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Effect of salinity and seawater calcite saturation state on Mg and Sr incorporation in cultured planktonic foraminifera

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ABSTRACT

Trace elements incorporated in planktonic foraminiferal test carbonate are commonly used as paleoproxies. For instance, Mg/Ca ratios are frequently used for reconstructing sea surface temperature and, together with the foraminiferal stable oxygen isotope ratios, are also used as paleosalinity proxy. Foraminiferal Sr/Ca ratios constitute another example of the application of trace elements in paleostudies since they may reflect the Sr/Ca values of seawater. However, over the past few decades it has been proven that the incorporation of trace elements in foraminiferal calcite is controlled by more than one environmental parameter. To quantify the effect of salinity on Mg and Sr incorporation planktonic foraminifera *Globigerinoides sacculifer* (sensu stricto) were grown in the laboratory under different environmental conditions. Laboratory experiments allowed us to separate a direct salinity effect from a possible independent impact through differences in the calcite saturation state of the seawater (Ω). Although the temperature effect is more important than the salinity effect, a change of 4 salinity units is equivalent to a 1 °C bias on Mg/Ca-based temperatures. This effect of salinity on Mg incorporation is minor. However, when using Mg/Ca-based temperatures in combination with foraminiferal $\delta^{18}O$ to calculate salinity, it cannot be neglected. The present study shows salinity as the overriding control on Mg incorporation within the range of Ω studied (Ω between 5.25 and 6.50; $[CO_3^{2-}]$ between 218 and 270 µmol/kg) at a constant temperature of 26 °C. In contrast, Ω appears to be the main control on foraminiferal Sr incorporation (0.10 mmol/mol per 100 µmol/kg rise in $[CO_3^{2-}]$), whereas salinity has a non significant influence on Sr/Ca.

1. Introduction

Over the past two decades temperature reconstructions have become increasingly reliant on Mg/Ca ratios measured on planktonic and benthic foraminiferal shells (Nürnberg et al., 1996a; Lea et al., 1999; Russell et al., 2004; Rosenthal et al., 2006; Yu and Elderfield, 2008). The incorporation of Sr into foraminiferal calcite may also show temperature dependence (Lea et al., 1999; Reichart et al., 2003; Mortyn et al., 2005). However other parameters might also be involved in the control of foraminiferal Mg/Ca and Sr/Ca ratios namely pressure (Elderfield et al., 1996), calcite saturation (Lea, 1999), carbonate ion concentration ($[CO_3^{2-}]$) (Mortyn et al., 2005; Rosenthal et al., 2006; Hendry et al., 2009), ontogeny (Ni et al., 2007) and growth rate (Kısakürek et al., 2008). Accurate temperature reconstructions, therefore, rely on an understanding of these additional factors that can potentially influence the incorporation of Mg and Sr in foraminiferal calcite.

Through culture experiments salinity showed to exert a small but consistent influence on foraminiferal Mg/Ca ratios (Nürnberg et al., 1996a; Lea et al., 1999; Kısakürek et al., 2008). More recently, Ferguson etal. (2008) suggested that salinity had a greater than hitherto expected influence on foraminiferal Mg/Ca ratios (15 to 59% increase per salinity unit) using a core-top calibration from the Mediterranean Sea. Such a large salinity dependency would imply that Mg/Ca-based temperature reconstructions can be severely biased (e.g. Ferguson et al., 2008; Groeneveld et al., 2008). Foraminiferal Sr/Ca ratios are also affected by salinity (i.e. 4% increase per salinity unit in *Orbulina universa* (Lea et al., 1999)). However, it remains unclear whether salinity itself influences

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foraminiferal incorporation of these two trace elements or whether it is due to changes in the carbonate chemistry associated with the changes in salinity. Since both salinity and TA are conservative parameters, by increasing salinity, carbonate parameters, such as total alkalinity (TA), dissolved inorganic carbon (DIC) and [CO_3^{2-}] also increase.

Seawater carbonate ion concentration [CO_3^{2-}] has an effect on planktonic and benthic foraminiferal Mg/Ca and Sr/Ca ratios as well (Russell et al., 2004; Mortyn et al., 2005; Elderfield et al., 2006; Rosenthal et al., 2006; Kısakürek et al., 2008; Rathmann and Kuhnert, 2008; Yu and Elderfield, 2008; Raitzsch et al., 2008). When [CO_3^{2-}] of the seawater changes, the calcium carbonate saturation state of seawater (Ω) also changes since $\Omega = [Ca^{2+}]*[CO_3^{2-}]/K_{sp}$ (where K_{sp} represents the solubility product at the *in situ* conditions of temperature, salinity and pressure (Zeebe and Wolf-Gladrow, 2005)). The carbonate saturation state might control the rate of calcite precipitation, which in turn is known to influence trace metal incorporation. This varies depending on the particular trace element studied (Lorens, 1981; Nehrke et al., 2007).

Here we present the results of culture experiments in which salinity is kept constant while the [CO_3^{2-}] varies and vice versa. In the marine environment [CO_3^{2-}] covaries with salinity, making it difficult to evaluate these parameters independently. Moreover, since salinity not only affects [CO_3^{2-}], but also [Ca^{2+}], Ω changes quadratically, potentially enhancing its impact. Therefore, the aim of the present culture study is to separate the effects of both salinity and Ω. This way we are able to quantify independently the impact of these two parameters on the incorporation of Mg and Sr into the tests of the planktonic foraminiferal species *Globigerinoides sacculifer* (Brady).

2. Methodology

2.1. Foraminifera collection and culturing

Planktonic, symbiont-bearing foraminifera *G. sacculifer* (Fig. 1A) were collected from surface waters (2–6 m depth), 10 km off the southwest coast of Puerto Rico (17° 54′ 46″N, 66° 58′ 44″W) by scuba diving between April and June 2006. Specimens were brought back to the marine station at Magueyes Island (Department of Marine Sciences, University of Puerto Rico at Mayaguez), La Parguera, Puerto Rico where the culture experiments were setup. Surface seawater for culturing the foraminifera was collected at the dive site and filtered over 0.25 µm polycarbonate membrane filters.

We identified and measured individual foraminifera using an inverted light microscope subsequently transferring the specimens to 120-ml Pyrex bottles with one of two types of filtered seawater namely: 1. unmodified seawater (controls) or, 2. seawater whose

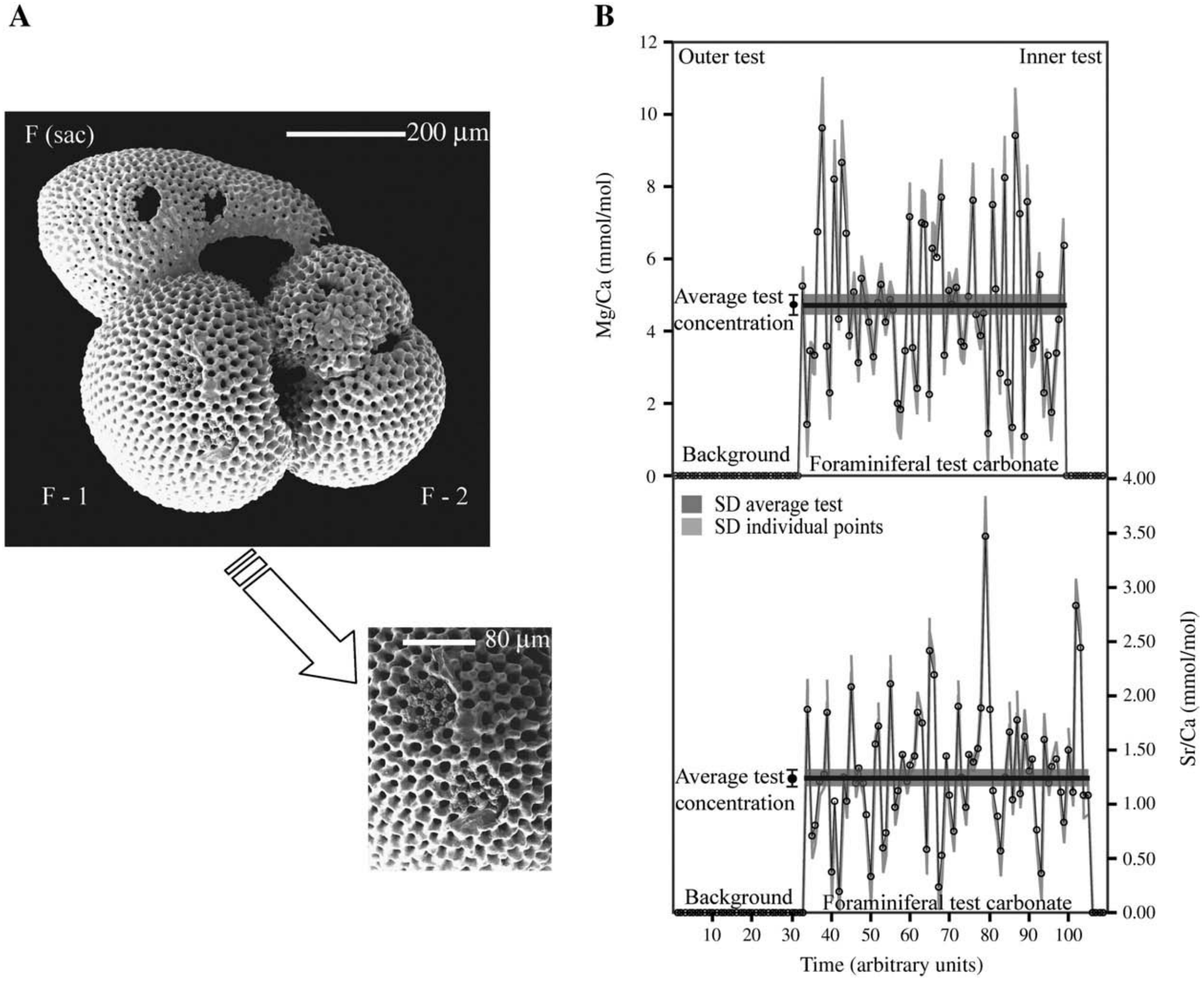

Fig. 1. A. Specimen of *G. sacculifer* (sac-like morphotype) analysed by Laser ablation ICP-MS; F (sac) = final chamber or in most of the cases chamber stage 19; F-1 = penultimate chamber or in most of the cases chamber stage 18; F-2 = third chamber/chamber stage 17 (older than F and F-1) and detail of LA crater; B. Laser ablation profile of foraminiferal calcite.

carbonate chemistry was modified. These bottles were incubated in water baths (0.50 × 1.50 m approx.) and kept at constant temperature (26 °C ± 0.5) and light intensity (12-h light:12-h dark cycle using a light intensity of 353 µE/m^2/s). Specimens were fed every third day with a 1-day-old nauplius of *Artemina salina*, starting on the day of collection. For these experiments, we used values of physical and chemical parameters within the tolerance ranges of *G. sacculifer* (Bijma et al., 1990) and followed the procedures outlined in Bijma et al. (1990) in order to minimize stressful conditions that could potentially affect foraminiferal trace element incorporation.

Specimens were monitored daily and usually underwent gametogenesis (GAM) between 7 and 14 days after collection. Then they were removed from the solutions and rinsed with deionised water. We measured individual chambers only from GAM specimens and archived them for later analysis. Foraminifera that built new chambers in the laboratory were identified by comparing their size at collection with the size of the test after gametogenesis. Foraminifera used for trace metal analyses formed 1 to 3 additional chambers during the incubations. Results presented here are based on analysis of the sac-like morphotype (= *G. sacculifer* sensu stricto (Bijma and Hemleben, 1994)).

According to the previously established correlation between chamber number and shell size of *G. sacculifer* (Hemleben and Bijma, 1994), almost all F-1 chambers (= penultimate) correspond to chamber stage 18 (referred as chambers 18) which corresponds to an average shell size of 545 µm. Consequently, chamber stage 19 refers in most of the cases to sac-like or F (ultimate) chambers.

Mg/Ca and Sr/Ca values used in this study are only from chambers 18 of *G. sacculifer* sensu stricto (sac-like morphotype) since we observed a more consistent relationship between this chamber stage and the carbonate parameters analysed (salinity, Ω and [CO$_3^{2-}$]) (Tables A3 and A4). Chambers corresponding to other life stages (i.e. 19 and 20) are not taken into account in the present study because ontogeny potentially plays an important role in determining their chemical composition (Nürnberg et al., 1996a; Eggins et al., 2003; Erez, 2003).

2.2. Carbonate chemistry of the culture solutions

Culture solutions with three different salinity values (30, 36 and 39) were prepared (Table 1). Carbonate parameters of all solutions were calculated using the CO$_2$sys program (version 01.05, Lewis and Wallace, 1998) and monitored every three days by measuring salinity, total alkalinity and pH (NBS scale) of the solutions (Table 2A). The corresponding Ω values were: 5.25 ± 0.03, 6.16 ± 0.02 and 6.50 ± 0.04. The majority of present-day surface water conditions fall within the limits of these values (Table 1). In order to obtain the target values (30 and 39), salinity was altered by adding deionised water or through evaporation by sub-boiling (at ~40 °C).

In order to reach the target Ω values, we varied the [CO$_3^{2-}$] of the seawater. Filtered seawater was modified to obtain a range of [CO$_3^{2-}$] by maintaining a constant dissolved inorganic carbon concentration – DIC – (closed system) and adjusting alkalinity with NaOH (1 M) or HCl (1 M). Under these conditions, both pH and [CO$_3^{2-}$] of the seawater increase or decrease with alkalinity.

Table 1
Overview of experiments with salinity and carbonate saturation state values.

Ω[a]/salinity	30	36	39
5.25 ± 0.03	Control 1		
6.16 ± 0.02		Control 2	
6.50 ± 0.04			Control 3

[a] Saturation values correspond to the values of the natural salinities (controls).

Table 2A
Experimental conditions.[a]

Experiment no.	Salinity	Alkalinity (µmol/kg)	pH (NBS scale)	Mean [CO$_3^{-2}$] (µmol/kg)	DIC (µmol/kg)	Ωc
1 (control 1)	30	2001 ± 29	8.29	208 ± 2	1702	5.2
2	36	2358 ± 13	8.17	222 ± 2	2046	5.3
3	39	2519 ± 26	8.10	224 ± 3	2210	5.2
4	30	2072 ± 32	8.37	246	1719	6.2
5 (control 2)	36	2416	8.24	258	2053	6.2
6	39	2558 ± 38	8.19	264 ± 2	2188	6.1
7	30	2113 ± 34	8.40	262 ± 3	1739	6.6
8	36	2440 ± 37	8.26	270 ± 2	2059	6.5
9 (control 3)	39	2603 ± 4	8.21	278 ± 2	2216	6.5

[a] All experiments were carried out at $T = 26$ °C and a light intensity = 353 µE/m^2/s. Mean [CO$_3^{-2}$] and DIC were calculated from alkalinity and pH measurements using the program CO2sys (Lewis and Wallace, version 01.05), with the CO$_2$ constants of Mehrbach et al. (1973) refit by Dickson and Millero (1987), and K$_{SO4}$ from Dickson (1990). Ωc refers to saturation state for calcite.

Table 2B
Summary of mean Mg/Ca and Sr/Ca ratios from chamber stage no. 18 in cultured planktonic foraminifera.

Experiment no.[a]	n[b]	Mg/Ca (mmol/mol)	Standard deviation of Mg/Ca	n[b]	Sr/Ca (mmol/mol)	Standard deviation of Sr/Ca
1 (control 1)	3	4.42	1.30	3	1.32	0.05
2	2	4.42	1.01	2	1.30	0.10
3	4	5.61	1.28	4	1.44	0.15
5 (control 2)	6	5.16	0.63	7	1.35	0.07
6	2	5.86	0.52	2	1.41	0.01
7	2	3.50	0.89	2	1.38	0.04
8	2	4.40	0.34	2	1.32	0.11
9 (control 3)	5	5.03	1.34	4	1.34	0.10

[a] Experiment No. 4 is not reported due to lack of analysis.

[b] n refers to the number of specimens used to calculate the mean and the standard deviation.

Seawater samples from control experiments (Ω not modified) were collected and brought to the home laboratory at Utrecht University to analyse DIC. This carbonate parameter was measured using a Total Organic Carbon Analyzer (Shimadzu, Model TOC-5050A). DIC results from these analyses and DIC values calculated via the CO$_2$sys program were not significantly different (Table A1).

2.3. Sample preparation and analysis

Specimens that grew new chambers were placed in a sodium hypochlorite bath (NaClO 5 %) for 20 min in order to remove any remaining organic material. The specimens were rinsed 3 times with deionised water afterward, carefully pipetting of the supernatant. Mg/Ca ratios were measured by laser ablation inductively coupled plasma-mass spectrometry (LA-ICP-MS, Micromass Platform). This technique allows us to measure the trace elemental concentrations of individual chambers from single specimens several times.

Individual foraminiferal chambers were ablated using a 193 nm laser (GeoLas 200Q Excimer) in a helium flushed ablation chamber which was coupled to the ICP-MS. A deep ultra violet-wavelength laser was used in order to guarantee the reproducibility of the ablation of the fragile tests. This type of laser was employed since carbonates do not absorb laser radiation well at higher wavelengths. Pulse repetition rate was set at 6 Hz with an energy density at the sample surface of 1 J/cm^2. Ablation craters were 80 µm in diameter and the ablated calcite was analysed with respect to time (Fig. 1A and B). Mg/Ca and Sr/Ca ratios correspond to the average value of single measurements during a laser ablation ICP-MS analysis (Fig. 1B). Calibration was performed against U.S. National Institute of Standards and Technology (NIST) SRM 610 glass with ^{44}Ca as an internal standard. Using calcium as an internal standard is ideal as this element is present at a constant concentration of 40%. This also allows direct comparison with the more traditional wet chemical analyses (Reichart et al., 2003). A collision and reaction cell was used to give improved results by reducing spectral interferences on the minor isotopes of Ca (^{42}Ca, ^{43}Ca, and ^{44}Ca). The glass standard SRM 610 was measured with a higher energy density (4 J/cm^2) than the calcite samples.

To check whether using different ablation energy biases the analyses, a matrix matched standard was included. This standard is an uncommonly homogeneous calcite crystal (Icelandspar) that was analysed by LA-ICP-MS. Subsamples taken from this calcite crystal were also dissolved in ultra clean HNO_3 and subsequently analysed using an ICP-AES (Spectro CIROS CCD). A comparison between these two analyses shows that, although a different energy density was used for the glass and calcite standard, Mg/Ca and Sr/Ca values are statistically identical (Table A2). Based on repetitive analyses of the calcite standard throughout the analytical period, relative precission of the LA-ICP-MS analyses for Mg and Sr was around 3% and 3.6% respectively. Monitoring simultaneously ^{42}Ca, ^{43}Ca and ^{44}Ca showed isotopic ratios expected on the basis of their natural relative abundances. Normalization to an international standard is also recommended for the analysis of Mg/Ca and Sr/Ca ratios apart from the use of SRM 610 glass (NIST) and homogeneous calcite crystal (Icelandspar) standards. This is suggested in order to avoid methodological variations that could bias the response of these foraminiferal elements to seawater physical and chemical parameters.

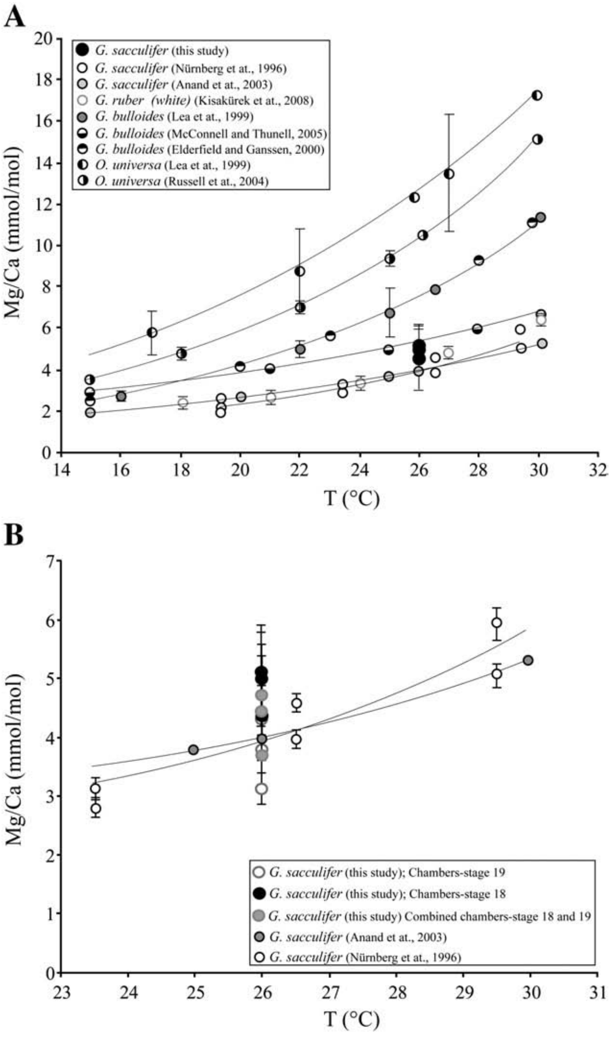

Fig. 2. A. Mg/Ca ratios of diverse planktonic foraminiferal shells from laboratory cultures and sediment traps vs. temperature. Salinity values vary from 30 to 39; B. Mg/Ca of *G. sacculifer* from three different studies (data from present study correspond to chambers 18 and 19 from control experiments in which Ω was not modified; experiments 1, 5 and 9 from Tables 2A and B).

Accuracy for each individual analysis was calculated using the Glitter computer program, which we also used to calculate elemental concentrations (Glitter, LA-ICP/MS Data Reduction and Display, GEMOC, CSIRO, Maquarie Research Limited, 1999–2000). The intervals of the acquired data used to calculate concentrations were selected avoiding sections with high Al and/or Pb counts. Although the foraminifera were never in contact with sediments as a source for contamination, this ensures that an (unknown) phase does not introduce errors in the trace metal analyses. An analysis of variance (ANOVA) was used in order to determine how well a regression model would predict Mg/Ca and Sr/Ca variability under the conditions analysed.

3. Results

Overall the test carbonate of *G. sacculifer* has lower Mg/Ca ratios (this study; Nürnberg et al., 1996b; Anand et al., 2003) compared to *O. universa* and *Globigerinoides bulloides* (Lea et al., 1999; Elderfield and Ganssen, 2000; McConnell and Thunell, 2005; Russell et al., 2004). Mg/Ca ratios in the tests of *G. sacculifer* and *Globigerinoides ruber* (white) (Kısakürek et al., 2008) show similar values (Fig. 2A). Our results correspond to a temperature of 26 °C and variable salinities, ranging from 30 to 39. Mg/Ca ratios measured on chambers 18 of *G. sacculifer* show slightly higher values compared to previous studies (Nürnberg et al., 1996b; Anand et al., 2003) (Fig. 2A). However, the combined Mg/Ca values of chambers 18 and 19 correspond well to previous correlations between seawater temperatures and foraminiferal Mg incorporation (Fig. 2B). The observed variability between Mg/Ca values of individual specimens grown at constant environmental conditions is larger than the analytical precision of the LA-ICP-MS analyses (Table A4).

3.1. Effect of salinity on Mg/Ca and Sr/Ca ratios

We combined the Mg/Ca ratio data of each salinity experiment (30, 36 and 39), regardless of their Ω value (Tables 1 and 2A and B; Fig. 3), in order to improve the statistical basis of the Mg/Ca-salinity correlation. The average Mg/Ca values of *G. sacculifer* increase at higher salinities despite the relatively large interindividual variability (Figs. 3 and 4; Table A4). The Mg/Ca ratios of the foraminiferal tests grown in the experiments show a slightly larger response to changes in salinity than previously observed by Nürnberg et al. (1996a) (values only from GAM specimens and chambers built during culture; in Table 1 data from Figs. 3 and 6) (Fig. 4A). The changes in Mg/Ca ratios due to a salinity increase observed in the present study are smaller than the ones found in the planktonic foraminifera *O. universa* and *G. ruber* (white) (Lea et al., 1999; Kısakürek et al., 2008, respectively) (Fig. 4A). The incorporation of Sr increases slightly with salinity (Figs. 3D and E and 4B). In contrast, existing calibrations by Lea et al. (1999) and Kısakürek et al. (2008) showed that in cultured *O. universa* and *G. ruber*

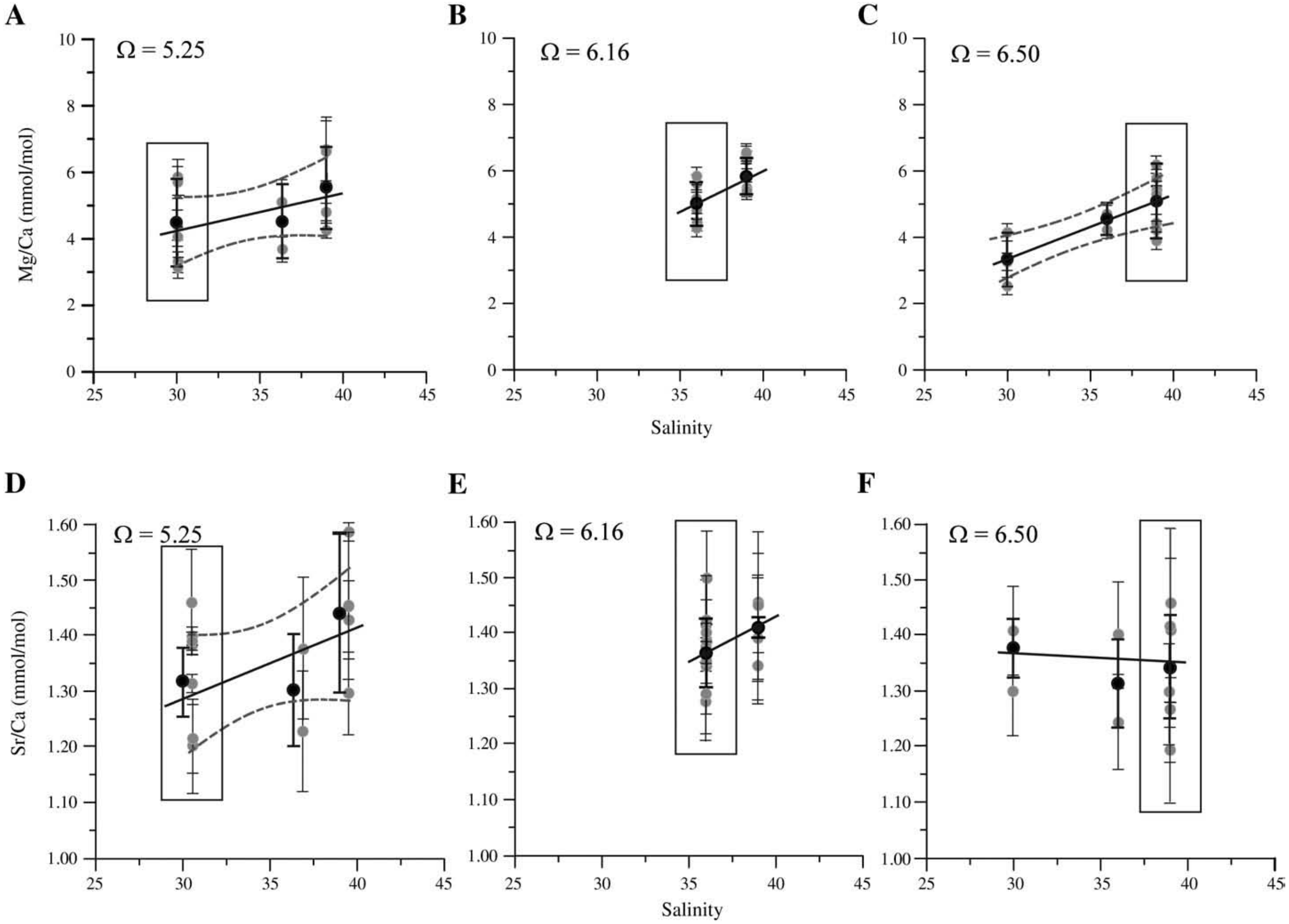

Fig. 3. Foraminiferal Mg/Ca and Sr/Ca ratios at three different salinity values and a constant *Ω*. Mg/Ca: A. *Ω* = 5.25 ± 0.03 (from Tables 1 and 2A and B experiment nos. 1, 2 and 3); B. *Ω* = 6.16 ± 0.02 (from Tables 1 and 2A and B experiment nos. 4, 5 and 6) and, C. *Ω* = 6.50 ± 0.04 (from Tables 1 and 2A and B experiment nos. 7, 8 and 9); Sr/Ca ratios from D to F, figures have the same correspondence as Mg/Ca ratios. Closed circles correspond to average Mg/Ca and Sr/Ca ratios of chamber 18; grey circles correspond to individual laser ablation analyses. Control experiments are marked with a square (from Tables 1 and 2A and B experiment nos. 1, 5 and 9). The 95% confidence limits of the curve fit are shown by dashed lines.

(white) Sr incorporation increases more rapidly with salinity changes (Fig. 4B).

3.2. Effect of calcium carbonate saturation state (*Ω*) on Mg/Ca and Sr/Ca ratios

The incorporation of Mg into foraminiferal test carbonate increases at higher salinity values when *Ω* is kept constant (Fig. 3A, B and C). In contrast, no consistent relationship is observed between Sr/Ca ratios and salinity when *Ω* is kept constant at three different salinity values (Fig. 3D, E and F). When plotting foraminiferal Mg/Ca ratios versus *Ω* and $[CO_3^{2-}]$ no correlation is observed (Fig. 5A and B, respectively). In comparison, there is a clear effect of *Ω* and $[CO_3^{2-}]$ on foraminiferal Sr/Ca when salinity is kept constant at 36 (0.04 mmol/mol per *Ω* unit and 0.10 mmol/mol per 100 µmol/kg rise in $[CO_3^{2-}]$ (Table 3; Fig. 5C and D).

4. Discussion

Several planktonic foraminiferal Mg/Ca–temperature calibrations have been published for different species during the last decade (Fig. 2A) (Nürnberg et al., 1996b; Lea et al., 1999; Elderfield and Ganssen, 2000; Anand et al., 2003; Russell et al., 2004; McConnell and Thunell, 2005; Kısakürek et al., 2008). All these calibrations show an increase in the incorporation of Mg into foraminiferal test carbonate with increasing temperatures. Whereas most calibrations are based on traditional wet chemical analyses of complete dissolved foraminifera we used laser ablation ICP-MS for analysing the trace elemental composition of single foraminiferal test chambers. We observe that Mg/Ca values measured on chambers 18 are higher whereas Mg/Ca values from chambers 19 are lower in comparison to existing *G. sacculifer* calibrations (Nürnberg et al., 1996b; Anand et al., 2003) (Fig. 2B). Average Mg/Ca values, based on combining the data from chambers 18 and 19, are consistent with the existing whole-foraminifera calibrations (Fig. 2B). This implies that differences in trace element composition between chambers can significantly affect trace element/Ca calibrations. Such an impact of intra-chamber heterogeneity of foraminiferal Mg incorporation on reconstructions of past seawater temperatures was previously suggested by Eggins et al. (2003), Sadekov et al. (2005), Hintz et al. (2006) and Sadekov et al. (2008). Nevertheless results from traditional Mg/Ca analyses (Anand et al., 2003) and laser ablation ICP-MS (this study) suggest similar foraminiferal Mg/Ca ratios for *G. sacculifer* at the same temperature

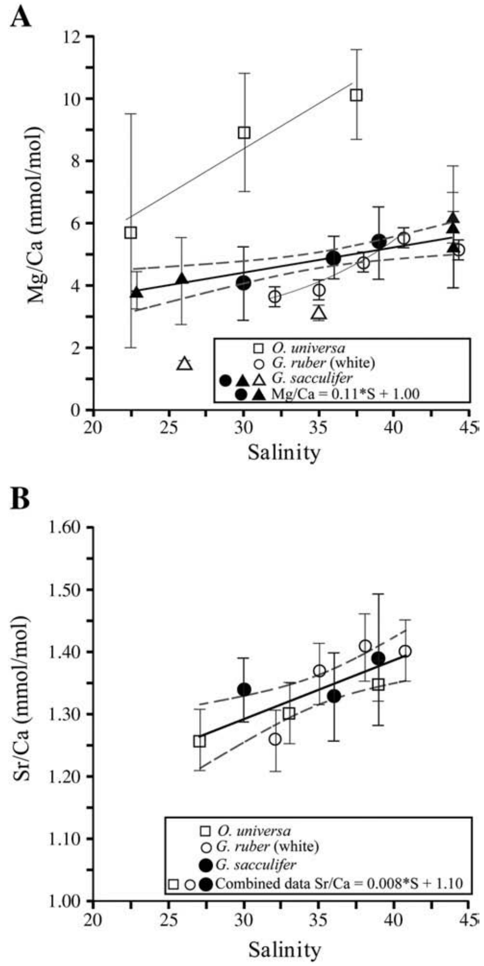

Fig. 4. A. Foraminiferal Mg/Ca ratio vs. salinity; closed circles correspond to data from present study; open and closed triangles correspond to NO GAM and GAM specimens respectively of *G. sacculifer* obtained from Nürnberg et al. (1996a) and Nürnberg's pers. comm; open circles correspond to data from Kısakürek et al. (2008) (*G. ruber* — white); open squares correspond to data from Lea et al. (1999) (*O. universa*); B. Foraminiferal Sr/Ca ratio vs. salinity; symbols have the same correspondence as in panel A. Data from present study correspond to Mg/Ca and Sr/Ca averages of every salinity experiment regardless of the Ω value (Tables 2A and B). Error bars are standard deviations based on the spread and number of analyses per experiment (applied to all figures). The 95% confidence limits of the curve fit are shown by dashed lines.

(Fig. 2B). Moreover, calibrations based on culturing experiments (this study and Nürnberg et al., 1996b) and those using core-top samples (Anand et al., 2003) are comparable (Fig. 2B). This implies that laser ablation ICP-MS as well as culture experiments are suitable approaches for investigating Mg incorporation in foraminiferal carbonate.

4.1. Effect of salinity on Mg/Ca and Sr/Ca ratios

The observed changes in planktonic foraminiferal Mg incorporation in response to different salinity values (Fig. 3) can be due to a direct effect or to the related effect of salinity changes on the carbonate parameters. When we compare the results from the control experiments to those in which Ω was kept constant by modifying the carbonate chemistry over a salinity range (Table 2B; Fig. 3A to C), we observe a similar positive correlation between Mg/Ca ratios and salinity. We also observe there is no significant effect of Ω within the range analysed (5.25–6.50) on foraminiferal Mg incorporation ($p > 0.05$) while a significant effect of salinity is found ($p < 0.05$). These results imply that changes in Mg/Ca ratios of *G. sacculifer* are mainly caused by salinity itself rather than Ω and the $[CO_3^{2-}]$ within the interval considered (salinity between 30 and 39). Therefore, the results discussed in the following section are based on the combined data of every salinity experiment (30, 36 and 39) regardless of their Ω value (Tables 2A and B; Fig. 4A).

When plotting the data of Nürnberg et al. (1996a) together with our data, we observe that the Mg/Ca values of NO GAM specimens are below the general trend (points at salinities 26 and 35; Fig. 4A). On the other hand, the Mg/Ca ratios at salinity “35” come from chambers that were grown in the natural environment which considerably varied in salinity (from 31.2 to 36) and temperature (from 26.2 to 29.7 °C). This might explain the overall lower values at this salinity. Therefore, we have used the data of Nürnberg et al. (1996a) coming from GAM specimens and chambers grown in controlled culture experiments.

The increase in Mg/Ca ratios observed at higher salinities in our results (Fig. 4A) is slightly larger than the one observed in a previous salinity calibration for *G. sacculifer* (Nürnberg et al., 1996a; from Table 1 data belonging to Figs. 3 and 6). However, the 95% confidence interval from our data encompasses the Mg/Ca-salinity correlation of Nürnberg et al. (1996a). This indicates that the Mg incorporation observed in these two studies (present study and Nürnberg et al., 1996a) is – at least statistically – comparable (Fig. 4A). When we combine Mg/Ca ratios from the present study with the data from Nürnberg et al. (1996a), the resulting Mg/Ca-salinity relationship indicates that salinity only accounts for 30% of the variation in Mg/Ca ratios ($r^2 = 0.30$; Table 3). This means that 70% of the variation in Mg/Ca ratios cannot be explained by salinity. Therefore, there must be other variables that have an influence too. We also observe that the regression model used provides a solid statistical basis to quantify the effect of salinity on Mg/Ca ($p < 0.05$; Table 3). From this Mg/Ca-salinity relationship, based on the combined data of this study and Nürnberg et al. (1996a), an increase of 0.11 mmol/mol of Mg/Ca per salinity unit is estimated (Table 3) which corresponds to an increase of 2.3% in Mg/Ca per salinity unit. This Mg/Ca response to salinity is slightly smaller than the ones found in the planktonic foraminifera *O. universa* and *G. ruber*, where Mg/Ca ratio dependencies on salinity of 4 and 5% respectively, are reported (Lea et al., 1999; Kısakürek et al., 2008; Fig. 4A).

The most important factor influencing Mg/Ca ratio of inorganic calcite is the Mg/Ca ratio of the parent solution from which calcite precipitates (Mucci and Morse, 1983). The much depleted Mg/Ca ratios of foraminiferal calcite, therefore, suggest a strongly modified internal calcification pool. Although increasing salinity of the seawater does not change the Mg/Ca ratio of the parent solution, absolute concentrations of these two elements do change. According to Bentov and Erez (2006), there is an inward driving force for Mg^{2+} from the extracellular environment into the cell. This has only been established in eukaryotic cells but it is probable that this is not significantly different in foraminifera. In order to keep a low intracellular Mg^{2+} activity, as other eukaryotes do, foraminifera may use different cellular mechanisms such as Mg protein-based transport systems, cellular buffering and/or sequestration within cellular organelles (Bentov and Erez, 2006). At higher salinities the parent solution from which calcite precipitates has a higher Mg^{2+} concentration. This parent solution is kept in an extracellular space from which the cell will extract Mg^{2+} to the cytoplasm by using Mg channels and pumps (Erez, 2003). We assume that the amount of Mg^{2+} extracted from this parent solution does not vary and the foraminifer continues to use the same amount of energy in order to lower the Mg^{2+} of the parent solution. If this is the case, higher Mg^{2+} in the parent solution would be reflected in the shell composition (Segev and Erez, 2006), which is in line with the

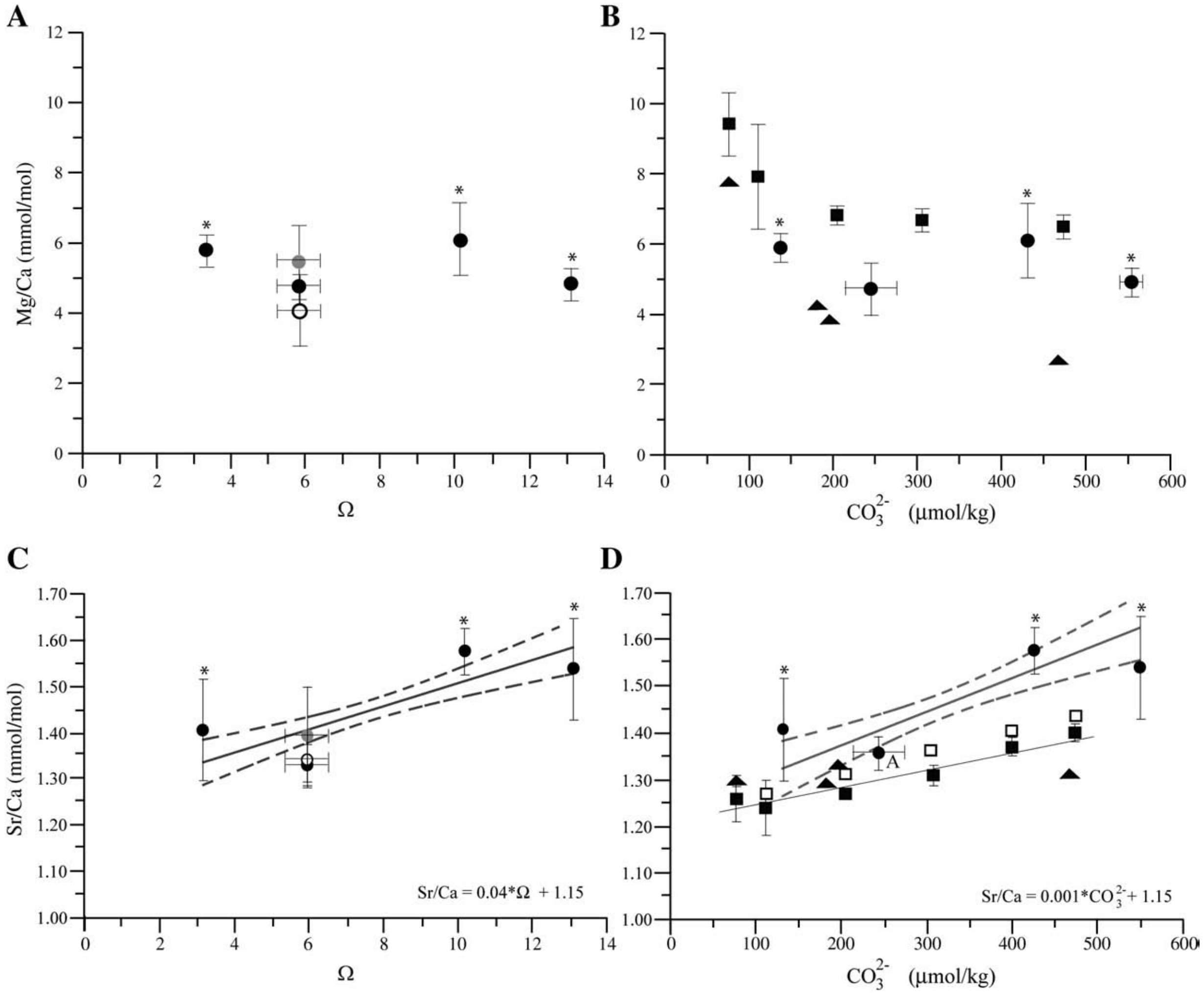

Fig. 5. A and B. Foraminiferal Mg/Ca ratio at different Ω and [CO$_3^{2-}$] values respectively (constant temperature of 26 °C); C and D. Foraminiferal Sr/Ca ratio at different Ω and [CO$_3^{2-}$] values (constant temperature of 26 °C). Open, closed and grey circles correspond to average Mg/Ca and Sr/Ca ratios of salinities 30, 36 and 39 respectively from the present study (Table 2B); closed squares correspond to data from Russell et al. (2004) (*O. universa*); open squares correspond to data from Russell et al. (2004) (*O. universa*) at the same temperature and salinity as in our study (26 °C and 36 salinity units); closed triangles correspond to data from Russell et al. (2004) (*G. bulloides*). The 95% confidence limit of the curve fit is shown by dashed lines. ^{A}Average of Sr/Ca of salinities 30, 36 and 39; * data from da Rocha et al. in prep (Table A6).

observations from the present study (Figs. 3A, B and C and 4A). Higher amounts of extracted Mg^{2+} – coming from the parent solution into the foraminifer cytoplasm – would mean an increase in the energy required to extrude this excess of Mg^{2+} from the cell (Zeebe and Sanyal, 2002). This strategy would not be as cost-effective as extracting the same [Mg^{2+}] regardless of the element concentration of the parent solution. This assumption is in line with inorganic calcite precipitation studies by Zeebe and Sanyal (2002) in which it is shown that, in order to initiate the calcification process at biogenic precipitation rates, the best cost-effective strategy is H$^+$ removal from the calcifying area. Therefore, the foraminifer would not change the rate at which Mg^{2+} is extruded from the cell in order to calcify.

The mechanisms responsible for the influence of salinity on Mg/Ca ratios are not well understood. Therefore, the difference found in the Mg/Ca ratio response to salinity changes between planktonic foraminiferal species cannot be explained with confidence (Fig. 4A). We suggest it might be due to the specific effect that a salinity increase has on the cellular Mg channels and pumps. In the case of *O. universa* and *G. ruber* (white), the biological pumps in charge of extracting Mg^{2+} from the parent solution could be less tolerant and therefore, less efficient at higher salinities than the Mg pumps of *G. sacculifer*. This would cause higher than usual Mg^{2+} in the parent solution that would be reflected in the biogenic calcite. We also suggest that the different Mg/Ca-salinity correlations found in planktonic foraminifera might be caused by the difference in salinity dependence between Mg/Ca ratios of GAM calcite and Mg/Ca values of NO GAM calcite. The Mg/Ca ratios from chamber stage 18 (F-1 chambers) have a significant correlation with salinity (Table 3), while Mg/Ca ratios from chamber stage 19 (sac-like/F chambers — most of it GAM calcite) do not show significant correlation with this parameter (data not included). This indicates that the response of Mg/Ca ratios from chambers 18 to salinity might be slightly biased by the presence of GAM calcite. As a result, the differences in Mg/Ca response to salinity between planktonic species with and without GAM calcite become larger (e.g. Mg/Ca-salinity dependencies of 2.3% and 5% per salinity unit for *G. sacculifer* and *G. ruber*, respectively).

There is no consistent relationship between Sr/Ca ratios and salinity within the salinity range analysed — 30 to 39 (Table 2B, Figs. 3D, E and F and 4A). We also observe that there is no significant effect of Ω within the range analysed (5.25–6.50) on foraminiferal Sr

Table 3
Summary of Mg/Ca and Sr/Ca relationships with salinity, Ω and $[CO_3^{2-}]$.[a]

	Regression	*n*	r^2	*F*	*p*	Experimentally determined responses (in mmol/mol) per salinity unit
Salinity vs. Mg/Ca (data from this study and Nürnberg et al. 1996a,b)	$Mg/Ca = 0.11 * S + 1.00$	31	0.30	10.43	0.003	0.11
	Regression	*n*	r^2	*F*	*p*	Experimentally determined responses (in mmol/mol) per 100 µmol/kg of $[CO_3^{2-}]$ and per Ω unit
$[CO_3^{2-}]$ vs. Sr/Ca	$Sr/Ca = 0.001 * [CO_3^{2-}] + 1.15$	29	0.40	18.30	<0.001	0.10
Ω vs. Sr/Ca	$Sr/Ca = 0.04 * \Omega + 1.15$	29	0.40	18.04	<0.001	0.04

[a] Temperature was kept constant at 26 °C. All regressions and statistics are based on individual analyses (not means as plotted in the figures); "*n*" refers to the number of laser ablation analyses used to calculate the mean and the standard deviation. Only relationships that are statistically significant ($p<0.001$; $p<0.05$) are included, resulting in the exclusion of Mg/Ca vs. salinity at constant Ω values and Sr/Ca vs. salinity at constant Ω values; *p* indicates that there is less than a 0.1% or 5% chance that the high *F*-ratios obtained would happen by chance alone. This means that a regression model overall predicts Mg/Ca and Sr/Ca variability significantly well under the conditions analysed.

incorporation ($p>0.05$). Therefore, the results discussed in the following section are based on the combined data of every salinity experiment, regardless of their Ω values (Tables 2A and B; Fig. 4B). A slight increase in Sr incorporation in the test carbonate of *G. sacculifer* is observed at higher salinity values (Fig. 4B). Lea et al. (1999) and Kısakürek et al. (2008) also reported a positive correlation between Sr/Ca and salinity for the planktonic foraminifera *O. universa* and *G. ruber* (white) respectively.

The 95% confidence interval from our data encompasses the previous Sr/Ca-salinity correlations of the two other species of planktonic foraminifera which means that Sr incorporation in these three planktonic species is – at least statistically – comparable and thus potentially reflects similar processes related to salinity increases. When combining Sr/Ca ratios from the present study with the data from Lea et al. (1999) and Kısakürek et al. (2008) (Fig. 4B), an increase of 0.008 mmol/mol of Sr/Ca per salinity unit is observed. According to Lorens (1981), Nehrke et al. (2007) and Tang et al. (2008), higher calcite Sr/Ca ratios are mainly associated with higher growth rates in inorganic calcite which is also observed in biogenic calcite (Kısakürek et al., 2008). Results from inorganic experiments (Zuddas and Mucci, 1998) furthermore indicate a positive linear correlation between calcite precipitation rate and ionic strength of the solution. Therefore, the observed increase in Sr/Ca values at higher salinities (Fig. 4B) might primarily result from an increased in Ω.

4.2. Effect of calcium carbonate saturation state (Ω) on Mg/Ca and Sr/Ca ratios

There is no clear effect of either Ω or $[CO_3^{2-}]$ on foraminiferal Mg incorporation when salinity is kept constant at 36 (Fig. 5A and B). This might be due to the selected range of Ω and $[CO_3^{2-}]$ values, which was rather limited (Table 2A). Russell et al. (2004) showed that the Mg/Ca-$[CO_3^{2-}]$ relationship in planktonic foraminifera tends to have a relatively constant value above CO_3^{2-} concentrations of 200 µmol/kg (Fig. 5B), while below this concentration it rapidly increases. Most of the Mg/Ca ratio data for *G. sacculifer* fall in the interval where no significant variations are expected (Fig. 5B).

Omega has a stronger effect on Sr incorporation in *G. sacculifer* than salinity with an increase of 0.04 mmol/mol of Sr/Ca per Ω unit for a constant salinity of 36 compared to 0.008 mmol/mol of Sr/Ca per salinity unit (Table 3; Figs. 4B and 5C). This Sr/Ca–Ω relationship indicates that Ω accounts for almost half of the variation in Sr/Ca ratios ($r^2 = 0.40$; Table 3). We also observe that the regression model predicts a statistically significant Sr/Ca variability due to changes in Ω ($p<0.001$; Table 3). In inorganic and biogenic calcite, the Sr/Ca ratio is known to increase with growth rate (Lorens, 1981; Nehrke et al., 2007; Tang et al., 2008; Kısakürek et al., 2008), and a higher $[CO_3^{2-}]$ increases foraminiferal shell weight and possibly calcification rates (Bijma et al., 1999). Therefore, the observed correlation is possibly linked to the calcification rate.

We observe a positive correlation between Sr/Ca ratios and $[CO_3^{2-}]$ (or pH) in the planktonic species *G. sacculifer* (this study) and *O. universa* (Russell et al., 2004) (Fig. 5D). However, the Sr/Ca sensitivities found in these two planktonic foraminifera are different (0.10 mmol/mol of Sr/Ca per 100 µmol/kg rise in $[CO_3^{2-}]$ for *G. sacculifer*, and 0.04 mmol/mol of Sr/Ca per 100 µmol/kg rise in $[CO_3^{2-}]$ for *O. universa*). *G. sacculifer* Sr/Ca values are also overall higher than those of *O. universa* (Fig. 5D). This interspecific variability in Sr/Ca is too large to be caused by differences in salinity and temperature between culturing experiments alone (Fig. 5D; cf. Russell et al. (2004) for temperature effect and Lea et al. (1999) for salinity effect on Sr/Ca in *O. universa*). Therefore, the response of Sr/Ca to increasing $[CO_3^{2-}]$ appears to be species-specific. According to Anderson and Faber (1984), Lea et al. (1995) and Bijma et al. (1999), *G. sacculifer* has a higher calcification rate than *O. universa*. The rates at which these two species calcify might thus be responsible for both the difference in Sr/Ca ratios and the different response of foraminiferal Sr incorporation to $[CO_3^{2-}]$ changes (Fig. 5D). The presence and size of internal DIC and calcium pools in the cytoplasm of *G. sacculifer* might also affect Sr incorporation into the test calcite and could also be responsible for the interspecies variability in Sr/Ca ratios. However, precipitation of trace elements from these internal reservoirs and their chemical composition are difficult to study (Erez, 2003).

Differences in the response of Sr/Ca to $[CO_3^{2-}]$ (or pH) between *G. sacculifer* and *G. bulloides* (Fig. 5D) might be related to the impact of photosynthetic symbionts in *G. sacculifer* on the calcification rate. According to the foraminiferal biomineralization model proposed by Wolf-Gladrow et al. (1999), photosynthetic uptake of CO_2 in the foraminifer microenvironment leads to an increase in pH and $[CO_3^{2-}]$, which potentially contributes to overall higher calcification rates (Bijma et al., 1999; Lea et al., 1999).

4.3. Paleoceanographic implications

Combining present and published data, we infer that a salinity increase of 4 units is equivalent to a 1 °C temperature increase, using the Mg/Ca ratio — temperature calibration from Nürnberg et al. (1996b): $Mg/Ca = 0.3948 \exp(0.0888 * T)$. Although smaller than the effect of sea surface temperature (SST), salinity has a limited though significant impact on planktonic foraminiferal Mg incorporation, sometimes with important consequences for the interpretation of paleorecords (e.g. Groeneveld et al., 2008). According to Groeneveld et al. (2008), salinity variations of up to three units, based on the planktonic $\delta^{18}O$ record during the latest Miocene/early Pliocene (5.6–3.9 Ma), would result in an overestimation of foraminiferal Mg/Ca ratios of ~21%. This offset is based on the original Mg/Ca-salinity calibration of Nürnberg et al. (1996a,b) using both GAM and NO GAM specimens (Table 1 data belonging to Fig. 6). Our salinity calibration, based on culture experiments of GAM specimens of *G. sacculifer*, suggests a more modest overestimation of Mg/Ca ratios of ~7%. This implies that, although the overall conclusion remains the same, the Caribbean temperature increase after 4.5 Ma was probably somewhat larger.

Although the impact of salinity on Mg/Ca ratios is limited in today's ocean, glacial–interglacial changes in salinity potentially offset temperature reconstructions. Whereas the effect of ice volume alone is about 1.2 salinity units, the drier tropical climate regionally impacted salinity as well. In the tropical western Atlantic Ocean, for instance, sea surface salinity changed between 1 and 3 units approximately from the Last Glacial Maximum (LGM) to the Holocene (Toledo, Costa, and Pivel, 2007; Weldeab et al., 2006). When comparing these salinity changes with reconstructed equatorial Atlantic SSTs during the LGM (3 °C cooler than SSTs during the Holocene) (Toledo et al., 2007; Weldeab et al., 2006), we find that 25% of this temperature decrease might actually be caused by salinity. According to our results, an increase of 4 salinity units is equivalent to about 1 °C warming, in terms of Mg/Ca ratios. Consequently, a change of 3 salinity units – during the LGM-Holocene period – would cause an offset of 0.75 °C, which equals 25% of the total $\Delta SST_{Mg/Ca}$ observed between Holocene and LGM sea surface temperatures. We consider this a significant offset that should be taken into account when calculating the $SST_{Mg/Ca}$ of the tropical Atlantic Ocean during the LGM.

The positive correlation between Ω ($[CO_3^{2-}]$ and thus pH) and Sr/Ca of symbiont-bearing planktonic species such as *G. sacculifer* (present study) and *O. universa* (Russell et al., 2004) could potentially bias data of past changes in seawater Sr/Ca (Stoll et al., 1999). The observed changes in foraminiferal Sr/Ca ratios coinciding with glacial–interglacial cycles are partly attributed to past changes in seawater Sr/Ca ratios and partly to changes in pCO_2. Our results as well as the culture study by Russell et al. (2004) demonstrate that changes in Ω ($[CO_3^{2-}]$ and hence pH) have an important impact on foraminiferal Sr incorporation: almost half of the Sr/Ca variability observed in the present study can be explained by changes in Ω ($[CO_3^{2-}]$ and thus pH) (Fig. 5C and D; Table 3). Reconstructions of glacial–interglacial changes in seawater $[CO_3^{2-}]$, based on $\delta^{13}C$ records of *G. sacculifer* and *G. ruber* (Spero et al., 1999), indicate a broad variation in $[CO_3^{2-}]$ of about 50 µmol/kg during the Last Glacial Maximum (LGM). According to the present study, this change in seawater $[CO_3^{2-}]$ would correspond to a 0.05 mmol/mol change in foraminiferal Sr/Ca ratios from *G. sacculifer*. Our correlation between Sr/Ca and seawater $[CO_3^{2-}]$ could potentially fully explain the observed glacial to interglacial changes in Sr/Ca ratios of *G. sacculifer* (Stoll et al., 1999), without changing seawater Sr/Ca ratios. Therefore, when studying foraminiferal Sr/Ca records on longer timescales, changes in ocean carbonate chemistry must be considered in addition to variations in seawater Sr/Ca.

5. Conclusions

The present study corroborates the presence of a salinity effect on Mg incorporation in *G. sacculifer*, although it is considerably smaller than the temperature effect (only 30% of the variation in Mg/Ca ratios in our experiments is due to salinity). The Mg/Ca ratios of *G. sacculifer* – this study and Nürnberg et al. (1996a) – show a 0.11 mmol/mol increase per salinity unit, which implies a salinity increase of 4 units corresponding to a 1 °C temperature increase. This effect of salinity on Mg incorporation is relatively modest. However, when using Mg/Ca-based temperatures in combination with foraminiferal $\delta^{18}O$ to calculate salinity, it cannot be neglected. Due to changes in salinity, on a glacial–interglacial timescale tropical $\Delta SST_{Mg/Ca}$ can be offset by up to 1 °C which corresponds to a 25% bias in the total $\Delta SST_{Mg/Ca}$. At constant temperature of 26 °C, our results suggest that salinity controls the Mg incorporation, as no impact of carbonate chemistry was observed within the range of Ω and $[CO_3^{2-}]$ analysed ($\Omega = 5.25$ to 6.50; $[CO_3^{2-}] = 218$ to 270 µmol/kg). Seawater Ω ($[CO_3^{2-}]$ and thus pH) appears to be the main control on shell Sr incorporation (0.10 mmol/mol per 100 µmol/kg rise in $[CO_3^{2-}]$), resulting in an indirect salinity dependence of Sr/Ca of 0.008 mmol/mol per salinity unit.

Calcite precipitation and growth rates of planktonic foraminifera *G. sacculifer* might be influenced by changes in environmental conditions, processes that may be reflected in the Sr composition of their carbonate test. Variations in the environment of calcification such as changes in carbonate chemistry of the surrounding seawater can also potentially offset past reconstructions of glacial–interglacial seawater Sr/Ca ratios. This might be caused by the influence of Ω and the $[CO_3^{2-}]$ on foraminiferal Sr incorporation (0.04 mmol/mol per Ω unit and 0.10 mmol/mol per 100 µmol/kg rise in $[CO_3^{2-}]$; for the planktonic species *G. sacculifer*).

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Appendix A

Table A1
DIC values obtained by two methods.

Controls (salinity)	DIC[a] analyzer	SD[b]	DIC CO_2sys[a] program	SD[b]	RSD (%)[c]
30	1772	2	1702	28	1.4
36	2099	10	2053	36	0.8
39	2191	4	2216	6	0.4

[a] Values in µmol/kg.
[b] Standard deviation.
[c] Relative Standard Deviation = absolute value of the coefficient of variation expressed as a percentage. It quantifies the precision of the measurements (a percentage lower than 3% indicates an acceptable precision).

Table A2
Mg and Sr concentrations in ppm of the calcite crystal (Icelandspar) obtained by LA-ICP-MS and ICP-AES.

Elements	LA-ICP-MS[a]	SD[b]	ICP-AES[c]	SD[b]
Mg	674	57	656	62
Sr	184	15	160	8

[a] Data calculated from 643 analyses taken during a 4 year period.
[b] Standard deviation.
[c] Data calculated from 10 analyses.

Table A3
Mg/Ca of cultured foraminifera shells (Chambers stage 19; control culture experiments.

Experiment no.	Salinity	Specimen no.	Mg/Ca (mmol/mol)	SD[a] of single values	Mg/Ca (mmol/mol)[b]	SD[a]	Average Mg/Ca (mmol/mol)	SD[a]
1 (control 1)	30	1	3.54	0.34				
			3.33	0.10	3.43	0.24		
		2	3.22	0.10				
			2.95	0.14	3.08	0.19		
		3	3.29	0.07				
			2.64	0.13	2.97	0.39	3.16	0.24
5 (control 2)	36	1	3.53	0.12	3.51	0.37		
			3.89	0.14				
			3.10	0.20				
		2	3.99	0.08				
			4.24	0.03	4.12	0.15		
		3	5.16	0.27	5.16	0.27		
		4	4.26	0.07				
			3.93	0.08	4.09	0.20		
		5	3.06	0.01				
			2.81	0.07	2.94	0.15		
		6	4.78	0.04				
			4.44	0.13	4.61	0.27		
		7	2.43	0.19				
			2.75	0.12	2.59	0.23		
		8	3.29	0.10				
			3.47	0.09	3.38	0.13	3.80	0.86
9 (control 3)	39	1	3.22	0.11	3.22	0.11		
		2	4.14	0.31	4.14	0.31		
		3	5.57	0.16	5.57	0.16		
		4	5.31	0.47	5.31	0.47	4.56	0.96

Data shown in Fig. 2B).

[a] Standard deviation.

[b] Values correspond to one laser ablation (LA) analysis or the average of two to three LA analyses.

Table A4
Mg/Ca of cultured foraminifera shells (chambers stage 18, all culture experiments).[a]

Experiment no.	Salinity	Specimen no.	Mg/Ca (mmol/mol)	SD[b] of single values	Mg/Ca (mmol/mol)[c]	SD[b]	Average Mg/Ca (mmol/mol)	SD[b]
1 (control 1)	30	1	4.45	0.13				
			4.06	0.02	4.25	0.24		
		2	5.72	0.11				
			5.86	0.31	5.79	0.20		
		3	3.14	0.26				
			3.27	0.54	3.21	0.36	4.42	1.30
2	36	1	3.71	0.03	3.71	0.03		
		2	5.14	0.23	5.14	0.23	4.42	1.01
3	39	1	6.71	0.51	6.71	0.51		
		2	6.69	0.72	6.69	0.72		
		3	4.24	0.67	4.24	0.67		
		4	4.81	0.40	4.81	0.40	5.61	1.28
5 (control 2)	36	1	5.20	0.01				
			5.22	0.28	5.21	0.16		
		2	5.65	0.15	5.65	0.15		
		3	5.19	0.03				
			5.24	0.06	5.21	0.05		
		4	4.09	0.02				
			4.44	0.13	4.27	0.21		
		5	5.97	0.01	5.97	0.01		
		6	4.79	0.17				
			4.45	0.01	4.62	0.22	5.16	0.63
6	39	1	6.28	0.05				
			6.25	0.32				
			6.16	0.51	6.23	0.27		
		2	5.48	0.20				
			5.51	0.27	5.50	0.20	5.86	0.52
7	30	1	4.13	0.21	4.13	0.21		
		2	2.57	0.14				
			3.17	0.18	2.87	0.11	3.50	0.89
8	36	1	4.64	0.27	4.64	0.27		
		2	4.16	0.19	4.16	0.19	4.40	0.34
9 (control 3)	39	1	3.53	0.05				
			3.28	0.35	3.41	0.25		
		2	3.97	0.27	3.97	0.27		
		3	6.10	0.21	6.10	0.21		
		4	6.53	0.20	6.53	0.20		
		5	5.08	0.41				
			5.16	0.19	5.12	0.27	5.03	1.34

[a] Experiment no. 4 is not reported due to lack of specimens.

[b] Standard deviation.

[c] Values correspond to one laser ablation (LA) analysis or the average of two to three LA analyses.

Table A5
Sr/Ca of cultured foraminifera shells (Chambers stage 18, all culture experiments).[a]

Experiment no.	Salinity	Specimen no.	Sr/Ca (mmol/mol)	SD[b] of single values	Sr/Ca (mmol/mol)[c]	SD[b]	Average Sr/Ca (mmol/mol)	SD[b]
1 (control 1)	30	1	1.46	0.11				
			1.22	0.09	1.34	0.17		
		2	1.38	0.16				
			1.33	0.15	1.36	0.04		
		3	1.2	0.15				
			1.32	0.17	1.26	0.08	1.32	0.05
2	36	1	1.23	0.18	1.23	0.18		
		2	1.37	0.24	1.37	0.24	1.30	0.10
3	39	1	1.64	0.33	1.64	0.33		
		2	1.45	0.3	1.45	0.30		
		3	1.37	0.07	1.37	0.07		
		4	1.29	0.07	1.29	0.07	1.44	0.15
5 (control 2)	36	1	1.4	0.09				
			1.43	0.09	1.42	0.02		
		2	1.29	0.06				
			1.38	0.07	1.34	0.06		
		3	1.35	0.05				
			1.37	0.06	1.36	0.01		
		4	1.29	0.09				
			1.34	0.1	1.32	0.04		
		5	1.29	0.07	1.29	0.07		
		6	1.27	0.07	1.27	0.07		
		7	1.5	0.08				
			1.42	0.08	1.46	0.06	1.35	0.07
6	39	1	1.41	0.12				
			1.39	0.12				
			1.45	0.13	1.42	0.03		
		2	1.46	0.08				
			1.34	0.07	1.40	0.08	1.41	0.01
7	30	1	1.41	0.08	1.41	0.08		
		2	1.3	0.08				
			1.41	0.09	1.36	0.08	1.38	0.04
8	36	1	1.4	0.1	1.40	0.10		
		2	1.24	0.1	1.24	0.10	1.32	0.11
9 (control 3)	39	1	1.19	0.09				
			1.26	0.09	1.23	0.05		
		2	1.3	0.11	1.30	0.11		
		3	1.41	0.13	1.41	0.13		
		4	1.42	0.21				
			1.46	0.21	1.44	0.03	1.34	0.10

[a] Experiment No. 4 is not reported due to lack of analysis.

[b] Standard deviation.

[c] Values correspond to one laser ablation (LA) analysis or the average of two to three LA analyses.

Table A6
Mg/Ca and Sr/Ca of cultured foraminifera shells (chamber stage 18, data shown in Fig. 5 A, B, C and D).

Ω[a]	[CO$_3^{2-}$] (µmol/kg)	Specimen no.	Mg/Ca (mmol/mol)	STDV	Average Mg/Ca (mmol/mol)	SD[b]	Sr/Ca (mmol/mol)	SD[b]	Average Mg/Ca (mmol/mol)	SD[b]
3.16	133	1	5.82	0.33	5.82	0.33	1.41	0.11	1.41	0.11
10.16	426	1	6.76	0.32			1.66	0.07		
		2	6.67	0.26			1.62	0.04		
		3	4.12	0.39			1.39	0.03		
		4	5.56	0.21			1.58	0.06		
		5	6.94	0.49	6.01	1.19	1.63	0.06	1.58	0.11
13.09	550	1	4.80	0.44	4.80	0.44	1.54	0.11	1.54	0.11

[a] From da Rocha et al. in prep.

[b] Standard deviation.

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