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**Trace metal/Ca in
benthic foraminifera**

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Trace metal/Ca ratios in benthic foraminifera: the potential to reconstruct past variations in temperature and hypoxia in shelf regions

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Abstract

Shelf and coastal regions are exceptionally important for many countries as they provide the main habitat for many economically important fish and shellfish species. With ongoing global warming and human-induced eutrophication the shelf regions are especially affected, resulting in increased temperatures and stratification as well as oxygen depletion of the bottom waters. In order to be able to predict the magnitude of these changes in the future it is necessary to study how they varied in the past. Commonly used foraminiferal climate and environmental proxies, e.g. stable isotopes and trace metal/Ca ratios, which are applied in open-ocean settings are not necessarily applicable in shelf regions, either as faunas are completely different or as conditions change a-typical compared to the open-ocean. In this study we explore the use of Mg/Ca as paleothermometer and Mn/Ca as a potential proxy for changing dissolved oxygen conditions on the benthic foraminifera *Bulimina marginata* and *Globobulimina turgida*. Living specimens were collected from the Skagerrak and the Gullmar Fjord (SW-Sweden); the latter is hypoxic for several months a year. As the specimens were alive when collected it can be excluded that any diagenetic coatings have affected the trace metal/Ca ratios. The Mg/Ca ratios are similar to previously published values from the literature but display much larger variation than would be expected from the annual temperature change of less than 2 °C. An additional impact of the difference in the carbonate ion saturation state between the Skagerrak and the Gullmar Fjord could explain the results. Mn/Ca ratios on *Globobulimina turgida* potentially record variations in dissolved oxygen of the habitat where the foraminifera calcify. Samples from the Skagerrak display increased Mn/Ca in specimens which lived deeper in the sediment than those that lived near the surface. *Globobulimina turgida* samples from the lower oxygen Gullmar Fjord showed significantly increased Mn/Ca, being highest when dissolved oxygen was at a minimum. Our study suggests that trace metal/Ca ratios in benthic foraminifera from shelf regions have the potential to record past variations in bottom water temperature and

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dissolved oxygen concentrations but care needs to be taken to constrain the carbonate system.

1 Introduction

Traditional, open-ocean proxies, which for example are based on the calcite shells of marine organisms like foraminifera, are applied to a lesser extent in shelf and fjord environments as changes in the water mass conditions vary on a much larger scale than in the open ocean. The larger seasonal variability as well as the higher impacts from terrestrial and anthropogenic factors make it sometimes difficult to disentangle local and regional patterns, which hampers the possibility of obtaining a large scale pattern.

Shelf and fjord environments, however, are particularly affected by ongoing global warming. Shelf environments are economically important, being for instance important habitats for many fish and shellfish species. With ongoing global warming, changes in circulation, and as a consequence of eutrophication, dissolved oxygen concentrations are predicted to decrease in many areas (Sarmiento et al., 1998; Keeling and Garcia, 2002; Meier et al., 2011). Hypoxic conditions ($< 2 \text{ mL}^{-1}$ dissolved oxygen) are already widespread in the world's ocean often occurring naturally under high productivity areas like for example in the Arabian Sea (Reichart et al., 1998; Helly and Levin, 2004). But increasingly (seasonal) hypoxic conditions are caused or strengthened by anthropogenic induced eutrophication, especially in semi-enclosed basins like, for example, the Gulf of Mexico (Osterman et al., 2005; Platon et al., 2005), the Baltic Sea (Diaz and Rosenberg, 2008; Conley et al., 2011), and (Scandinavian) fjords (Gustafsson and Nordberg, 2000; Filipsson and Nordberg, 2004a). Ocean general circulation models predict that with ongoing global warming dissolved oxygen concentrations are going down as less oxygen can be dissolved into warmer water and stratification increases (Keeling and Garcia, 2002; and references therein). A matter of debate is still if these recent decreases in dissolved oxygen, especially in the Baltic Sea area, are

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purely anthropogenic or only intensifying natural variations (Zillén and Conley, 2010). Zillén and Conley (2010) showed that especially during the last two millennia phases of hypoxia like during the early-Medieval expansion (700–1300 AD), seem to correlate with major changes in population and land use in Scandinavia instead of correlating with natural climate variations. Some of the fjords of southwest Sweden, e.g. the Gullmar Fjord off the Skagerrak, have been studied extensively, especially with respect to changes in hydrography, eutrophication, and oxygen status (Gustafsson and Nordberg, 1999, 2000, 2001; Nordberg et al., 2001; Eriksson et al., 2002; Filipsson and Nordberg 2004a, b; 2010; Filipsson et al., 2004; Erlandsson et al., 2006; Polovodova et al., 2011). Due to their relatively small area fjords provide ideal settings for monitoring changes in water mass conditions. The Gullmar Fjord has been subject to regular environmental monitoring since 1958. The Swedish Meteorological and Hydrological Institute's (SMHI) SHARK hydrographic data base provides a time-series to perform further investigations on. Based on sediment cores covering the last century, using benthic foraminiferal assemblages, Filipsson and Nordberg (2004a) indicated that the faunal switches which have taken place between a common Skagerrak-Kattegat fauna and a more low-oxygen adapted fauna can be correlated to switches in North Atlantic Oscillation (NAO). During positive NAO periods, like the beginning and the end of the 20th century, strong westerly winds prevented the stagnant water from the Gullmar Fjord to be refreshed leading to more hypoxic conditions (Nordberg et al., 2000; Filipsson and Nordberg, 2004a).

Most deep-sea benthic foraminiferal species do not occur in proximal and relatively shallow and more brackish settings like the Skagerrak or the Gullmar Fjord. Similarly, planktonic foraminifera become increasingly less abundant closer to the coast. Lower salinities as well as potentially lower oxygen concentrations, and larger seasonal variability in temperature and salinity result in a different foraminiferal fauna compared to the open ocean. As such, commonly used proxies like foraminiferal Mg/Ca to reconstruct paleo-temperatures or potential new proxies like foraminiferal Mn/Ca to reconstruct paleo-oxygen conditions need to be applied on selected species which are

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common in shallow settings and/or more tolerant to low oxygen, i.e. hypoxic, conditions. Additionally, the mechanisms of these proxies need to be verified ensuring they are working in these proximal settings similar as they do in open-ocean settings.

Bulimina marginata is a common, shallow infaunal species of benthic foraminifera occurring in a wide range of settings from the deep sea, to open-shelf areas like the Skagerrak and stratified fjords like the Gullmar Fjord (Condradsen et al., 1994; Gustafsson and Nordberg, 2000). Additionally, it is tolerant to a wide range of microhabitats, from fully oxygenated down to hypoxic or even anoxic (Ohga and Kitazato, 1997; Fontanier et al., 2002) and it is relatively easy to culture (Hintz et al., 2006a, b; Barras et al., 2010; Filipsson et al., 2010). This makes *B. marginata* a very promising candidate to be used more often in paleoceanographic reconstructions.

Another common benthic foraminiferal species tolerant of low-oxygen contents is *Globobulimina turgida* (Jorissen et al., 1995; Kitazato et al., 2000; Fontanier et al., 2003; Koho and Piña-Ochoa, 2012). Although *Globobulimina* spp. are usually reported as deeper infaunal, *G. turgida* in the Gullmar Fjord is mostly found < 2 cm depth in the sediment (Risgaard-Petersen et al., 2006; Koho et al., 2008). In general the chemocline in the Gullmar Fjord is located shallower in the seasonally hypoxic fjord's sediment which explains the shallower habitat depth of *G. turgida* in the fjord compared to more oxygenated open-ocean settings.

In this study we explore the potential of using Mg/Ca and Mn/Ca in the benthic foraminiferal species *Bulimina marginata* and *Globobulimina turgida* from the Skagerrak and the Gullmar Fjord as proxies for reconstructing changes in sea water temperature and dissolved oxygen content.

1.1 Hydrography

The Skagerrak is located between Denmark, Sweden, and Norway forming the connection between the North Sea in the west and the Kattegat in the south, which then continues on towards the Baltic Sea (Fig. 1). Water masses enter the Skagerrak both from the south and the northwest. The brackish Baltic Current (salinity ~ 20) usually

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follows the Swedish west coast transporting surface water from the Baltic Sea northwards. The Northern Jutland Current enters the Skagerrak from the west consisting of water masses from the North Sea (salinity of 31–35) as well as from the Atlantic (salinity > 35). These two water masses mix into the Norwegian Coastal Current which leaves the Skagerrak in the northwest creating a cyclonic circulation pattern (Rodhe, 1996).

The Gullmar Fjord is located in southwestern Sweden off the Skagerrak (Fig. 1). The fjord is connected to the Skagerrak by a sill at 42 m water depth. The fjord itself is almost 30 km long and 1–2 km wide. The maximum water depth reaches 120 m. The water masses in the fjord can be divided into three separate layers, including a brackish surface layer consisting of low saline Baltic Current and river water; an intermediate layer from 15–50 m which is in connection with the Skagerrak having salinities of 32–33; and a deep (> 50 m) layer with salinities of 34–35 which is blocked from free exchange with the Skagerrak by the sill. Usually the deep water is renewed once a year in late winter/early spring (Filipsson and Nordberg, 2004a) bringing the oxygen concentration back to open Skagerrak values of $\sim 6 \text{ mL}^{-1}$ (Fig. 2b). After the spring bloom oxygen concentrations start to decrease reaching minimum, hypoxic, values of less than 2 mL^{-1} towards the end of the year (Filipsson and Nordberg, 2004a).

2 Material/methods

2.1 Samples

The samples were collected from site FÅ17 (SMHI site Å17), from the NE Skagerrak ($58^{\circ} 16.50' \text{ N}/10^{\circ} 30.80' \text{ E}$, 330 m water depth) and from site FG113 from the deepest part of the Gullmar Fjord ($58^{\circ} 19.40' \text{ N}/11^{\circ} 32.8' \text{ E}$, 118 m water depth) by a Gemini corer (Fig. 1). The stations were visited in June 2006 (called no. 1); August 2006 (no. 2), January 2007 (no. 3); June 2007 (no. 4) and September 2007 (no. 5), using R/V *Arne Tiselius*. At each station two replicate cores were recovered, labeled A and B.

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The uppermost two cm of the sediment cores were sectioned into 1 cm slices. The vital fluorogenic probe CellTrackerGreen CMFDA (hereafter CellTracker Green) (final concentration 1 μM in ambient sea-water) was added to all samples, except for station no. 1 where Rose Bengal was used instead; the CellTracker Green samples were incubated for 12 h at 7°C, following the protocol by Bernhard et al. (2006). The samples were preserved using 3.8 % borax-buffered formalin. At time of analysis an aliquant of the sediment was carefully sieved over 63 μm screen and selected foraminiferal species were wet-picked under an epifluorescence stereo microscope (Nikon SMZ 1500, filters 485-nm excitation, 520-nm emission) and air dried for geochemical analysis.

Traditionally, Rose Bengal-staining of the samples is used to identify living foraminifera as it fixes onto proteins and colors the protoplasm pink (Walton, 1952; Bernhard, 2000). However, the use of Rose Bengal often results in an overestimation of number of living specimens (Bernhard et al., 2006). This can particular be an issue in low-oxygen environments, where the foraminiferal protoplasm is either slowly or not at all re-mineralized, leading to the occasional staining of dead specimens. In seasonal and short-term studies, as presented here, where comparisons are made to in-situ measurements of the environmental conditions, a much higher accuracy is needed. Therefore, we used the CellTracker Green method (Bernhard et al., 2006; Pucci et al., 2009), which is a non-terminal method, meaning it can be applied during the life stage without killing the specimens. This is not only important in allowing comparison with water mass conditions during the time of sampling but also for interpreting the geochemical results. Especially for Mn/Ca high values are commonly associated with Mn-oxhydroxides and Mn-carbonates which form on the test of buried foraminifera (Boyle, 1983; Barker et al., 2003). The CellTracker Green method ensures that the Mn/Ca values in this study are part of the foraminiferal test itself and not associated with diagenetic coatings.

2.2 Mg/Ca and Mn/Ca

Between 3 and 20 tests of *B. marginata* and *G. turgida* were selected from each sample and gently crushed. Due to the low number of living specimens available the size range of the selected specimens was not restricted. The shell fragments were then cleaned according to the standard cleaning protocol for foraminiferal Mg/Ca analyses (Barker et al., 2003). The tests underwent ultrasonic cleaning alternated with washes in deionized water (Seralpur) and methanol, before an oxidizing step was applied, which was neutralized with multiple deionized water washes (Seralpur). After transfer into clean vials a weak acid leach (0.001 M QD HNO₃) was applied, and samples were dissolved in 0.075 M QD HNO₃. Samples were centrifuged for 10 min (6000 rpm) to exclude any remaining insoluble particles from the analyses. Finally, samples were diluted with Seralpur water and analyzed with an ICP-OES (Perkin Elmer Optima 3300RL with autosampler and ultrasonic nebulizer U5000 AT, Cetac Technologies Inc.) at the Faculty of Geosciences, University of Bremen, Germany. Instrumental precision of the ICP-OES was monitored by analysis of an in-house standard solution with a Mg/Ca of 2.93 mmol mol⁻¹ after every five samples (longterm standard deviation of 0.026 mmol mol⁻¹ or 0.91 %). To allow interlaboratory comparison we analyzed an international limestone standard (ECRM752-1) with a reported Mg/Ca of 3.75 mmol mol⁻¹ (Greaves et al., 2008). The longterm average of the ECRM752-1 standard, which is routinely analysed twice before each batch of 50 samples in every session, is 3.78 mmol mol⁻¹ ($1\sigma = 0.066$ mmol mol⁻¹). Analytical precision based on three replicate measurements of each sample for *B. marginata* and *G. turgida* was 0.39 % ($1\sigma = 0.009$ mmol mol⁻¹) and 0.43 % ($1\sigma = 0.016$ mmol mol⁻¹) for Mg/Ca, resp.; for *G. turgida* Mn/Ca the analytical precision was 1.02 % ($1\sigma = 0.06$ mmol mol⁻¹) for the Gullmar Fjord and 5.52 % ($1\sigma = 0.001$ mmol mol⁻¹) for the Skagerrak. Due the low number of specimens no replicates were measured. Mn/Ca of the samples was corrected offline based on analyses of the ECRM752-1 standard with a published Mn/Ca of 0.132 mmol mol⁻¹ (Greaves et al., 2008).

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the resulting calibrations are different from each other and also from the core top study. Another option to calculate temperatures is the multi-species calibration of Lear et al. (2002) as it also includes analyses on *Bulimina* spp. Temperature fluctuations in the Skagerrak at 300 m water depth during 2007 were less than 2 °C with an average of 7.3 °C (Fig. 2a; SMHI). Applying the different calibrations gives average Mg/Ca-based temperatures of 9.0 °C (Lear et al., 2002), 16.4 °C (Wit et al., 2012), and 3.4 °C (Hintz et al., 2006a), respectively. Comparing our core top Mg/Ca data with the results of cultured *B. marginata* of Filipsson et al. (2010) would suggest temperatures below zero.

Mg/Ca for *G. turgida* ($n = 8$) varies between 3.41 and 4.96 mmol mol⁻¹ (Fig. 3b). The *G. turgida* samples are evenly split between samples taken at 0–1 cm and 1–2 cm, but no systematic difference between the two depths is present. Our results confirm that *Globobulimina* Mg/Ca values are in general significantly higher (Table 2a) than those of commonly used benthic genera like *Cibicidoides*, *Uvigerina*, or also *Bulimina* (Lear et al., 2002; Raitzsch et al., 2008; Huang et al., 2012), but similar to *Globobulimina affinis* (Skinner et al., 2003). The Mg/Ca vs. temperature calibration of *Globobulimina affinis* from Skinner et al. (2003), based on a downcore record from the Iberian Margin, is the only species-specific equation for the genus *Globobulimina*. Applying the species-specific and the multi-species calibrations to our *G. turgida* Mg/Ca data gives average temperatures of 4.2 °C (Skinner et al., 2003) and 14.2 °C (Lear et al., 2002), respectively, compared to the average temperature of 7.3 °C (Fig. 2a; SMHI) for measured Skagerrak water.

3.2 Gullmar Fjord

Only one sample from a slightly shallower location at 70 m water depth yielded enough specimens of *B. marginata* to analyze. Mg/Ca of this sample was 2.15 mmol mol⁻¹ similar to the Mg/Ca from the Skagerrak samples. The in-situ temperature during June 2007 when the samples were collected was 6.7 °C, similar to the temperatures from the Skagerrak.

Due to the matrix effect when analyzing low concentrations on the ICP-OES only three out of eight samples for *G. turgida* yielded enough material for reliable Mg/Ca values (Table 2b). In-situ temperatures at the time of sampling varied between 5.4 and 6.0 °C, slightly lower than the Skagerrak temperatures. Mg/Ca values varied between 2.80 and 2.97 mmol mol⁻¹, i.e. significantly lower than those from the Skagerrak samples, which would mean temperatures around the freezing point when applying the calibration of Skinner et al. (2003).

3.3 Mn/Ca

3.3.1 Skagerrak

Measurable Mn-concentrations were only present in *G. turgida*. Mn/Ca for *G. turgida* varied between 0 and 0.09 mmol mol⁻¹ with the samples from 1–2 cm depth having higher Mn/Ca than the samples from 0–1 cm depth sampled at the same time (Fig. 4a). Oxygen concentrations between 5.5 and 6.5 mL L⁻¹ confirm that the bottom water in the Skagerrak was well oxygenated throughout the year. An extensive collection of pore water profiles from the Skagerrak shows that oxygen penetration generally occurs down to 5–20 mm sediment depth (Bakker and Helder, 1993). As such, it is likely that the *G. turgida* specimens from 1–2 cm sediment depth calcified under lower oxygen conditions than those from 0–1 cm sediment depth.

3.3.2 Gullmar Fjord

Mn/Ca for *G. turgida* in the Gullmar samples was significantly higher than for the samples from the Skagerrak. Mn/Ca varied between 0.20 and 5.8 mmol mol⁻¹ (Fig. 4b), while two additional samples, which yielded Mn/Ca of 7.94 and 10.58 mmol mol⁻¹, are likely to be biased by a matrix effect (Table 2b). Bottom water oxygen concentrations in 2006 and 2007 followed the common pattern for the fjord, which includes one annual deep water exchange with subsequent replenished dissolved oxygen content

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(5–6 mL L⁻¹) (Fig. 4b). The oxygen concentrations decrease for the remainder of the year again towards hypoxic values. The water samples with the highest Mn/Ca were collected during the time with the lowest oxygen concentrations (January 2007 and April 2007).

4 Discussion

Before the trace metal/Ca results are discussed it is necessary to consider the life cycle of *B. marginata* and *G. turgida*. Unlike shallow-dwelling planktonic foraminifera that live and calcify only for two to four weeks, many benthic foraminiferal species survive longer. It is essential to know for the discussion of any geochemistry data, especially acquired from benthic foraminifera from variable environments, when the used species calcify as this determines the geochemical signal. Culturing experiments under anoxic conditions showed that *G. turgida* survived for several months, utilizing their internal nitrogen pool (Risgaard-Nielsen et al., 2006; Piña-Ochoa et al., 2010). A remaining question is, however, if the specimens also grow during the anoxic conditions. Reproduction and increased abundances often seem to follow peaks in food supply (Gustafsson and Nordberg, 1999, 2000, 2001; Kitazato et al., 2000; Fontanier et al., 2003; Filipsson et al., 2004; Koho and Piña-Ochoa, 2012). In the Havstens and Gullmar Fjords a strong correlation between the occurrence of the benthic foraminifera *Stainforthia fusiformis* and chlorophyll-*a* concentrations was found, which was also imprinted onto the $\delta^{13}\text{C}$ values (Filipsson et al., 2004). This would suggest that the geochemical signal which is actually recorded into the foraminiferal tests is representative for a restricted period directly after maximum food supply.

In the following chapters we discuss the analysed variations in Mg/Ca and Mn/Ca for *B. marginata* and *G. turgida* with respect to proposed impact factors like temperature, salinity, carbonate system, and dissolved oxygen concentration.

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4.1 Mg/Ca

4.1.1 Mg/Ca in *B. marginata*

As mentioned in the results converting our Mg/Ca data of *B. marginata* into calcification temperatures using existing calibrations does not give temperatures which are similar to the in-situ measured temperatures (Fig. 3a). The calibration of Wit et al. (2012) results in temperatures which are 5–14 °C higher than measured, while the calibration of Hintz et al. (2006a) gives temperatures which are 5 °C or more colder. Why are these differences in Mg/Ca between the different studies so large assuming that Mg/Ca in *B. marginata* in the Skagerrak is indeed representative of calcification temperatures? And additionally, why is the scatter in the results much larger than is commonly seen in planktonic or deep sea benthic foraminiferal Mg/Ca?

One possible reason for the large spread in Mg/Ca could be the heterogeneous Mg distribution between and within shells from the same sample (e.g. Eggins et al., 2003; Rathmann et al., 2004; Hathorne et al., 2009). Hintz et al. (2006a) showed a range in the distribution coefficient of Mg from 0.5–15.1. It is therefore necessary to include a sufficient number of shells when analyzing Mg/Ca to obtain a representative average value for the sample. Although this would explain the scatter in the data which result from using Laser Ablation it does not explain why absolute values between the different studies have different ranges. It is likely that the data from Filipsson et al. (2010) included excess Mg due to the presence of either organic material or an amorphous calcium carbonate phase (Barker et al., 2003; Addadi et al., 2003; Hathorne et al., 2009). Due to the fragility of the cultured tests no cleaning was performed before raster analyses on the newly grown calcite resulting in Mg/Ca being higher than in any of our samples (Fig. 3a). The data of Wit et al. (2012) on the other hand, are all lower than our results apart from the specimens grown at 14 °C, which is ~ 7 °C higher than our settings. The number of specimens used for our bulk analyses ranged between 6 and 15, providing an average signal that should be representative of the water mass temperature (7.1–7.3 °C) at the time of collection. The variation in Mg/Ca, however,

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suggests a spread of 3.9–6.0 °C depending on the choice of calibration, which is significantly larger than the error commonly associated with benthic Mg/Ca vs. temperature calibrations of 0.5–1.5 °C (e.g. Lear et al., 2002; Huang et al., 2012).

Additionally, it cannot be excluded that there is a size effect imprinted on the results.

5 Due to the low number of living specimens available for analyses we combined all size fractions possibly introducing a bias. Mg/Ca of the bulk results of Hintz et al. (2006) varied between 2.0–4.8 mmol mol⁻¹ (not including a possible outlier) with no apparent size trend, which was also suggested by Wit et al. (2012). The Mg/Ca of the single specimens, on the other hand, showed a significant size effect with specimens in the
10 mid-range size (150–225 µm) having much higher values (Hintz et al., 2006b). This suggests that also for benthic foraminifera it is necessary to limit the analyses to narrow size fractions. Apart from the relatively large scatter in the data the bulk analyses of Hintz et al. (2006b) and our bulk Mg/Ca results do cover the same range of values providing a basis to construct a future bulk Mg/Ca vs. calcification temperature
15 calibration.

As the scatter in the data cannot directly be linked to variations in temperature, additional factors of influence on Mg/Ca have to be considered. The impact of salinity on Mg/Ca has recently received significant attention based both on benthic and planktonic foraminifera (e.g. Nürnberg, 1995; Lea et al., 1999; Kisakürek et al., 2008; Groeneveld et al., 2008; Mathien-Blard and Bassinot, 2009; Dissard et al., 2010; Diz et al., 2012). In
20 general, higher salinity leads to higher Mg/Ca, although with a highly variable impact of increasing Mg/Ca with just 5% per salinity unit (Kisakürek et al., 2008) to increases as high as 15–59% per salinity unit (Ferguson et al., 2008). Especially when considering the variations in salinity in natural settings, most of the determined changes in Mg/Ca were found either over very large salinity ranges (> 10) or at high salinity settings like
25 the Red Sea or the Mediterranean (Ferguson et al., 2008; Hoogakker et al., 2009; Van Raden et al., 2011). Salinity in the deep Skagerrak is very stable throughout the year (35.0–35.3) both similar to open-ocean settings and to the culturing experiments of Hintz et al. (2006a). The impact of low-salinity (20–30) surface water coming from the

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Baltic proper, via the Baltic Current, does not reach the deep Skagerrak. Thus, it can be excluded that salinity is having a significant impact on our Mg/Ca results.

A factor known to be significant in the deep-sea is the carbonate ion effect (Elderfield et al., 2006; Raitzsch et al., 2008). Especially at water temperatures $< 3\text{--}4^\circ\text{C}$ the change in the carbonate ion concentration dominates over the influence of temperature on Mg/Ca, mainly caused by increasing water depth, i.e. pressure, and the impact of water masses with a different carbonate ion saturation state, e.g. North Atlantic Deep Water vs. Antarctic Bottom Water (Raitzsch et al., 2008). Conditions in the deep Skagerrak show over-saturated values with $\Omega = 2.77$ and a carbonate saturation state of $77.1 \mu\text{mol kg}^{-1}$ (values calculated with CO_2SYS ; Pierrot et al., 2006; Table 1). In combination with a water temperature of $\sim 7^\circ\text{C}$ this does not directly suggest a significant impact on the Mg/Ca results.

4.1.2 Mg/Ca in *Globobulimina turgida*

Our Mg/Ca results on *G. turgida* are among the first data available for *Globobulimina* spp. covering the same range of values as for *Globobulimina affinis* between $2.2\text{--}4.9 \text{ mmol mol}^{-1}$ (Skinner et al., 2003). The Mg/Ca calibration of *G. affinis* (Skinner et al., 2003), however, was defined for a temperature range between -1.8°C and $\sim 3.5^\circ\text{C}$ resulting in too low temperatures when applied to our data. Applying the Skinner et al. (2003) calibration equation would result in an average temperature for the Skagerrak of 4.2°C and for the Gullmar Fjord of $\sim 0^\circ\text{C}$ while in-situ temperatures at the time of collection were 7.3°C and 6.2°C , respectively. Due to generally higher Mg/Ca ratios of *Globobulimina* spp. the multi-species calibration of Lear et al. (2002) gives temperatures between $10\text{--}14^\circ\text{C}$ which are significantly higher than measured. As with *B. marginata* the lack of available published data for comparison makes it difficult to determine if the Mg/Ca is indeed showing temperature or also showing the influence of other factors. The spread in the Skagerrak *G. turgida* Mg/Ca data is as large as for *B. marginata* (Fig. 3b), possibly also being caused by the large size range of the analyzed specimens. Even though the bottom water mass characteristics between Skagerrak

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and the Gullmar Fjord concerning temperature and salinity are similar (Table 1), there is a significant difference in Mg/Ca of *G. turgida*. Average foraminiferal Mg/Ca in the Skagerrak is 4.11 mmol mol⁻¹ vs. 2.90 mmol mol⁻¹ in the Gullmar Fjord. This difference cannot be explained by the small temperature difference of 1.1 °C implying that another factor is present.

Bottom water temperature in the Gullmar Fjord is higher (6.2 °C) than the threshold of 3–4 °C, suggested for the carbonate ion saturation state to have a major influence on Mg/Ca (Elderfield et al., 2006; Raitzsch et al., 2008). Nevertheless, the parameters of the carbonate system are different in the deep Gullmar Fjord when compared to the deep Skagerrak. The carbonate ion concentration is significantly lower, leading to less saturated bottom water with respect to calcite ($\Omega = 1.7$ and $\Delta(\text{CO}_3^{2-}) = 29.1 \mu\text{mol kg}^{-1}$; values calculated with CO₂SYS; Pierrot et al., 2006; Table 1). If we hypothetically study the differences between the deep water in the Gullmar Fjord and the Skagerrak using existing dependencies for benthic foraminifera, the difference in Mg/Ca of *G. turgida* from the Skagerrak and the Gullmar Fjord could be explained as a combined effect of temperature and $\Delta(\text{CO}_3^{2-})$. The difference in temperature of 1.1 °C would account for a $\Delta(\text{Mg}/\text{Ca})$ of 0.35–0.45 mmol mol⁻¹ (Lear et al., 2002; Skinner et al., 2003). Using the dependency of $\Delta(\text{Mg}/\text{Ca})$ on $\Delta(\text{CO}_3^{2-})$ determined on *Cibicides mundulus* and *Planulina wuellerstorfi* (Raitzsch et al., 2008) the difference in Mg/Ca is 0.48–0.82 mmol mol⁻¹. Taken together, the expected difference between Mg/Ca in the Skagerrak and Gullmar Fjord would be 0.83–1.27 mmol mol⁻¹. This would be sufficient to explain the observed difference for *G. turgida* Mg/Ca of 1.21 mmol mol⁻¹.

Additionally, a bottom water mass characteristic which is also significantly different between the Skagerrak and the Gullmar Fjord is the low dissolved oxygen content during most of the year in the fjord. *Globobulimina turgida* is a foraminifera species which is able to respire nitrate through denitrification via an internal nitrate pool (Risgaard-Petersen et al., 2006; Piña-Ochoa et al., 2010) allowing its survival in low-oxygen environments. But although it was shown in culturing experiments that *G. turgida* maintains its metabolic activity, it remains unclear if the foraminifera also calcify and reproduce

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under these conditions (Piña-Ochoa et al., 2010). Elderfield et al. (1996) hypothesized that there is a direct link between growth rate and partitioning coefficients of trace metal incorporation into calcite, although this was not shown explicitly for Mg. We speculate that in a lower oxygen environment the lower metabolic activity might also decrease the distribution coefficient of Mg which could potentially have contributed to the lower *G. turgida* Mg/Ca in the Gullmar Fjord compared with the Skagerrak.

4.2 Mn/Ca as a potential proxy for oxygen concentration

Traditionally, Mn/Ca in foraminifera is used as an indicator for the presence of diagenetic Mn-oxyhydroxides and Mn-carbonates, which can contain significant concentrations of other trace metals like Cd and Mg (Boyle, 1983; Barker et al., 2003). Typical Mn/Ca in cleaned foraminifera, which are considered not to be significantly affected by coatings, is $< 0.1 \text{ mmol mol}^{-1}$ (Barker et al., 2003). For the development of Mn/Ca as proxy for oxygen concentrations it is essential to determine where the Mn is located in the foraminifera, either incorporated into the tests or as coating. Manganese is a redox-sensitive element, mainly present as Mn^{2+} in sea water but precipitating as a Mn-oxyhydroxide under oxygenated conditions. However, under low oxygen conditions the Mn-oxyhydroxide is reduced and the Mn^{2+} concentration increases (Tribovillard et al., 2006). Especially pore water can become super-saturated with respect to Mn under low-oxygen conditions resulting in inorganic precipitation of Mn-carbonate (MnCO_3 or Rhodochrosite). Thus, when benthic foraminifera live and precipitate calcium carbonate under lower oxygen conditions, more Mn will be incorporated into their calcite tests and Mn/Ca may therefore be used as a proxy for low oxygen conditions. Based on theory and precipitation experiments of inorganic calcite the variation in distribution coefficients for Mn into inorganic calcite varies between 1 and 1700 (e.g. Pingitore, 1978; Franklin and Morse, 1983), and laser ablation measurements on benthic foraminifera from the Arabian Sea and the Gulf of Biscay confirmed the incorporation of Mn into foraminiferal calcite (Reichert et al., 2003). The large spread in distribution coefficients for Mn is also suggested for benthic foraminifera, which display a large variation of

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Mn/Ca ratios. A laser ablation study on the aragonitic *Hoeglundina elegans* for sapropel S1 from the Mediterranean showed Mn/Ca between 1–10 $\mu\text{mol mol}^{-1}$ (Flaithearta et al., 2010) similar to Mn/Ca in the calcitic *Uvigerina peregrina* and *Bolivina spissa* from below the Oxygen Minimum Zone (OMZ) off the coast of Peru determined by secondary ion mass spectrometry (SIMS; Glock et al., 2012). Mn/Ca of *U. peregrina* from below the OMZ in the eastern tropical North Pacific, on the other hand, varied from 0.05–0.2 mmol mol^{-1} , analysed using Flow Through time-resolved analysis (Klinkhammer et al., 2009). The different analytical techniques used in these studies all aimed at preventing to have Mn related diagenetic coatings included in the analyses. This is also the reason why the specimens used in this study have all been labeled by Cell-Tracker Green ensuring that they were alive during collection and, thus, could not yet have been affected by diagenetic coatings.

4.2.1 Mn/Ca in the Skagerrak

Bottom water in the Skagerrak is well oxygenated throughout the year with oxygen concentrations varying between 5.5–7 mL L^{-1} (Fig. 2a; SMHI, 2007, 2008). The Mn concentrations in the bottom water were all below the detection limit. *B. marginata* tends to live closer to the sediment surface where abundant oxygen is available (e.g. Gustafsson and Nordberg, 2000), while *G. turgida* extends its microhabitat down into the sediment (e.g., Jorissen et al., 1995). The lack of Mn in the bottom water translates to negligible Mn/Ca in the tests of *B. marginata* (Table 2). For *G. turgida* on the other hand, even though the specimens were sampled from the same horizons as *B. marginata*, a clear distinction is present between the samples from 0–1 cm and those from 1–2 cm. Opposite to *B. marginata* the Mn/Ca in the surface *G. turgida* has values up to 0.07 mmol mol^{-1} . The results from the deeper (1–2 cm) samples all display higher Mn/Ca, reaching 0.09 mmol mol^{-1} (Fig. 4a; Table 2). Bakker and Helder (1993) analysed oxygen concentrations in pore water profiles from a collection of sediment cores from the Skagerrak. Typical profiles show oxygen penetration into the sediment from < 5 mm for the shallower locations to 15–20 mm for deeper locations. Comparing their

profiles with the water depth of our study location suggests that oxygen penetrated the sediment down to ~ 5 mm. Accordingly, Mn-concentrations in the pore water increase to values of $50\text{--}100 \mu\text{mol L}^{-1}$ below the oxic zone in the sediment (Bakker and Helder, 1993) leading to increased incorporation of Mn into the tests of *G. turgida*. Additionally, we speculate that the *B. marginata* specimens were mainly living in the upper 5 mm while *G. turgida* might have been restricted to 5–10 mm when considering the 0–1 cm sample and their respective Mn/Ca. These Mn/Ca values are still significantly lower than the values from the Gullmar Fjord where hypoxic conditions prevail for a large part of the year.

4.2.2 Mn/Ca in Gullmar Fjord

Mn/Ca in the Gullmar Fjord samples is significantly different from the Skagerrak samples with partially extremely high foraminiferal Mn/Ca values (Fig. 4b; Table 2). As the deeper water in the Gullmar Fjord only gets re-noved once a year usually during late winter/early spring, oxygen concentrations decrease during the rest of the year, being lower than 2 mL L^{-1} during autumn and winter (Fig. 2b). As dissolved Mn is not directly precipitated as it is under lower oxygenated conditions, higher Mn-concentrations occur in the bottom water of the Gullmar Fjord than in the Skagerrak, reaching concentrations between $3.7\text{--}10.3 \mu\text{mol L}^{-1}$, rapidly increasing to $30\text{--}250 \mu\text{mol L}^{-1}$ in the pore water (Goldberg et al., 2012). Due to the already low bottom water oxygen concentrations *G. turgida* tends to live near the sediment surface (Risgaard-Petersen et al., 2006). The Mn/Ca values in *G. turgida* vary between $0.2\text{--}5.8 \text{ mmol mol}^{-1}$, significantly higher than those in the Skagerrak (Table 2). As CellTracker Green was used to ensure that analysed specimens were alive during collection we can make a direct link between Mn/Ca in the bottom water and in the foraminiferal calcite determining distribution coefficients. A potential bias on the possible distribution coefficients is caused by the lack of pore water chemistry data with likely even higher Mn-concentrations than the bottom water itself (Goldberg et al., 2012). The calculated distribution coefficient for Mn into the tests of *G. turgida* varies between 264 and 712. For the low-oxygen conditions encountered

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in this study the distribution coefficient for Mn is similar to that of inorganic calcite, but significantly higher than the value of 2.4 estimated for cultured *Ammonia tepida* (e.g. Pingitore, 1978; Franklin and Morse, 1983; Munsel et al., 2010).

4.3 Potential as paleoproxies

4.3.1 Mg/Ca

The Mg/Ca results in this study are difficult to align with currently existing Mg/Ca vs. temperature calibrations. Likely reasons for this are the different ways the different studies have achieved their results, from laser ablation analyses on chamber parts of cultured specimens to bulk analyses on specimens from core tops. The conditions in the Skagerrak are similar to open-ocean conditions suggesting, especially for *B. marginata*, that the application of Mg/Ca as paleothermometer as hitherto commonly applied has the potential to work. What is needed though is an effort to establish a Mg/Ca vs. calcification temperature calibration based on multi-specimen samples with a narrow size fraction from a wide range of core top settings. In the case of *G. turgida* the significant difference between the samples from the Skagerrak and the Gullmar Fjord can be explained by a significant impact of differences in the carbonate system between the Skagerrak and the Gullmar Fjord. Additionally, we speculate that there might be an additional impact of the low-oxygen conditions on the incorporation of Mg in foraminiferal tests.

4.3.2 Mn/Ca

Mn/Ca in benthic foraminifera which are tolerant of low-oxygen conditions like *G. turgida* has the potential to be developed as an indirect proxy to variations in low dissolved oxygen conditions. Our results show a clear increase in foraminiferal Mn/Ca with increasing Mn concentrations in the bottom water due to low-oxygen conditions. The redox behavior of Mn, however, provides some issues for reconstructing downcore

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records which need to be resolved. The specimens used in this study were alive when collected and, thus, were not affected by diagenetic coatings. But after burial in the sediment the tests can be covered with diagenetic coatings either resulting from oxidizing conditions (Mn-oxyhydroxides) or reducing conditions (Mn-carbonates) (Boyle, 1983; Klinkhammer et al., 2009). So, not only is it necessary to determine when and/or where in the sediment the foraminifera incorporate the Mn-signal into their tests, this primary signal also needs to be extracted without including the additional Mn from any coatings. The traditional reduction step for cleaning foraminiferal samples only removes the Mn-oxyhydroxides (Boyle, 1983; Barker et al., 2003). Different techniques have made an effort to exclude the Mn-carbonate from the analyses, although the resulting Mn/Ca in the foraminifera still varied several orders of magnitude (Klinkhammer et al., 2009; Flaithearta et al., 2010; Glock et al., 2012). To determine whether this range in values is indeed primary or an artifact of the analytical techniques, culturing experiments under varying oxygen conditions might provide the opportunity to restrict the range of Mn/Ca values which can be expected in downcore records.

5 Conclusions

In this study we have explored variations of Mg/Ca and Mn/Ca in live benthic foraminifera, *Bulimina marginata* and *Globobulimina turgida*, with respect to changes in bottom water temperature and dissolved oxygen content in shelf regions. Foraminifera and water samples were collected from the Skagerrak and the Gullmar Fjord (SW Sweden) over a two-year period. Staining with CellTracker Green ensured that the specimens were alive when collected and, thus, were not affected by diagenetic coatings.

The range in Mg/Ca of 1.90–2.85 mmolmol⁻¹ for *B. marginata*, and 2.80–4.96 mmolmol⁻¹ for *G. turgida* is similar to other studies. However, Mg/Ca results for both species show much more variation than would be expected from the influence of the annual temperature change of less than 2 °C using existing Mg/Ca vs. temperature

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calibrations. Taking differences in the carbonate ion system between the Skagerrak and the Gullmar Fjord into account would explain these results.

Mn/Ca results on *G. turgida* show potential to record variations in dissolved oxygen content of the habitat where they calcify. Samples from the Skagerrak show increased Mn/Ca (on average $0.05 \text{ mmol mol}^{-1}$) in specimens which lived deeper in the sediment (1–2 cm) than those that lived near the surface (on average $0.02 \text{ mmol mol}^{-1}$; 0–1 cm). *G. turgida* samples from the lower oxygen Gullmar Fjord showed significantly increased Mn/Ca, being highest ($10.58 \text{ mmol mol}^{-1}$) when dissolved oxygen was lowest.

Our study shows that trace metal/Ca ratios in benthic foraminifera from shelf regions have the potential to record past variations in bottom water temperature and dissolved oxygen concentrations, but calibration studies based on both core tops and culturing are needed to resolve the possible extend of variation.

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H. L. Filipsson**Table 1.** Average water mass characteristics, including temperature, salinity, carbonate system parameters, sea water Mg/Ca, and Mn/Ca for the Skagerrak (Site FÅ17) and the Gullmar Fjord (Site FG113).

	Skagerrak	Gullmar Fjord
Temperature	7.3 °C	6.2 °C
Salinity	35.21	34.46
Dissolved Oxygen	6.03 mL L ⁻¹	0.9–6.6 mL L ⁻¹
Total Alkalinity	¹ 2319 μmol kg ⁻¹	² 2300 μmol kg ⁻¹
Dissolved Inorganic Carbon	¹ 2151 μmol kg ⁻¹	² 2225 μmol kg ⁻¹
⁴ CO ₃ ⁼	120.6	70.6
⁴ Δ(CO ₃ ⁼)	77.1	29.1
⁴ Omega	2.77	1.7
⁴ pH	8.15	7.91
Mg/Ca	5.04 mol mol ⁻¹	5.00 mol mol ⁻¹
Mn/Ca ³	n.a.	6.07 μmol mol ⁻¹

¹ Hjalmarsson et al. (2010).² Ulfssbo, MSc, Univ. Göteborg (2009).³ Mn concentrations were below the detection limit for the Skagerrak and 3.7–10.3 μmol L⁻¹ for the Gullmar Fjord.⁴ Carbonate system parameters were calculated using the CO₂SYS (Pierrot et al., 2006) with the constants of Mehrbach et al. (1973) and pH on the sea water scale.

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H. L. Filipsson**Table 2a.** Water mass characteristics during sample collection in the Skagerrak and Mg/Ca and Mn/Ca for *B. marginata* and *G. turgida*.

Sample ¹	Sediment depth (cm)	Temperature (in-situ, °C)	Salinity	Oxygen (mL L ⁻¹)	<i>B. marginata</i> Mg/Ca (mmol mol ⁻¹)	<i>B. marginata</i> Mn/Ca (mmol mol ⁻¹)	<i>G. turgida</i> Mg/Ca (mmol mol ⁻¹)	<i>G. turgida</i> Mn/Ca (mmol mol ⁻¹)
4A	0–1	7.31	35.22	6.14	1.90	b.d.l. ²	3.41	b.d.l.
4B	0–1	7.31	35.22	6.14	2.18	b.d.l.	4.07	0.072
5A	0–1	7.09	35.20	5.85	2.85	b.d.l.	4.15	0.003
5B	0–1	7.09	35.20	5.85	2.27	b.d.l.	4.96	0.020
4A	1–2	7.31	35.22	6.14	2.42	b.d.l.	3.79	0.058
4B	1–2	7.31	35.22	6.14	n.a. ³	n.a.	4.30	0.090
5A	1–2	7.09	35.20	5.85	n.a.	n.a.	3.94	0.015
5B	1–2	7.09	35.20	5.85	n.a.	n.a.	4.25	0.035

¹ Samples 4A and 4B were collected June 2007; samples 5A and 5B were collected September 2007.² b.d.l. = Mn concentrations below the detection limit.³ n.a. = No *B. marginata* specimens were available in these samples.

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H. L. Filipsson**Table 2b.** Water mass characteristics during sample collection in the Gullmar Fjord and Mg/Ca and Mn/Ca for *G. turgida*.

Sample	Date of collection	Temperature (in-situ, °C)	Salinity	Oxygen (mL L ⁻¹)	Ca conc. (ppm)	<i>G. turgida</i> Mg/Ca (mmol mol ⁻¹)	<i>G. turgida</i> Mn/Ca (mmol mol ⁻¹)
2	Feb 2006	5.41	34.45	6.59	59.55	2.93	2.73
8	Aug 2006	5.97	34.50	3.54	28.68	2.97	0.20
13	Jan 2007	5.95	34.41	1.6	0.82 ¹		10.58 ¹
13	Jan 2007	5.95	34.41	1.6	1.88 ¹		7.94 ¹
18	Jun 2007	6.69	34.46	3.7	7.65 ¹		4.30 ¹
18	Jun 2007	6.69	34.46	3.7	3.38 ¹		4.16 ¹
21	Sep 2007	6.75	34.46	1.5	4.89 ¹		5.80 ¹
21	Sep 2007	6.75	34.46	1.5	13.01	2.80	4.76
18	Jun 2007	6.69	34.43	3.6	17.51	2.15 ²	b.d.l.

¹ Due to the matrix effect on very small samples Mg/Ca on samples with Ca concentrations lower than 5–10 ppm are considered to be biased; Mn/Ca on samples with Ca concentrations lower than 3 ppm are considered to be biased. The two samples with highest Mn/Ca and lowest Ca concentrations are considered to be under-estimated based on a matrix experiment using the ECRM752-1 standard of Greaves et al. (2008).

² From 70 m water depth (Site FG70) one sample yielded enough *B. marginata* ($n = 10$) for analysis.

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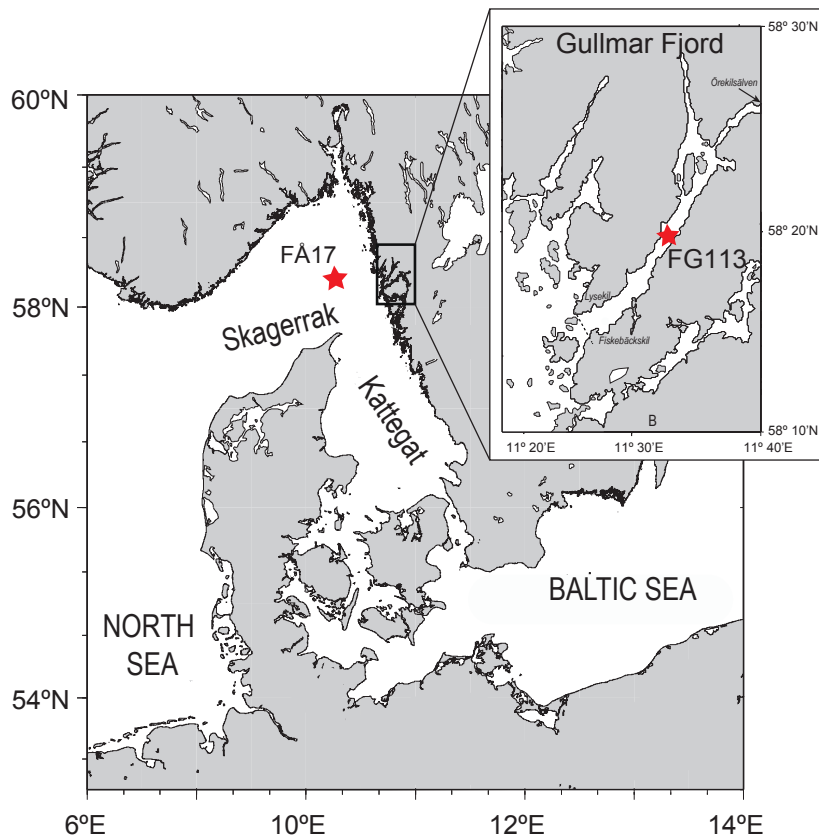


Fig. 1. Map showing the locations of sites FÅ17 and FG113 (inset) where samples were collected. Site FÅ17 is located in the Skagerrak, while Site FG113 is from the deepest, central part of the Gullmar Fjord.

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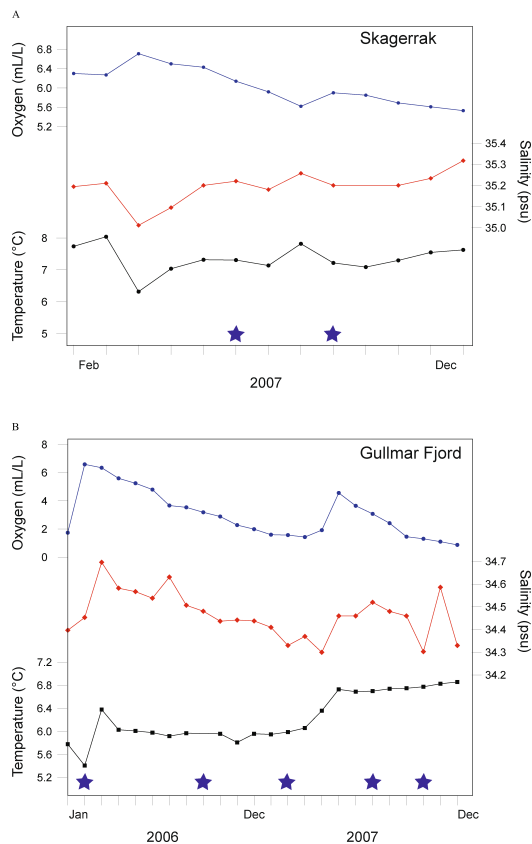
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Fig. 2. (a) Annual cycle of temperature, salinity, and dissolved oxygen at 300 m water depth at the location of Site FÅ17 in the Skagerrak for 2007. (b) Temperature, salinity, and dissolved oxygen at 110 m water depth at the location of Site FG113 in the Gullmar Fjord for 2006 and 2007. Stars (lila) indicate the month of sampling of the living foraminifera.

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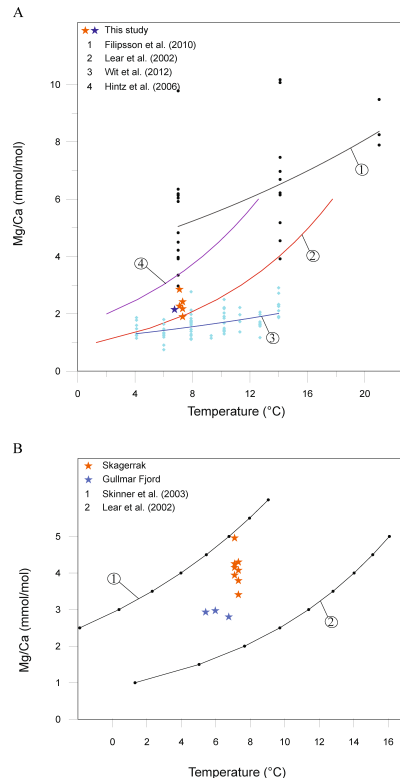
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Fig. 3. (A) Mg/Ca vs. temperature for *B. marginata* from this study (stars; orange (Skagerrak), blue (Gullmar Fjord)) in comparison with published data for *B. marginata/aculeata* (Filipsson et al., 2010; “1”); multiple species (Lear et al., 2002; “2”); *B. marginata* (Wit et al., 2012; “3”); and *B. marginata* (Hintz et al., 2006a; “4”). Single measurements of Filipsson et al. (2010) and Wit et al. (2010) were included to show the scatter when Laser Ablation is used on single specimens/chambers in comparison with our bulk measurements. Note that the results of the lowest temperature experiment of Filipsson et al. (2010) are omitted from the figure. Error bars are too small to be displayed (see Sect. 2). **(B)** Mg/Ca vs. temperature for *G. turgida* from this study from the Skagerrak (stars; orange) and the Gullmar Fjord (stars; blue) in comparison with the calibrations for *G. affinis* (Skinner et al., 2003; “1”); and for multiple species (Lear et al., 2002; “2”). Note that the points in the two calibrations do not represent samples. Error bars are too small to be displayed (see Sect. 2).

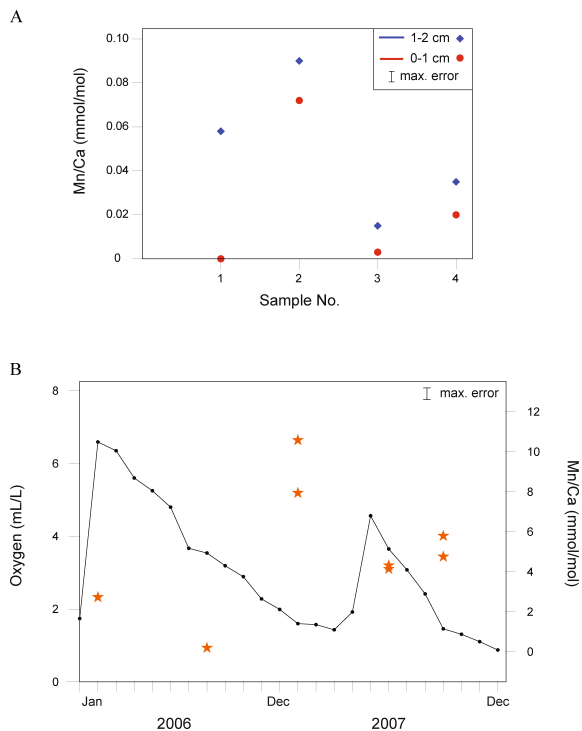


Fig. 4. (A) Mn/Ca of *G. turgida* for the Skagerrak for 0–1 cm (circles; red) and 1–2 cm (diamonds; blue) sediment depth, showing higher values deeper in the sediment corresponding to lower dissolved oxygen concentrations deeper in the sediment. Maximum error corresponds to the largest analytical error for the displayed samples. **(B)** Mn/Ca of *G. turgida* (stars; orange) from the Gullmar Fjord vs. dissolved oxygen concentrations in the water column for 2006 and 2007. When bottom water dissolved oxygen decreases, Mn/Ca in *G. turgida* increases. Maximum error corresponds to the largest analytical error for the displayed samples.