data collected for the same carbonate species, over the wide pH range covered by these planktic and benthic foraminifer samples it appears that both methods yield data that are consistent with the boron isotope fractionation after Klochko et al. [2006]. In contrast, **c** shows the empirical calibrations from Fig. 2 relative to the $\delta^{11}B_{borate}$ after Klochko et al. [2006]. This comparison demonstrates that the difference in slopes and offsets between species only become apparent when the same marine carbonate species is grown over a wide pH range. No such empirical calibrations exist from MC-ICP-MS measurements. Note that the isotopic composition of borate predicted from a 27.2‰ fractionation does not take into account a temperature effect on the aqueous boron isotope fractionation.

experiments, where a much larger pH-range (>1 pH units) can be established under otherwise constant conditions (Fig. 2). Figure 3c shows all available empirical calibrations relative to $\delta^{11}B_{borate}$ after Klochko et al. [2006]. This figure demonstrates that it is only over a wide pH range where the difference in slopes between $\delta^{11}B$ in marine carbonates and $\delta^{11}B_{borate}$ becomes apparent. As yet there are no equivalent culture data analyzed by MC-ICP-MS. To solve this issue an international interlaboratory comparison effort is underway to compare the same samples measured by different techniques (Foster, Hönisch, Gaillardet, Dwyer, Vengosh, Eggins). So far the group has analyzed boric acids and single samples of various marine carbonates. No differences were observed for the boric acids over the $\delta^{11}B$ range of 50‰. There do appear to be differences between the marine carbonates, which may be caused by matrix effects. Continued efforts of comparing the same carbonate species grown over a wide pH range are underway and will be completed by the same group of analysts. When these intercalibration efforts are complete it will be possible for the community to decide (a) whether one analysis method is preferable or (b) how to make sure the different methods yield the same pH-estimate from the same samples. Despite the existing apparent discrepancies the value of a paleo-pH proxy makes this effort worthwhile. In the context of this proposed study our analyses will be performed using both techniques, by NTIMS at LDEO and by MC-ICP-MS at Bristol University and subsequently at LDEO. The pH range covered by our modern and cultured sample (pH 7.6-8.1) set is much larger than available for foraminifers from coretop sediments, and thus will allow us to establish calibration curves over a wide pH-range, and to directly compare NTIMS and MC-ICP-MS results. The large sample sizes available will also allow subsamples to be sent to other laboratories at their request to continue intercalibration efforts into the future.

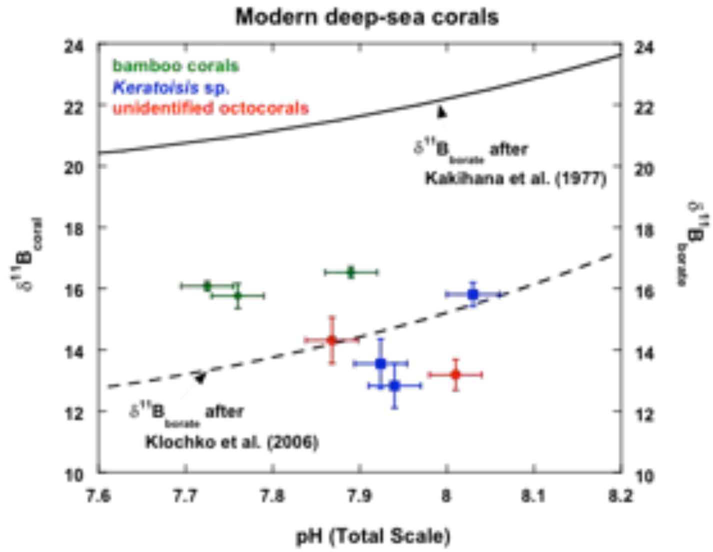

**Figure 4.** Preliminary boron isotope analyses of various octocoral species suggest that their isotopic composition is similar to that of benthic foraminifers [*Foster*, 2008; *Hönisch et al.*, 2008] and falls close to the boron isotopic composition of dissolved borate in seawater. Some of the variability in this data set is likely due to species-specific vital effects but the scatter in the *Keratoisis* data is most likely due to sampling off the skeletal surface rather than a selective microsampling along the major growth axis of the coral skeletons.

Preliminary analyses of deep-sea octocorals have been performed by graduate student Sarah Hale and Marie-Tharp fellow Laura Robinson on the NTIMS at LDEO and show low $\delta^{11}B$ data similar to benthic foraminifers (Figure 4). As yet we have only analyzed one species over a pH range, the other analyses were from different species. *Keratoisis* sp., a cosmopolitan species, shows a ~3‰ increase in $\delta^{11}B$ over a ~ 0.1 pH change.

During these preliminary analyses we observed variability greater than analytical precision in replicate analyses. These preliminary data have been collected on material that was scraped off the outer surface of these coral skeletons and we suspect this variability would be reduced with an improved sampling strategy. As part of the propsed study we will establish the best sampling strategy for boron isotopes in deep-sea corals. This will include sampling with a microdrill along the growth lines of the calcitic internodes and sampling of transects from the central axis to the outer surface (Fig. 5).

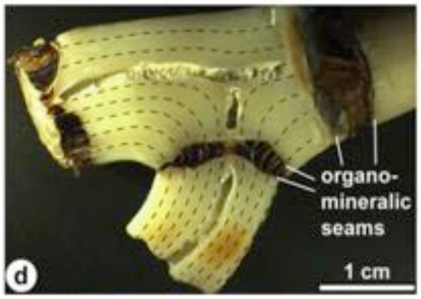

**Fig. 5.** Longitudinal section of an octocoral skeletal fragment. Dotted lines show growth increments of the calcitic internodes, dark bands are the organic nodes typical for bamboo corals [*Noé and Dullo*, 2006]. Sampling will track the dotted lines and cross them from the central axis to the outer surface.

## SAMPLES

### Boron isotope/paleo-pH calibration

The main focus of this study is to calibrate and assess the reliability of deep-sea corals in recording paleo-pH through the boron isotopic composition of their skeletons. We have assembled a range of modern corals for this study from recent oceanographic research cruises in the North Atlantic, Southern Ocean, and the Mediterranean. The samples come from depths of 20-3460 m, covering a pH-range of 7.7–8.1, a temperature-range of -0.9 to 13.8ºC, and salinity 32.9-38.8 (Fig. 6 and Table 1 in the supplementary material). The octocorals *Keratoisis sp.* and *Corallium sp.* dominate the sample set and in particular a comprehensive set of recent and fossil *Keratoisis* sp. from the New England Seamounts that cover depths of 1334-2584 m. The calibration will focus on all the species within these genus and the locations available to us. One of the goals of this study is to identify specific offsets between different species of the same genus. Identifying fossil specimens to the species level is challenging and species-specific offsets need to be identified to gauge the feasibility of undertaking paleoreconstructions with unidentified species.

In addition to specimens collected from their natural environment, we also have available cultured specimens of the scleractinian *Lophelia pertusa*. Specimens were grown by Armin Form for 10 months in the laboratory at IfM GEOMAR in Kiel, Germany. Four pH-treatments were accomplished (pH 7.7, 7.8, 7.9 and 8.0) under controlled conditions, and the experimental onset was marked by alizarin staining. *Lophelia pertusa* secretes rather small corallites, providing little material for geochemical analyses. However, the B concentration in *L. pertusa* skeletons measured by Blamart et al. [2007] and López Correa et al. [2010] ranges from 53-122 ppm. With such high B concentrations, sample material required for boron isotope analyses by NTIMS and MC-ICP-MS is smaller than 0.5 mg and can easily be collected by microdrilling.

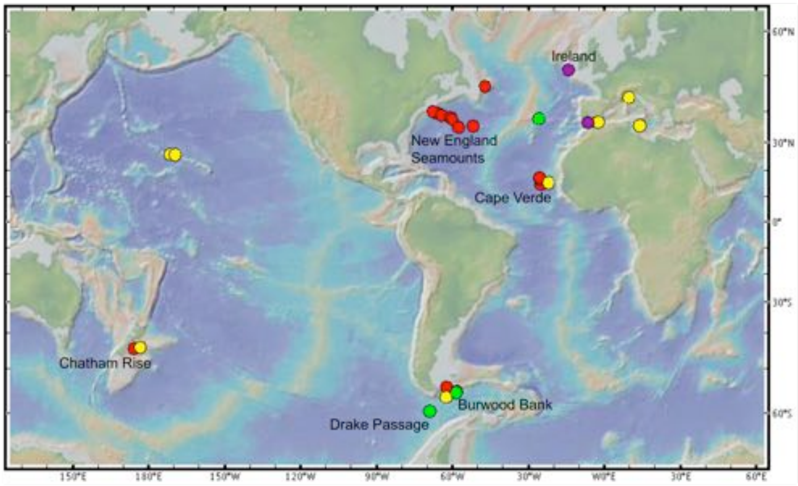

**Fig. 6:** Map showing the modern and fossil deep sea coral samples available to this study. Colors indicate coral genus: red – *Keratoisis*, yellow – *Corallium*, green – stylasterids, purple - *Lophelia pertusa*.

### Paleoreconstructions: 1. Anthropogenic ocean acidification

Our first goal is to reconstruct pH change at intermediate depths, where significant anthropogenic $CO_2$ invasion has been documented in the North Atlantic and the Southern Ocean [*Feely et al.*, 2004; *Sabine et al.*, 2004]. Although few samples are available from suitable depths in the North Atlantic, we have identified sufficient samples to provide the first data of this kind. In addition, future cruises are planned to fill this gap. For example, Robinson & Waller have a pending return to the Southern Ocean, which is focused towards collecting coral and water samples in a transect across the Drake Passage. This cruise would provide GRA Sarah Hale with the opportunity to go to sea, participate in sample collection and measure carbonate chemistry directly at the collection sites. Even without these potential samples we have already identified samples to meet the goals of this project. Through collaborations and loans from museums we have access to a number of samples that should have recorded an anthropogenic $CO_2$ invasion signal, and "control" samples that should not. A unique set of *Keratoisis ornata* samples is available from the Smithsonian National Museum of Natural History, collected alive from the Nova Scotia Banquereau Bank, Georges Bank and Nantucket Shoals in the Western North Atlantic in years 1878-1884 at 366-2789 m water depth. Thick branches of 15-40 cm length are available from these specimens, allowing for detailed microsampling. A recent modern companion sample is available from the Canadian Southern Grand Banks at 743 m water depth. The collection sites lie in the heart of the strongest region of anthropogenic $CO_2$ invasion. We will employ the 1994 GLODAP carbonate chemistry data set with anthropogenic $CO_2$ estimates after Sabine et al. [2004] (Fig. 7) to estimate pre-anthropogenic dissolved inorganic carbon (DIC) concentrations at the collection sites. Pairing these DIC with modern alkalinity estimates ($CO_2$ invasion does not affect seawater alkalinity) then allows for estimation of past pH conditions at the sampling location. With 30 $\mu mol\ kg^{-1}$ anthropogenic $CO_2$, seawater-pH at 700 m water depth in the Northwest Atlantic should have dropped by ~0.06 units, a change large enough to be resolved by boron isotopes (2s analytical uncertainty is ±0.03 pH units). In fact, the GLODAP data are from 1994, so the actual anthropogenic $CO_2$ signal at these sites should now be even larger than estimated by Sabine et al. in [2004] and thus resolvable with the modern coral specimens already in hand. Ocean going efforts simply provide snapshots of pH in the ocean at discrete data points. Our high temporal resolution record of pH from B isotopes will be the first continuous time series of pH in the North Atlantic that will document both the timing and amplitude of pH change in this sensitive location.

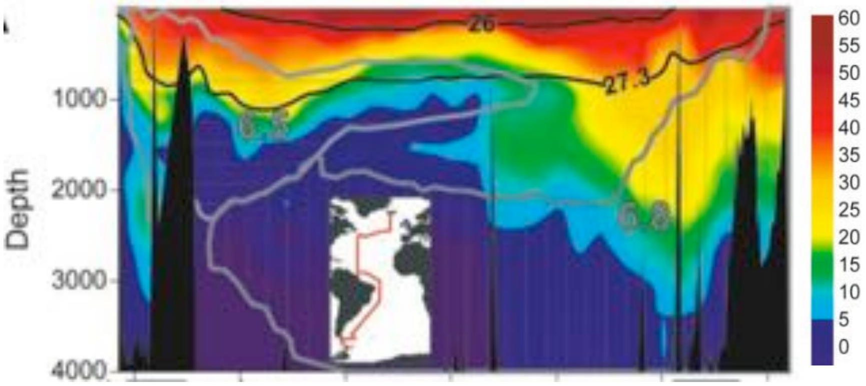

**Fig. 7.** Anthropogenic $CO_2$ ($\mu$mol $kg^{-1}$) in the Atlantic Ocean from *Sabine at al.* [2004]. Note the deeper penetration in the North Atlantic and Southern Ocean, where many of our samples were collected.

*Lophelia pertusa* specimens from 600-800 m water depth off the coast of Ireland will be provided by Dierk Hebbeln (see attached letter of collaboration). Subrecent samples are already available at Bremen University and modern specimens will be collected by Hebbeln on a cruise planned for June/July 2010. Estimates for the anthropogenic acidification at this location yield -0.065 pH units.

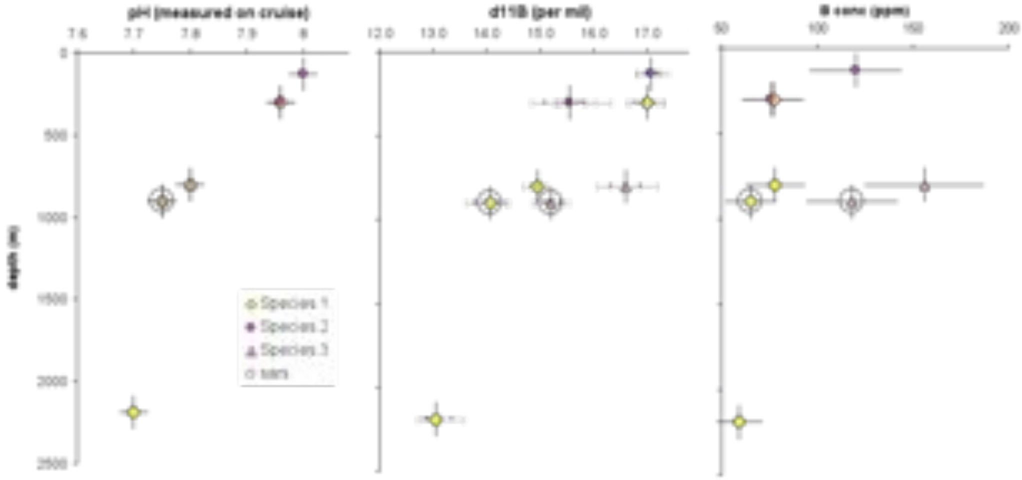

**Fig. 8.** Depth transect from the Drake Passage, showing **a** pH measured at sea, **b** boron isotopic composition and **c** boron concentration measured in various recent stylasterids. Note that the species of these stylasterids have not yet been identified and specimens have only been classified according to the color of their skeleton. The boron isotopic composition and boron concentration increase with seawater pH, consistent with these geochemical signals being controlled by seawater carbonate chemistry. Radiocarbon dating of the shallow pink specimen has shown bomb radiocarbon throughout the skeleton, suggesting an age younger than 50 years. The large size and banding pattern of this specimen make it ideal for high-resolution analysis of anthropogenic $CO_2$ invasion at this shallow site.

Abundant stylasterid corals were retrieved from the Drake Passage and Burdwood Bank (the shelf off southern Argentina). These stylasterids appear to thrive where other corals are not present. During her stay at LDEO as a Marie Tharp fellow Laura Robinson analyzed a depth transect of these specimens (Fig. 8). The stylasterid species have not yet been identified and a distinction was made according to the color of their skeleton and polyp morphology. However, it is obvious that the boron isotopic composition and boron concentration of these specimens increase at higher pH (shallower water depths), suggesting that pH is the controlling parameter. pH was analyzed from water collected on a CTD cast during NBP-0805 cruise to the region. One specimen has a diameter of ~3cm, with clear pink and white banding, and was collected from 130 m on the Burdwood Bank. Radiocarbon dating revealed that the entire skeleton contains bomb radiocarbon, suggesting it is younger than ~50 years in age. We propose to subsample across the bands using a micro-mill to generate the first high-resolution annually resolved pH record for this region.

**In summary, time series of boron isotopes and B/Ca will be established from subrecent and recent specimens from these locations. The subrecent samples will serve as a reference and to evaluate potential seasonal or annual variations. The difference between the subrecent and modern samples will then allow interpretation for anthropogenic $CO_2$ invasion in the North Atlantic. Dating of *L. pertusa* from off Ireland will further allow for interpretation of abundance patterns through time, noting this scleractinian is one of the most vulnerable to ocean acidification.**

Recent analysis of $CO_2$ invasion to the western North Atlantic [Fig. 7, *Sabine et al.*, 2004] suggests that the maximum depth of penetration is about 1,500 m. If this analysis is correct, our corals from the New England Seamounts in the Western North Atlantic (>1,300 m water depth) should show no or only very little and recent change over time if the $CO_2$ has in fact reached these sites. Our samples from Cape Verde in the Eastern equatorial Atlantic should not have experienced any pH drop, as they are not located within the flow path of North Atlantic Deep Water. Fossil specimens of these collections will be used as a reference for non-ocean acidification locations.

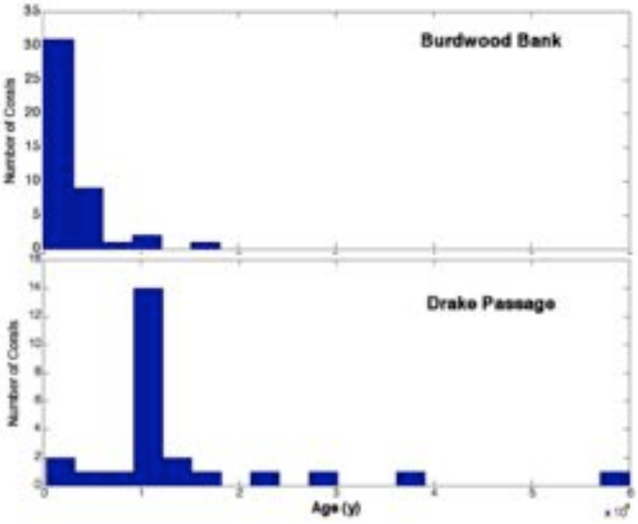

**Fig. 9**. Age histograms of *D. dianthus* corals from the Burdwood Bank and Seamounts in the Drake Passage (note scale in $10^4$ years, so each x axis goes to 60,000 years ago). Most fossil corals from Burdwood lived in the last 1000 years. By contrast peak abundance in the Drake Passage was about 10,000 years ago.

### Paleoreconstructions: 2. Last glacial and deglaciation in the Southern Ocean

Some deep-sea corals, such as *Lophelia pertusa*, are colonial and form carbonate mounds [*Freiwald*, 2003]. Cores drilled into these mounds can be dated at selected depths and the age can be interpolated between these depths to produce a preliminary chronology [e.g., *Frank et al.*, 2004; *Kano et al.*, 2007]. Deep-sea corals collected from the New England Seamounts in the Western North Atlantic reveal that most of the corals grew during glacial conditions at times of abrupt climate change, indicating a potential connection between ocean circulation changes and coral growth [*Robinson et al.*, 2007]. In a similar fashion, the ages of deep-sea corals collected from the seafloor in the Northeast Atlantic appear to exhibit a latitudinal dependence. Corals from north of 50°N are all of Holocene age, whereas corals from lower latitudes (from seamounts off Northwest Africa and from the Mediterranean) have a broader age distribution, with apparently continuous coral growth for the past 53.5 thousand years (ky) [*Schröder-Ritzrau et al.*, 2005]. Recent analyses of corals from the Drake Passage show that the deep-sea coral diversity and populations in the central Drake Passage have clearly changed over time [*Burke et al.*, submitted]. The *Desmophyllum dianthus* fossils from Burdwood Bank are biased towards modern ages, with more than 70% of the fossils collected having calendar ages between 0 and 2,500 years (Fig. 9). In contrast to Burdwood Bank, more than half of the fossils from the Drake Passage have late deglacial/early Holocene calendar ages of between 10 and 12.5 ka (Fig. 9). Changes in environmental parameters may provide a control on the distributions of these communities, perhaps through availability of detrital food or the seawater saturation state. In particular the large abundance peak in the Drake Passage between 10 and 12.5 ka indicates that conditions may only have been favorable for a short period. Geochemical analysis of such corals provides an opportunity to gain insight into the environmental controls of deep coral reef growth.

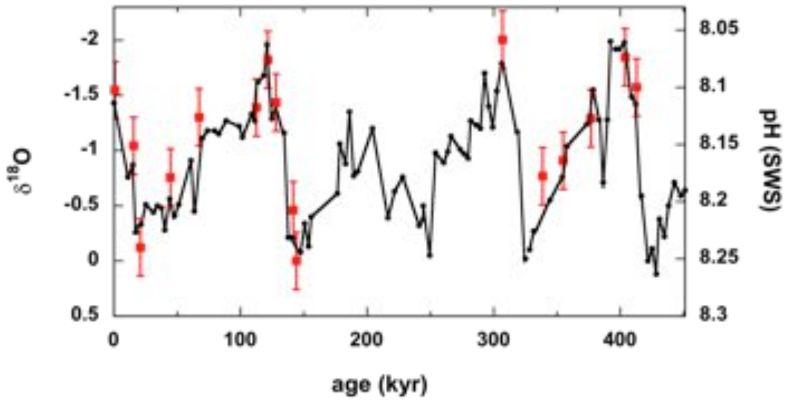

**Fig. 10.** Planktic oxygen isotope record (black symbols) and surface ocean pH variation reconstructed from boron isotopes in planktic foraminifers [red squares, after *Hönisch and Hemming*, 2005]. Glacial pH is systematically higher than interglacial pH by ~0.15 units.

The age distribution of *Desmophyllum dianthus* specimens demonstrates that corals have lived in these two locations for tens of thousands of years. Given the large chemical heterogeneity observed in cold-water scleractinians we prefer to focus our approach on gorgonians, which occur in plentiful abundance in the same site samples. We will generate an age survey on gorgonians similar to the one already established on *D. dianthus*, and then select material for isotope and B/Ca analysis. Specific focus will be placed on material from the last glacial and the deglaciation. Boron isotope measurements of planktic and benthic foraminifers have demonstrated that surface seawater pH during glacial times was 0.15 units higher [Fig. 10, *Foster*, 2008; *Hönisch and Hemming*, 2005; *Sanyal et al.*, 1995], respectively bottom water pH was ~0.1 pH units higher [*Hönisch et al.*, 2008] than during interglacial times. In comparison to modern deep-sea coral specimens, the largest pH changes therefore should have been experienced by their glacial and deglacial relatives. The proposed reconstructions in the Drake Passage and on Burdwood Bank will then allow for interpretation of the variations in scleractinian abundance in the light of changing seawater carbonate chemistry.

A final important sample set exists from Chatham Rise off New Zealand, where modern and fossil (3.9 ka) *Keratoisis* specimens have been retrieved from 594-740 m water depth [*Noe et al.*, 2008]. These corals are radiocarbon dated and high-resolution (micro-milled) stable isotope profiles ($\delta^{13}C/\delta^{18}O$) and trace element (LA-ICP-MS) profiles are already available (Noé, López Correa and Montagna, unpublished data) and provide a base to study $\delta^{11}B$ variations in parallel micromill profiles. The recent and mid-Holocene *Keratoisis* specimens provide centennial scale longevity and serve as an ideal target to test potential pH changes associated with natural Holocene variability and associated to the recent anthropogenic $CO_2$ invasion at these relatively shallow sites.

### GEOCHEMICAL ANALYSES

The analyses of all these specimens will include boron isotopes and B/Ca ratios. Little is known about the geochemical composition of deep-sea corals and this comprehensive sample set aims to gain better insight into the controlling parameters. Boron isotopes are the best-understood and most direct pH-proxy in marine carbonates (Fig. 2, 10 and references therein) and our pilot data (Fig. 4 and Fig. 8) show great promise that it will also work in deep-sea corals.

Trace metals in corals, and in particular in deep-sea corals, are poorly understood and we do not aim to study them in great detail. However, as shown in Fig. 8, the boron concentration of the stylasterids from the Drake Passage show significantly higher values in samples with a higher boron isotopic composition. This pattern is consistent with the idea that borate is the dominant boron species incorporated into marine carbonates [*Hemming and Hanson*, 1992] and because the dissolved borate concentration in seawater increases with seawater pH, more B is expected to be incorporated at higher pH. This hypothesis has been studied in planktic and benthic foraminifers [*Foster*, 2008; *Yu et al.*, 2007] and although discussions about the actual controlling parameters (pH, carbonate ion, temperature, carbonate saturation, growth rate) remain unresolved, the variation in boron concentration measured in deep-sea corals (Fig. 8) appears systematic and much larger than observed in foraminifers. We therefore propose to measure B/Ca ratios by solution ICP-MS at Rutgers University. Measuring metal/Ca ratios will yield more accurate results than the isotope dilution technique applied for the B concentration measurements on the stylasterids (Fig. 9). Other elements (Mg, Sr, U, Ba, Li, Cd, Mn and Fe) will be collected as well but are not the focus of this study.

The corals will be sampled using a microdrill. The powders will then be homogenized and split into subsamples for the various analyses. NTIMS and MC-ICP-MS techniques will be compared for boron isotope analyses, following the procedures outlined in Hönisch et al. [2007] and Foster [2008], respectively. Boron isotope analyses will be performed on the Triton TIMS at LDEO and on the Neptune MC-ICP-MS at Bristol University and subsequently at LDEO.

#### Dating

One difficulty with using fossil corals is that using current techniques it is impractical to date large numbers of corals to determine which specimens have the appropriate ages for radiocarbon reconstructions. Through ongoing collaboration with NOSAMS staff, continued development of reconnaissance dating will be undertaken and applied to age screening of our sample set [*Burke et al.*, submitted]. Approximately 10 mg of carbonate is burned in an Elemental Analyzer before combustion to graphite in sealed tubes based on a method by [*Xu et al.*, 2007]. Replicate samples from a single coral yielded a standard deviation of 81 years (n=9), with no correlation to sample mass or cleaning protocol, and a 1:1 correlation with analyses made following standard procedures. These percent level analyses provide an age screening method for determination of corals suited to the reconstructions to be made in this proposal. The largest uncertainty comes from assumptions of the reservoir age when converting to calendar age. For precise radiocarbon analyses, each coral sample will be thoroughly cleaned and, immediately prior to phosphoric acid dissolution, leached in HCl to remove ~50% of its total mass [*Adkins et al.*, 2002]. Select samples for paleoreconstructions will be analyzed multiple times to determine the growth period of the specimens and to identify the significance of their growth bands. All samples will be analyzed at NOSAMS.

The high U-content in aragonitic scleractinian species allows accurate dating back to ~500ka with ~1% error by ICP-MS [*Cheng et al.*, 2000; *Lomitschka and Mangini*, 1999; *Robinson et al.*, 2006]. To measure the age, the ingrowth of $^{230}$Th from uranium will be used as a chronometer. A 1.0 g sample of coral carbonate will be physically and chemically cleaned to remove contaminating phases [*Cheng et al.*, 2000; *Shen and Boyle*, 1988]. Dissolved samples will be spiked with a mixed $^{229}$Th - $^{236}$U spike and U, and Th will be separated and purified by anion-exchange chemistry [*Edwards et al.*, 1986]. Purified aliquots of U and Th will be measured by Neptune MC-ICP-MS at WHOI or Bristol [*Robinson et al.*, 2002; *Robinson et al.*, 2005]. The $^{232}$Th concentration in each sample will be monitored to assess the contribution of $^{230}$Th that is not a product of ingrowth. The potential effects of diagenesis will be monitored using the $^{234}$U/$^{238}$U ratio, and all analyses lying more than 6‰ from the closed system evolution will be rejected.

The dating part of the study will be guided by Laura Robinson at WHOI and Bristol University.

## DATA MANAGEMENT PLAN

The data generated from this proposed research will be published in peer-reviewed journals and the NOAA paleo data base.

## BROADER IMPACTS

Finding proxies to measure past ocean acidification events is a fundamental component to better understand the impact of ocean acidification on calcifying ecosystems. By comparison of multiple analytical techniques this study will also advance the discussion whether there is a fundamental difference between boron isotope measurements by NTIMS and MC-ICP-MS. This is important to improve our confidence in past and future reconstructions using this valuable proxy.

The proposed work will be supervised by Bärbel Hönisch, who recently started her tenure track career as an Assistant Professor at the Lamont-Doherty Earth Observatory of Columbia University (LDEO). Employment of a lab assistant will not only facilitate the proposed boron isotope analyses, but also improve support for the growing number of visiting scientists (e.g. Gretta Bartoli, Gianluca Marino, Ryan Moyer) in the LDEO TIMS facility. LDEO has a strong commitment to improving diversity in the geosciences. Hönisch is a young female scientist, and the junior scientists working in the LDEO labs include at least 50% females. LDEO also annually participates in several summer internship programs, including the NSF Ocean Science Sponsored Summer Intern Program, and the Federal Work Study Program. These programs are designed to encourage outstanding undergraduate students to participate in a research program in order to gain insight into the daily work of an Earth scientist. Over the past 3 years Hönisch has supervised 3 undergraduate students funded by these programs, Steven Wang, Lucy Chen and Ashley Ellenson, who were trained in sediment studies, picking foraminifers, drilling coral samples and operating the mass spectrometer for boron isotope analyses. The proposed project has clear societal relevance and is perfect for a team of eager young scientists who are still finding their way but are keen to be involved.

Hönisch is active in organizing workshops and conference sessions on ocean acidification, including the NSF sponsored workshop on paleocean acidification to be held on Catalina Island in August 2010. She also gives talks in public schools, presents her research at LDEO open house and embraces opportunities to talk to the general public, e.g. at a public screening of "A Sea Change" in New York. Hönisch plans to continue these efforts, which will facilitate communication with the public and discussion with scientific peers.

## RESULTS FROM PRIOR NSF SUPPORT:

**OCE 06-23621** to B. Hönisch, N.G. Hemming, W.S. Broecker, "Reconstructing the role of $CO_2$ in climate change throughout the Cenozoic, Step 1: The mid-Pleistocene transition", amount: $196,581.-, period covered: 9/1/06-8/31/10.

A 2.1 Myr record of atmospheric $pCO_2$ has been estimated from boron isotopes in planktic foraminifer shells, showing relatively stable $pCO_2$ over the past 2.1 Myr, only glacial $pCO_2$ before 800 kyr was slightly elevated compared to the late Pleistocene [*Hönisch et al.*, 2009]. In 2008 the scope of this study was expanded to study boron isotopes in Cenozoic benthic foraminifers from the Pacific and Atlantic Ocean, to extract the presumably dominant contribution of the $\delta^{11}B_{seawater}$ signal from these shells. Postdoc Markus Raitzsch has analyzed three benthic foraminifer species from the Atlantic and Pacific ocean with a 3 million year resolution. The data are overall similar to modeling estimates by Lemarchand et al. [2000] but also show a pronounced step near the onset of Antarctic glaciation.

**OCE 07-51764** to B. Hönisch: "Validation of the B/Ca proxy for surface seawater pH and application to measure anthropogenic ocean acidification", amount: $337,724.-, period covered: 3/15/08-3/14/11.

Culture experiments with living planktic foraminifers have been performed in 2008 and 2010 to study the effect of carbonate chemistry, temperature, salinity, and boron concentration on the B/Ca ratio of *Orbulina universa*, *Globigerina bulloides*, *Globigerinoides sacculifer* and *G. ruber*. Graduate student Katherine Allen has analyzed shells from the 2008 cultures and is in the process of writing a manuscript for publication. She already presented trace element and calcification data at the AGU Fall Meetings 2008 and 2009. In addition, a 300-year sample set of planktic foraminifer shells from Pygmy Basin core PBBC-1 for B/Ca and boron isotope analyses. First boron isotope measurements have been performed and further analyses will follow when the study of cultured shells is completed.

**OCE 09-03014** to B. Hönisch: "Collaborative Research: Reconstructing deep sea acidification during the Paleocene-Eocene Thermal Maximum", amount: $68,185.-, period covered: 7/1/09-6/30/2011.

Up to three benthic foraminifer species have been measured for the period 55.3-54.7 Ma. The oldest and youngest samples, i.e. right before the carbon isotope excursion and after the recovery, give the highest isotopic composition. During the recovery phase the boron isotopic composition is as much as 2.5‰ lower, indicating significant bottom water acidification at that time. First data will be presented at ICP X.