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Uncertainties in seawater thermometry deriving from intratest and intertest Mg/Ca variability in *Globigerinoides ruber*

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[1] Laser ablation inductively coupled plasma–mass spectrometry microanalysis of fossil and live *Globigerinoides ruber* from the eastern Indian Ocean reveals large variations of Mg/Ca composition both within and between individual tests from core top or plankton pump samples. Although the extent of intertest and intratest compositional variability exceeds that attributable to calcification temperature, the pooled mean Mg/Ca molar values obtained for core top samples between the equator and >30°S form a strong exponential correlation with mean annual sea surface temperature (Mg/Ca mmol/mol = 0.52 $\exp^{0.076\mathrm{SST}°\mathrm{C}}$, $r^2 = 0.99$). The intertest Mg/Ca variability within these deep-sea core top samples is a source of significant uncertainty in Mg/Ca seawater temperature estimates and is notable for being site specific. Our results indicate that widely assumed uncertainties in Mg/Ca thermometry may be underestimated. We show that statistical power analysis can be used to evaluate the number of tests needed to achieve a target level of uncertainty on a sample by sample case. A varying bias also arises from the presence and varying mix of two morphotypes (*G. ruber ruber* and *G. ruber pyramidalis*), which have different mean Mg/Ca values. Estimated calcification temperature differences between these morphotypes range up to 5°C and are notable for correlating with the seasonal range in seawater temperature at different sites.

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1. Introduction

[2] Foraminiferal Mg/Ca seawater thermometry is a rapidly developing and increasingly widely used tool for paleoceanographic reconstruction [*Nürnberg et al.*, 1996; *Rosenthal et al.*, 1997; *Lea et al.*, 1999; *Elderfield and Ganssen*, 2000; *Lea et al.*, 2000; *Anand et al.*, 2003; *Barker et al.*, 2005]. The exponential increase of bulk test Mg/Ca composition with seawater temperature is well established from deep-sea sediment core top [*Rosenthal et al.*, 1997; *Hastings et al.*, 1998; *Elderfield and Ganssen*, 2000; *Lea et al.*, 2000; *Rosenthal et al.*, 2000; *Dekens et al.*, 2002; *Rosenthal and Lohmann*, 2002], plankton net and sediment trap samples [*Anand et al.*, 2003; *McKenna and Prell*, 2004], and laboratory culture studies [*Nürnberg et al.*, 1996; *Lea et al.*, 1999; *Mashiotta et al.*, 1999]. Reported Mg/Ca values however, are notable for being significantly dispersed about thermometer calibrations [*Elderfield et al.*, 2002; *Dekens et al.*, 2002]. This dispersion requires the presence of unaccounted for heterogeneity or biases within the bulk Mg/Ca composition of foraminifer tests that make up deep-sea core samples.

[3] Recent microanalysis studies have documented significant intratest and intertest Mg/Ca variation within sampled populations of various planktonic foraminifer species [*Nürnberg et al.*, 1996; *Eggins et al.*, 2004; *Anand and Elderfield*, 2005; *Sadekov et al.*, 2005; *Kunioka et al.*, 2006]. However, the extent to which this compositional heterogeneity influences the reproducibility of bulk test Mg/Ca analyses and the achievable precision and accuracy of seawater thermometry has not been rigorously assessed. Meanwhile the uncertainty of Mg/Ca paleotemperature estimates has been shown by studies employing bulk analysis techniques to decrease with increasing number of foraminiferal tests analyzed [*Anand and Elderfield*, 2005]. It has been suggested, that by pooling and analyzing at least 20 tests of *G. ruber*, the uncertainty of temperature estimates can be reduced to less than ±1°C (2σ) [*Barker et al.*, 2003; *Anand and Elderfield*, 2005]. To be generally applicable, this claim requires a similar population variance structure for the distribution of individual test Mg/Ca values in all cases. However, sample Mg/Ca variability is likely to be influenced by a range of site-specific biological and environmental factors, including the amplitude of seasonal and interannual temperature changes, differences in sedimentation rate and depth of bioturbation. Moreover, variation in the biological control of calcite biomineralization [*Bentov and Erez*, 2006], during both test growth and chamber wall thickening [*Sadekov et al.*, 2005], may also affect the Mg/Ca composition of individual foraminifer tests. Collectively, these factors may contribute to Mg/Ca variability within a population of tests and, thereby, increase the uncertainty of and bias paleoseawater temperature estimates.

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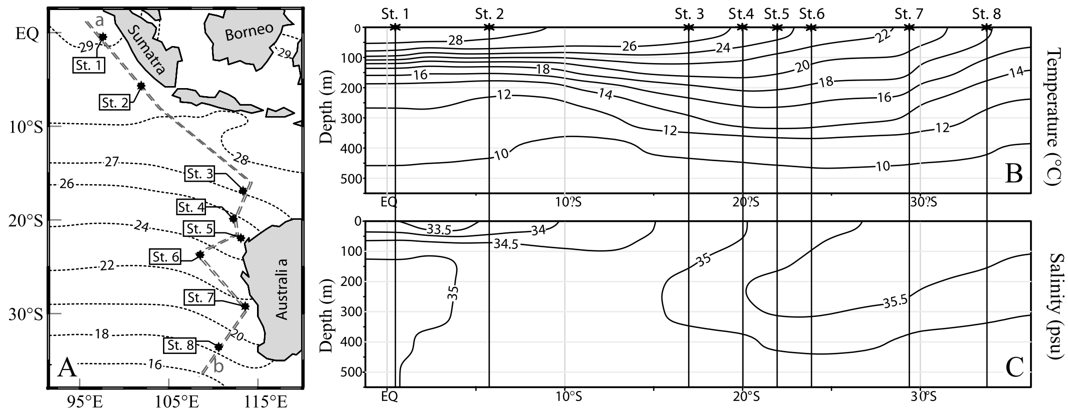

Figure 1. (a) Map showing the location of core top samples (stars) and mean annual sea surface temperature (dashed contour lines). Cross sections of subsurface annual (b) sea surface temperature and (c) salinity along the transect a–b shown on In Figure 1a. Data are from World Ocean Atlas 2001 [*Conkright et al.*, 2002].

[4] The primary aim of this study is to evaluate the extent to which Mg/Ca variability at the individual foraminifer and sample population levels impacts paleotemperature estimates based on the bulk (mean) Mg/Ca composition of a population of foraminifer tests. We have specifically targeted *G. ruber* because it is a key, mixed layer dwelling species that is widely used for sea surface temperature reconstruction and is considered to be little affected by calcite precipitation at lower temperatures within the thermocline.

2. Materials and Methods

[5] *Globigerinoides ruber* tests were obtained from eight core top samples spanning a large latitudinal range (equator to 35°S) and sea surface temperature range in the eastern Indian Ocean (Figure 1 and Table 1). These samples were selected based on (1) the absence of visible foraminiferal test dissolution; (2) low test fragmentation; and (3) retrieval from depths in the range 1000–2500 m. The annual sea surface temperature (SST) for each station was taken from the World Ocean Atlas 2001 [*Conkright et al.*, 2002] and ranged from 29.2°C near the equator to 18.5°C off the southwestern margin of Australia. In addition to these core top samples, we also analyzed several plankton pump samples from the Indian Ocean collected during the *Snellius II* expedition (Table 1, for details see [*Brummer and Kroon*, 1988]).

[6] Between 20 and 35 *G. ruber* tests were picked randomly from >250 μm sediment fractions in each core top sample. Foraminifer tests were cleaned prior to analysis by ultrasonicating individual chamber in methanol for 2–3 s to remove clay and other adhering detrital material. The tests surfaces were then examined under a high-magnifica-

Table 1. Core Top and Plankton Pump Sample Locations, Seafloor Depths, and Calibrated ^{14}C Age of the Core Top Samples[a]

Core Top Samples[b]	Latitude, deg	Longitude, °E	Depth, m	Annual SSS, ‰	Annual SST, C°	SD SST	SE SST	SST Seasonality, C°	Age, B.P.
BARP9411 (1)	−0.46	97.61	2055	33.4	29.16	0.84	0.30	1.8	2737 ± 46
SHIVA 9045 (2)	−5.65	101.90	2340	33.5	28.39	0.84	0.25	2.2	2005 ± 60
FR2/96-GC23 (3)	−16.91	113.34	1967	34.6	27.29	1.41	0.13	3.8	5071 ± 120
FR10/95-GC15 (4)	−19.90	112.22	1393	34.9	26.00	2.20	0.52	4.2	11245 ± 58
FR10/95-GC17 (5)	−22.13	113.50	1093	35.1	25.16	1.73	0.25	4.9	1187 ± 87
FR2/96-GC12 (6)	−23.74	108.53	2100	35.3	23.37	1.69	0.39	3.6	11487 ± 146
FR10/95-GC26 (7)	−29.24	113.56	1738	35.5	21.63	1.86	0.08	3.9	2240 ± 55
MD94-11 (8)	−33.58	110.58	2400	35.7	18.47	1.95	0.32	4.0	16885 ± 217
Plankton Pump Samples	**Latitude, deg**	**Longitude, deg**	**Depth, m**	**Measured SSS, ‰**	**Measured SST, C°**				
138	8.37	71.89	0–5	35.9	28.87				
145	7.98	76.14	0–5	35.7	28.88				
152	6.82	79.78	0–5	35.7	28.25				
154	5.56	80.94	0–5	35.3	28.38				
174	−1.20	94.08	0–5	34.8	29.43				
178	−2.33	95.69	0–5	34.6	28.94				
192	−6.24	103.15	0–5	34.3	29.29				

[a]Mean annual sea surface temperatures, their standard errors and standard deviations, and the salinity values for each core top sample have been taken from the World Ocean Atlas 2001 [*Conkright et al.*, 2002]. Abbreviations are SSS, sea surface salinity; SD, standard deviation; and SE, standard error.
[b]Station number is given in parentheses.

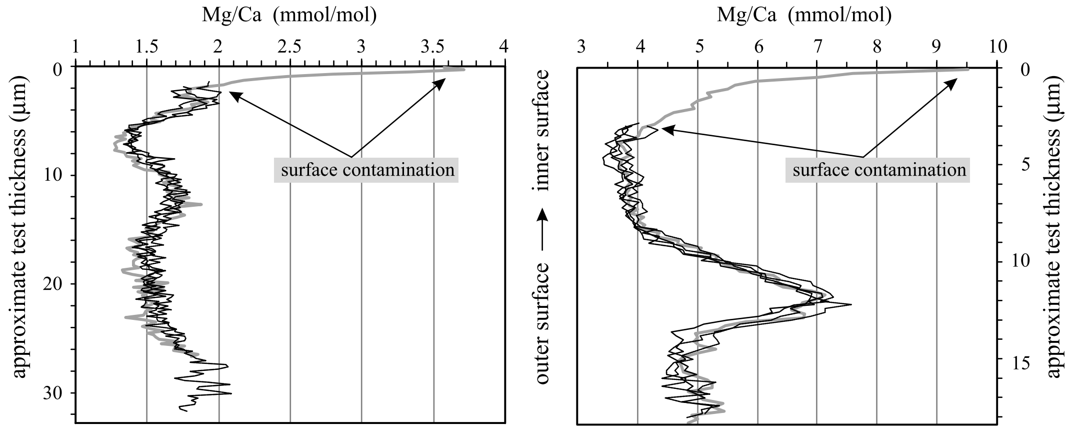

Figure 2. Examples of laser ablation ICP-MS profiles of Mg/Ca (mmol/mol) through the final chamber walls of *G. ruber* from representative temperate and tropical region core top samples (stations 8 and 2). Each line represents a single chamber wall profile analysis; shaded and black lines show Mg/Ca profiles for the same test chamber without and with laser ‘‘preablation’’ cleaning, respectively (see text for details).

tion stereomicroscope for the presence of any remaining surface contamination, and reultrasonicated if necessary. The last two to four chambers of each test were removed with a surgical scalpel, and mounted using carbon tape on a glass slide for analysis by laser ablation inductively coupled plasma–mass spectrometry (ICP-MS). A high-resolution depth profiling technique that employs a pulsed ArF excimer laser (λ = 193 nm) coupled to an Agilent 7500 s ICP-MS at the Research School of Earth Sciences ANU was used to measure depth profiles through test walls [*Eggins et al.*, 2003, 2004]. The isotopes ^{24}Mg, ^{25}Mg, ^{43}Ca, ^{44}Ca, ^{55}Mn, and ^{27}Al were measured during each depth profile analysis, which required only 20–120 s to acquire, and between two and seven profiles were generated for each test chamber. The horizontal and vertical resolution of the technique was optimized by ablating small-diameter spots (30 μm) at four laser pulses per second. Data reduction involved initial screening of spectra for outliers, subtraction of the mean background intensities (measured with the laser turned off) from the analyzed isotope intensities, internal standardization to ^{43}Ca and ^{44}Ca, and external standardization using the NIST-SRM610 glass reference material.

[7] Test surfaces were also ‘‘preablated’’ prior to each analysis by the application of 3–5 pulses to remove the outer 0.5 μm or so of the test surface. Significant enrichment in Mg and other trace elements was observed on test surfaces that were not ‘‘preablated’’ (Figure 2). High surface concentrations of Mg, Al and Mn have been previously reported by [*Eggins et al.*, 2003] and attributed to the presence of diagenetically modified surface calcite [*Pena et al.*, 2005]. The ‘‘pre ablation’’ technique provides a simple method of test cleaning, with some significant advantages over commonly used cleaning procedures [*Hastings et al.*, 1998; *Martin and Lea*, 2002; *Barker et al.*, 2003; *Rosenthal et al.*, 2004; *Pena et al.*, 2005; *Weldeab et al.*, 2006] in that it requires minimal time, and yet it effectively removes contaminated surface calcite material.

[8] Approximate ages for the core top samples were established from ^{14}C dates measured at the Poznañ Radiocarbon Laboratory, Potsdam, and the Australian Nuclear Science and Technology Organisation-ANSTO [*Olley et al.*, 2004] (also please see details of method used by *Fink et al.* [2004]) (reported in Table 1). We have used the CALIB5.0 program to calibrate δ^{14}C dates into calendar age, for marine samples.

3. Results

3.1. Intratest Variability

[9] In previous studies, we have shown that intratest Mg/Ca variability is linked to; (1) the presence of calcite layers with differing Mg/Ca compositions within chamber cross sections, and (2) variation in the arrangement and thickness of high– and low–Mg/Ca layers within different chambers of the same test and between individual tests of the same species [*Eggins et al.*, 2004; *Sadekov et al.*, 2005]. To characterize the Mg/Ca variability within individual foraminiferal tests of *G. ruber*, we have analyzed and compared the compositions of the final two to four chambers of tests from core top station 2 and plankton station 192 (see Figure 1 and Table 1; for details about plankton samples, see *Brummer and Kroon* [1988]).

[10] Examples of typical compositional profiles through the last four chambers are presented in Figure 3. All show the presence of intercalated layers with relatively low– and high–Mg/Ca compositions. The high–Mg/Ca layers are usually 1.5 to 2.5 times more enriched in Mg relative to low–Mg/Ca layers. The high–Mg/Ca layers also show differences in composition both within and between chambers, whereas the low-Mg calcite layers tend to be more consistent within individual test (Figure 3).

[11] Owing to the presence of intercalated high– and low–Mg/Ca layers, the mean Mg/Ca value of a chamber wall profile (hereafter referred to as the ‘‘profile mean’’) typically has a large standard deviation. However, repeat analyses of specific chambers show good reproducibility such that the average of the profile means for a particular chamber (hereafter referred to as the ‘‘chamber mean’’), exhibits a small standard deviation. We further note that the chamber mean Mg/Ca of the final chamber usually displays

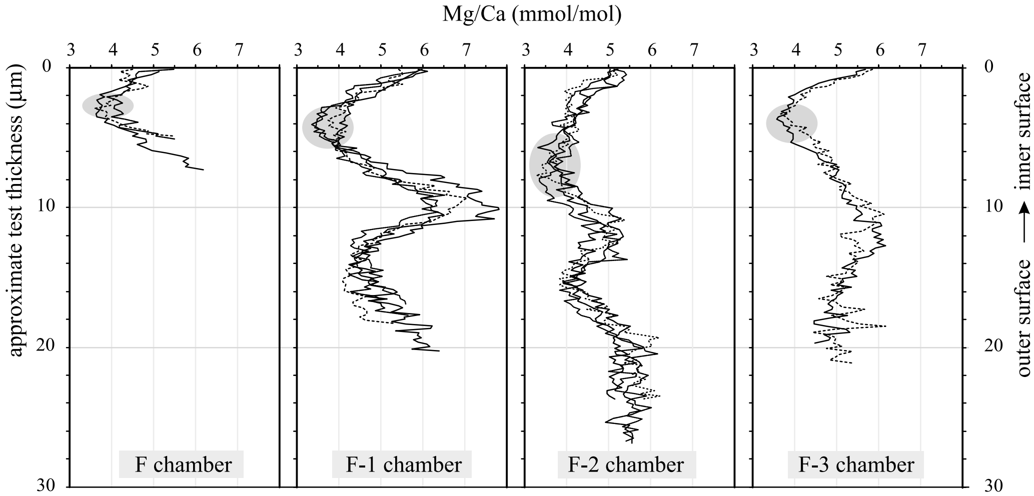

Figure 3. Mg/Ca profiles through the final four chambers of a single *G. ruber* test, showing the large variation of Mg/Ca values and the intercalation of low– and high–Mg/Ca layers. Note the first low–Mg/Ca calcite layer in each profile, which is marked by the shaded area, has a similar Mg/Ca concentration in all chambers.

the smallest standard deviation, which typically does not exceed 0.2 mmol/mol (Table 2).

[12] The mean Mg/Ca values of different chambers within the same test often display large differences (Figure 4), ranging up to 50% (compare chambers F-2 and F in test 8). Close inspection of the chamber wall profiles indicates that these differences are related to the development of high-Mg layers in the different chambers, with both the number and thickness of high-Mg layers varying from chamber to chamber. The final chamber usually has one low-Mg layer sandwiched between two high-Mg layers on both inner and outer surfaces (Figure 3), whereas the penultimate chambers have multiple high– and low–Mg/Ca layers. There is no direct relationship between the position of the chamber within the test whorl and the number of the high-Mg layers.

[13] We have not found any systematic relationship between a chamber's mean Mg/Ca value and its position within the test whorl (Figure 4). Some tests show an increase in the mean Mg/Ca value from the final chamber (F) to the second last chamber (F-1) and then the third last chamber (F-2), whereas others show the opposite trend or a random relationship. Importantly, however, no significant difference is observed between the average Mg/Ca composition of the final two or three chambers of the 17 tests used for this comparison (see insert on Figure 4).

3.2. Intertest Variability

[14] To characterize intertest variability, we analyzed between 20 and 35 *G. ruber* tests from each of the eight core tops. The Mg/Ca composition of the final chamber was used as a measure of the Mg/Ca value of each test, based on the observation that it exhibits the least intratest Mg/Ca variability (see above). The mean Mg/Ca value for each core top sample (hereinafter referred to as the "sample mean") was then calculated by averaging the chamber means obtained from each test. These results are summarized in Table 2.

[15] The distribution of the individual test Mg/Ca values relative to annual SST is shown in Figure 5. Each core top sample is characterized by a large range of individual test Mg/Ca values, for example, from 1 to 4.5 mmol/mol in the case of station 5. The standard deviations of the different Mg/Ca core top sample means vary significantly and range from 0.6 to 1.1 mmol/mol (average 0.8 mmol/mol). Importantly, the relative standard deviation calculated for the individual test Mg/Ca values increase with the size of the seasonal temperature range at each core top site (Figure 5).

[16] Although fewer tests were available for analysis, the plankton pump samples display a large dispersion of test compositions and have absolute Mg/Ca values that are consistent with the core top samples.

3.3. Relationship Between the Sample Mean Mg/Ca and Annual SST

[17] We have used simple linear regression modeling to assess the relationship between the sample mean Mg/Ca composition and annual SST. An exponential function was found to best describe the relationship (Figure 5) between the middle to late Holocene (e.g., younger 3 ka) core top samples as follows:

$$\mathrm{Mg/Ca} = 0.52\,\exp(0.076\mathrm{T}^\circ\mathrm{C})$$

This is equivalent to 7.6 ± 1.3% change in Mg/Ca composition for 1°C change in temperature. This regression model accounts for 95% of the observed variation of the sample mean Mg/Ca values. Full results of the regression analysis are summarized in Table 3.

3.4. Mg/Ca Thermometer Uncertainties: Sample Size

[18] To evaluate the dependence of the temperature uncertainty obtained using our Mg/Ca thermometer upon sample size (i.e., number of tests analyzed), we have determined the relationship between the standard error (SE) of the sample mean and sample size (the number of

Table 2. Summary of the Measured Mg/Ca Values of Individual *G. ruber* Tests[a]

	Specimen																					
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD of Average
Station 1																						
Number of tests	5	5	5	5	5	5	5	5	5	5	4	5	5	4	5	5	3	4	5	5		
Mean	5.19	4.82	5.37	3.31	4.10	3.53	4.84	4.63	5.12	5.33	4.56	5.84	5.74	4.63	5.40	5.41	4.43	5.56	4.79	4.03	4.83	0.70
Standard deviation	0.42	0.34	0.18	0.14	0.11	0.35	0.46	0.11	0.38	0.21	0.48	0.42	0.41	0.39	0.46	0.36	0.08	0.21	0.14	0.30	0.30	0.14
Morphotype	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r	r		
Station 2	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	4	3	3	2	4	4	4	5	4	2	2	3	2	3	3	3	2	3	3	3		
Mean	5.14	4.40	4.49	4.50	5.18	4.24	4.53	5.21	4.84	4.30	3.03	6.04	3.63	3.62	4.14	4.56	4.41	3.44	4.23	4.34		
Standard deviation	0.19	0.02	0.33	0.46	0.09	0.08	0.21	0.28	0.24	0.06	0.01	0.10	0.22	0.27	0.11	0.05	0.08	0.24	0.07	0.28		
Morphotype	r	r	r	p	r	r	p	r	r	r	r	r	r	r	r	r	p	p	p	p	4.45	0.89
Station2 continued	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35						0.23	0.17
Number of tests	2	3	2	3	2	7	2	2	3	5	4	4	5	4	3							
Mean	4.11	5.45	5.34	3.71	5.98	7.32	5.15	4.56	3.69	3.29	3.62	3.80	3.45	3.96	4.11							
Standard deviation	0.43	0.14	0.27	0.21	0.76	0.49	0.07	0.10	0.45	0.21	0.30	0.43	0.33	0.38	0.05							
Morphotype	r	r	r	r	r	r	r	r	r	r	r	p	p	r	r							
Station 3	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	4	4	4	4	4	4	5	5	5	5	4	5	4	4	4	5	5	4	5	5		
Mean	3.94	5.29	4.43	5.06	3.29	2.87	4.83	3.85	2.70	3.55	3.73	4.55	4.06	2.86	3.01	3.45	4.06	2.58	3.31	2.40	3.69	0.84
Standard deviation	0.46	0.40	0.11	0.54	0.08	0.13	0.18	0.14	0.10	0.15	0.16	0.19	0.11	0.18	0.08	0.35	0.36	0.13	0.12	0.31	0.21	0.14
Morphotype	r	r	r	r	r	p	r	r	p	p	r	r	r	p	p	r	r	r	r	r		
Station 4	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	5	5	5	5	5	5	5	5	5	5	5	4	4	5	5	3	4	5	5	5		
Mean	3.72	3.76	5.86	2.74	2.58	4.00	2.24	3.05	3.81	2.18	3.80	3.55	3.06	3.46	3.49	6.38	3.50	4.42	3.51	3.30	3.62	1.03
Standard deviation	0.24	0.16	0.91	0.05	0.17	0.25	0.19	0.34	0.64	0.18	0.17	0.20	0.43	0.10	0.24	0.09	0.25	0.09	0.17	0.06	0.25	0.21
Morphotype	r	r	r	r	r	r	p	p	r	p	r	r	p	r	r	r	r	r	r	r		
Station 5	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	3	5	4	3	3	4	5	5	4	5	4	4	4	5	5	5	4	4				
Mean	4.45	2.66	5.14	3.67	5.02	3.47	2.48	2.75	2.97	5.67	4.13	2.72	2.24	3.01	4.33	3.41	4.53	2.08			3.60	1.08
Standard deviation	0.42	0.23	0.31	0.09	0.59	0.07	0.12	0.11	0.27	0.43	0.72	0.11	0.09	0.18	0.21	0.12	0.46	0.13			0.26	0.19
Morphotype	r	r	r	p	r	p	p	r	p	p	r	p	p	r	r	r	p	p				
Station 6	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	3	3	5	3	2	5	5	5	5	5	5	5	5	4	5	4	5	4	3	3		
Mean	2.68	2.77	3.50	2.54	4.05	2.88	3.15	2.79	3.91	3.44	2.20	2.51	2.75	2.37	2.90	2.45	3.02	2.58	4.17	3.23	2.99	0.57
Standard deviation	0.18	0.23	0.21	0.09	0.21	0.04	0.23	0.09	0.20	0.19	0.06	0.12	0.04	0.07	0.12	0.06	0.16	0.06	0.32	0.19	0.14	0.08
Morphotype	r	r	r	p	r	r	r	r	r	p	r	r	p	p	r	p	r	r	r	r		
Station 7	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	4	4	4	4	2	4	5	5	5	4	4	2	5	4	4	4	4	4	5	4		
Mean	2.23	2.23	2.66	2.95	2.40	2.76	3.64	3.02	2.28	2.34	2.35	2.09	2.61	2.66	2.43	1.75	2.11	2.96	5.53	2.66	2.68	0.79
Standard deviation	0.14	0.18	0.15	0.10	0.23	0.12	0.20	0.11	0.28	0.03	0.46	0.11	0.15	0.03	0.08	0.10	0.10	0.06	0.41	0.23	0.16	0.11
Morphotype	r	r	r	r	p	p	r	r	r	r	r	p	p	r	p	p	r	p	r	r		
Station 8	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Average	SD
Number of tests	5	4	5	5	5	5	4	5	5	5	5	5	5	5	5	5	4	5	5	5		
Mean	2.81	4.46	2.26	2.45	2.91	1.30	1.58	3.09	2.28	1.36	1.31	1.24	1.47	1.43	2.18	2.25	1.61	2.91	2.56	1.07	2.13	0.85
Standard deviation	0.15	0.14	0.18	0.18	0.19	0.04	0.09	0.04	0.15	0.04	0.02	0.09	0.07	0.05	0.11	0.18	0.05	0.14	0.11	0.05	0.10	0.06
Morphotype	r	r	r	r	p	p	p	p	p	r	p	p	p	p	r	r	p	r	r	p		
Plankton samples	1	2	3	4	5	6	7	8	9	10	11	12	13								Average	SD
Station number	138	138	145	152	154	174	178	192	192	192	192	192	192								for st 192	
Number of tests	3	3	1	2	2	2	3	3	4	3	3	4	4									
Mean	4.11	3.56	3.62	5.19	3.46	5.52	4.06	4.28	4.58	3.89	5.72	4.98	5.40								4.81	0.71
Standard deviation	0.15	0.02	0.22	0.30	0.22	0.12	0.13	0.31	0.19	0.13	0.99	0.28	0.23								0.25	0.24

[a]Abbreviations are SD, standard deviation; r, morphotype *G. ruber ruber*; and p, morphotype *G. ruber pyramidalis*.

individuals averaged to estimate the sample mean). This simple approach has been used previously to characterize the error of Mg/Ca paleothermometry for two other planktonic foraminifer species, *Globorotalia truncatulinoides* and *Globigerina bulloides* [*Anand and Elderfield*, 2005]. Following recalculation of the standard error values into degrees Celsius using the regression result obtained in the previous paragraph, we find that all core top samples display a similar pattern of decreasing temperature uncertainty with the square root of the number of analyzed tests (Figure 6). However, the standard error of SST estimates differs significantly at a sample size of 20 tests, ranging between 0.8 and 2.4°C for the different core top samples (Figure 6). In the case of stations 6, 7, 1 and 2 an uncertainty of ±1°C (SE) is achieved with sample populations comprising only 20 tests, whereas for stations 3, 4, 5 and 8 significantly larger uncertainties ranging from ±1.3 up to ±2.4°C are achieved at the same sample size. No clear relationship is observed between the calculated temperature uncertainty (n = 20 sample) and core top sample ^{14}C age (Figure 6).

3.5. Mg/Ca of *G. ruber* Morphotypes (Subspecies)

[19] Four morphotypes of *G. ruber* have been identified as part of this study that are similar to the species *G. ruber, G. pyramidalis, G. elongatus* and *G. cyclostomatus,* previously described by *Saito et al.* [1981]. However, the latter three morphotypes have proved difficult to separate because

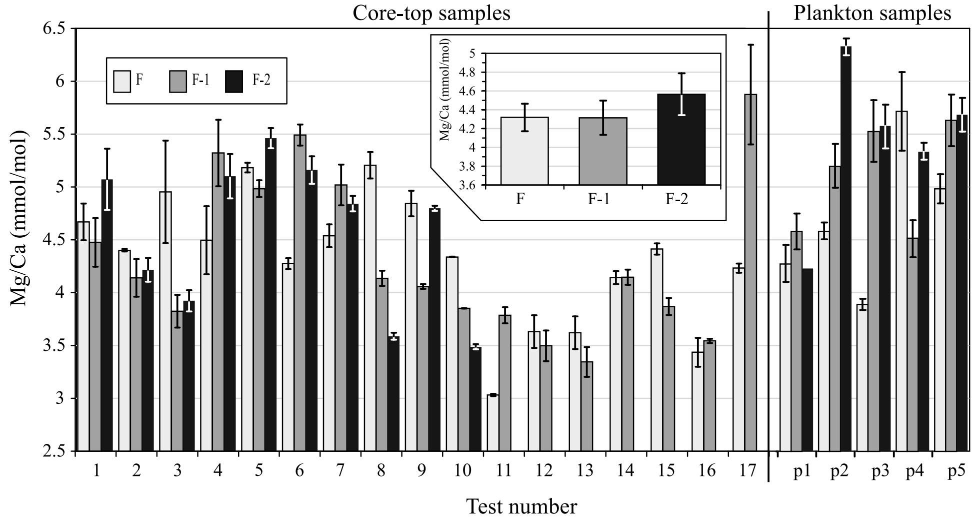

Figure 4. Comparison of the mean Mg/Ca values obtained for different chambers in 17 *G. ruber* tests obtained from a single core top sample (station 2) and five tests from a plankton pump sample (station 192). The labels F, F-1, and F-2 indicate the final, penultimate, and antepenultimate chambers. Inset shows a comparison of the mean Mg/Ca composition of the F, F-1, and F-2 chambers for the 17 tests from core top sample. Error bars indicate the size of the standard error of the chamber mean values in each test.

of intergradational changes in their test morphology [*Sadekov et al.*, 2006]. Accordingly, to evaluate the effect of test morphology on Mg/Ca variability, a simplified approach has been used that subdivides the tests into two morphotype groupings; *G. ruber ruber* and *G. ruber pyramidalis*, which are equivalent to the morphotypes *G. ruber* s. s. and *G ruber* s. l. previously used by *Wang* [2000], *Kuroyanagi and Kawahata* [2004], *Steinke et al.* [2005] and *Löwemark et al.* [2005]. Here we have employed the different taxon names because these morphotypes have distinct morphology and are exclusive of each other, hence the terms sensu lato and sensu stricto should not be used [*International Commission on Zoological Nomenclature*, 1999].

[20] Both morphotypes of *G. ruber* are present and relatively abundant in the studied samples, with the proportion of *G. ruber pyramidalis* appearing to increase as SST decreases, until it becomes dominant in station 8. Given all analyzed tests were randomly selected from the >150 μm size fraction in each core top sample, the observed relative proportions of these morphotypes provide estimates of those in the original sample. A comparison of the Mg/Ca sample means for the two subspecies from each core top sample is shown in Figure 7a. These results clearly indicate that *G. ruber pyramidalis* have consistently lower Mg/Ca compositions than *G. ruber ruber*. The deviation of *G. ruber pyramidalis* Mg/Ca values from those of *G. ruber ruber* are much larger for station 3, 4, 5 and 8 and correspond to 3 ± 2°C using the Mg/Ca thermometer calibration derived above.

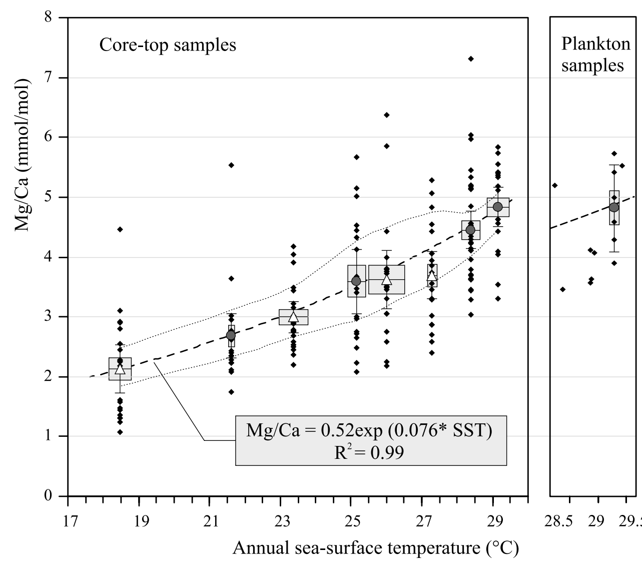

Figure 5. Plots of annual SST at each core top site versus the measured Mg/Ca composition of individual tests (diamonds) and the sample mean Mg/Ca composition for each core top (shaded circles, core tops younger than 3 ka; shaded triangles, core tops older than 3 ka). The standard errors of the sample mean Mg/Ca and the SST values for each core top sample are represented by the height and width of the boxes. Error bars indicate the 95% confidence interval for each sample mean Mg/Ca composition, and the dotted line outlines the envelope of predicted Mg/Ca values based on the seasonal SST range at each site (taken from World Ocean Atlas 2001 [*Conkright et al.*, 2002]).

4. Discussion

4.1. Factors Contributing to the Uncertainty of Mg/Ca Thermometry

[21] Our results indicate that the seasonal range in SST and the occurrence of varying proportions of *G. ruber*

Table 3. Summary of Linear Regression Results Obtained for Fits Between the Natural Logarithm of the Mean Sample Mg/Ca Value and Mean Annual SST of Different Groupings of Core Top Samples and Morphotypes

	Exponential Relationship Mg/Ca = Bexp(A(Temperature))					
	Sample Mean Mg/Ca[a]	Sample Mean Mg/Ca[a] (Fixed A = 0.09)	Sample Mean Mg/Ca[b]	Individual Test Means Mg/Ca[b]	Mg/Ca Values of *G. ruber pyramidalis*[b]	Mg/Ca Values of *G. ruber ruber*[b]
R	0.999	0.983	0.988	0.733	0.912	0.975
R square	0.997	0.966	0.976	0.538	0.832	0.950
ln(B)	−0.66	−1.02	−0.60	−0.74	−0.94	−0.22
Standard error of s(B)	0.08		0.12	0.14	0.38	0.14
95% confidence intervals	0.34		0.29	0.28	0.92	0.35
B	0.52	0.36	0.55	0.48	0.39	0.80
A	0.076	0.09	0.073	0.078	0.081	0.060
Standard error of A	0.002		0.005	0.006	0.015	0.006
95% confidence intervals	0.013		0.011	0.011	0.037	0.014

[a]Young core top samples <3 ka.
[b]All core top samples.

morphotypes, which have distinct Mg/Ca values, contribute to the uncertainty of SST estimates derived using foraminiferal Mg/Ca thermometry. These factors can be related to differences in calcification temperature (i.e., seasonal range or habitat depth) but cannot account for the full range of intertest Mg/Ca variation observed. If the sample variance (standard deviation) is regressed against the seasonal range in SST for each core top site, a positive correlation is found that has a significant nonzero intercept value (Figure 7b) and a slope that is equal to a little more than half the seasonal temperature range. The intercept value is notable for its consistency with the variance of the plankton pump sample (star label at Figure 7b), the latter representing a discrete time with a zero seasonal temperature range. If we make the reasonable assumption that intertest compositional variability in the plankton pump sample is due to biological differences between individual *G. ruber* tests, it follows that the regression intercept value represents the stochastic nature of "vital effect" influences upon individual test Mg/Ca compositions. The size of the intercept value indicates that this vital effect(s) contributes ∼1.6 ± 0.3°C to the apparent temperature variance (see Figure 7b) and possibly as much as ∼2.5 ± 0.3°C (upper 95% confidence limit). We suggest that this vital effect can be linked to the differential development of Mg/Ca banding in individual foraminifers and could be related to symbiont activity in *G. ruber* [see *Eggins et al.*, 2004] or differences in biomineralization processes during layers formation [*Erez*, 2003; *Bentov and Erez*, 2006]. It follows that seasonal changes in seawater temperature may be reconstructed from the variability of

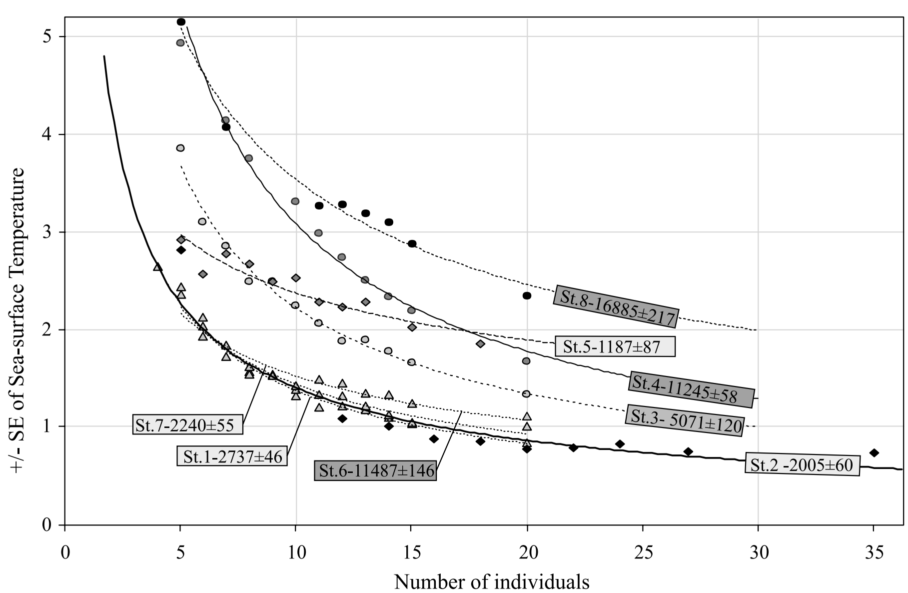

Figure 6. Plot showing the relationship between the standard error (SE) of the estimated SST and the number of analyzed tests from each core top sample. The standard errors have been calculated on a random subsample of tests (typically five) followed by the addition of individual tests until all analyzed tests from a core top sample have been included. Lines show best fit curves through the data for each core top sample. Numbers in boxes indicate the calibrated C^{14} age of each core top sample.

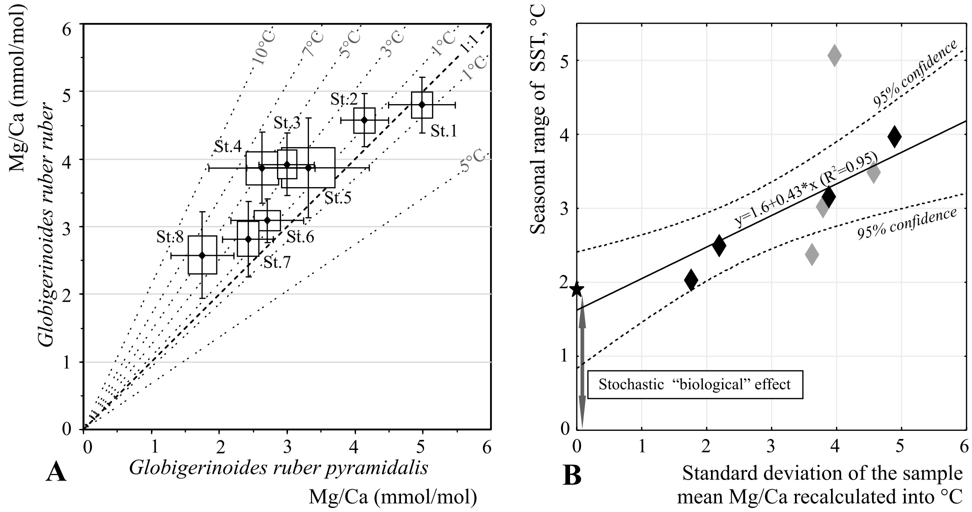

Figure 7. (a) Comparison of the mean Mg/Ca composition of tests of the two *G. ruber* morphotypes (*G. ruber ruber* and *G. ruber pyramidalis*). Boxes indicate the standard error, and error bars indicate the 95% confidence interval for the mean morphotype Mg/Ca composition from each core top sample. Dotted lines indicate the difference in the morphotype Mg/Ca compositions recalculated into SST degrees using the Mg/Ca thermometer calibration determined in this study. (b) Plot of the SST seasonality range at each sample site versus the standard deviation of each core top and plankton sample mean Mg/Ca values, recalculated into equivalent calcification temperature. The regression line and 95% confidence limits have been calculated for the young (3 ka) core top samples (solid diamonds) and plankton sample (star). The shaded diamonds show core top samples older than 3 ka.

Mg/Ca values if the "vital effect" component can be reliably quantified and subtracted. Further studies targeting this possibility are required to better understand the contributions of seasonality of SST and vital effects to the total Mg/Ca variability of foraminiferal tests.

[22] The consistently different Mg/Ca compositions of the *G. ruber* morphotypes suggest that their presence and any variation in the proportions will induce uncertainty in SST reconstructions. *Steinke et al.* [2005] have documented similar differences in Mg/Ca composition between the same *G. ruber* morphotypes we have analyzed in our study, and suggested these differences stem from the various subspecies inhabiting different water depths. Our results show that *G. ruber pyramidalis* have consistently lower Mg/Ca values and thus can be interpreted to have calcified at lower temperature and greater depth than *G. ruber ruber,* as has been suggested in previous studies [*Wang*, 2000; *Kuroyanagi and Kawahata*, 2004; *Löwemark et al.*, 2005; *Steinke et al.*, 2005]. However, we note that smaller differences between the morphotype Mg/Ca compositions occur in tropical locations, which have stronger and shallower thermoclines than higher-latitude locations (compare Figures 1b and 7a). Samples with the largest differences in estimated morphotype calcification temperatures are further noted to come from sites near frontal zones between water masses where large seasonal shifts in temperature occur (see Figure 1 stations 3, 4, 5 and 7). Accordingly, we propose that compositional differences between *G. ruber* morphotypes could reflect SST seasonality in addition to or rather than habitat depth. Similar seasonal changes in *G. ruber* morphology and δ^{18}O values of foraminiferal test have been documented previously by *Spero et al.* [1987].

[23] Irrespective of the origin of the Mg/Ca compositional difference, variation in the mix of *G. ruber* morphotypes within a sample population may significantly bias the value and influence the precision of any seawater temperature estimate. On the basis of the maximum difference in Mg/Ca composition observed in this study, variation in the proportion of these morphotypes could bias SST estimates by as much as 5°C. If on the other hand, SST estimates are derived from a single morphotype, an approach previously proposed to improve the precision of SST reconstruction [e.g., *Steinke et al.*, 2005], a seasonal bias could be imparted onto the derived temperature signal.

4.2. Comparison of Mg/Ca–SST Relationship With Previous Studies

[24] The relationship between sea surface temperature and the sample mean Mg/Ca value found in this study agrees very well with previously published Mg/Ca thermometers for *G. ruber* [*Lea et al.*, 2000; *Dekens et al.*, 2002; *Anand et al.*, 2003; *McConnell and Thunell*, 2005] (see Figure 8). Our calibration most closely matches that of *Dekens et al.* [2002], which is also derived from core top samples, although characterized by less steep exponential growth of Mg/Ca with annual SST (i.e., 7.6% versus 8.9% per °C). The smaller exponential coefficient obtained in this study could be partly due to the lower salinity of our equatorial stations, where salinity is reduced by ~2 salinity units compared to higher latitudes (Figure 1). The effect of salinity on the Mg/Ca composition of foraminiferal calcite is currently poorly constrained, with studies indicating an 11% increase in Mg/Ca per salinity unit for *G. sacculifer* [*Nürnberg et al.*, 1996] and 4% increase per salinity unit for

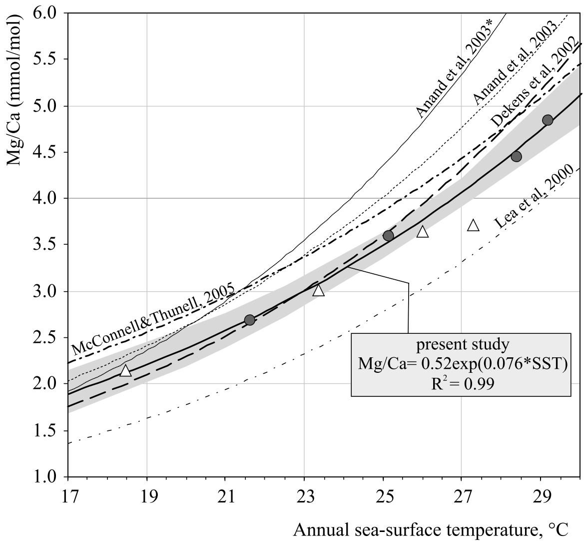

Figure 8. Comparison of published Mg/Ca seawater thermometers for *G. ruber* with the calibration obtained in this study. Core top sample symbols are as used in Figure 5. The shaded area delineates the 95% confidence band for the best fit exponential equation (see text); the asterisk denotes the Mg/Ca thermometer calibration obtained by *Anand et al.* [2003] on the large test size fraction.

O. universa [*Lea et al.*, 1999]. If a modest 5% increase in Mg/Ca per salinity unit is assumed, our equatorial sites would be shifted to ~10% higher Mg/Ca values and result in a significantly larger exponential coefficient (i.e., 9% per °C), in close agreement with the calibration of *Dekens et al.* [2002]. Other thermometer calibrations for *G. ruber* agree less well with our results. The *Anand et al.* [2003] plankton trap–based calibrations for *G. ruber* have a higher exponential growth and predict higher absolute Mg/Ca values by 0.5–1.0 mmol/mol. The *McConnell and Thunell* [2005] plankton trap calibration has a similar exponential growth rate but higher Mg/Ca values by approximately 0.5 mmol/mol. In contrast, the *Lea et al.* [2000] calibration, which is based on core top samples, is shift to lower Mg/Ca by 0.5 mmol/mol (Figure 8). These core top samples were taken from depths between 2050 and 3200 m where selective dissolution of more Mg-rich test calcite can shift residual test compositions toward lower Mg/Ca values [*Brown and Elderfield*, 1996; *Rosenthal et al.*, 2000; *Dekens et al.*, 2002; *Rosenthal and Lohmann*, 2002]. If the Mg/Ca compositions of our samples are corrected for water depth using the algorithm derived for the Pacific Ocean by [*Dekens et al.*, 2002], our recalculated Mg/Ca versus temperature calibration closely matches the plankton trap (dissolution unaffected) calibration of *McConnell and Thunell* [2005] (Figure 8). Collectively, these explanations provide a reassuring degree of accountability between our and the other thermometer calibrations that exist for *G. ruber*. Moreover, if the intercept constant (B) can be adjusted, almost all of the published equations might be located into 95% confidence interval of our regression line. Therefore it is possible that the application of different cleaning techniques play a significant role in determining the accuracy of calibration models. Additional studies are needed to compare cleaning techniques from bulk solution base methods with that of laser ablation ICP-MS.

4.3. Error Estimation for Mg/Ca Thermometry

[25] Our results indicate that *G. ruber* tests taken from different core top sediment samples are characterized by distinct intertest Mg/Ca composition distributions that can be related to multiple site-specific factors that include SST seasonality and the mix of foraminiferal morphotypes. It follows that the analysis of a prescribed number of tests may not everywhere achieve a particular desired level of uncertainty. Rather, a sample-by-sample assessment is preferred to determine the number of tests needed to achieve a specific temperature uncertainty. Power analysis, a statistical method based on the analysis of the standard deviation of sample means, is one approach to this problem that permits calculation of the sample size required to obtain a specified level of uncertainty. Examples showing how power analysis can be used to calculate the sample size (number of tests) required to obtain a specified SST uncertainty (e.g., ±1°C at the 95% confidence level) are given below.

[26] On the basis of our Mg/Ca–thermometer calibration, the expected sample mean Mg/Ca value for a seawater temperature of 28°C ($\mu^{t=28}$) is 4.37 mmol/mol. For a target temperature error (ΔT) of 1°C, the upper 95% confidence limit of the sample mean at 28°C is $\mu_0^{t=28}$ = Mg/Ca (T = 28 + 1°C) = 4.71 mmol/mol. The total variance of any sample can be derived from a combination of the intertest and intratest variances which, using the measured standard deviations of the chamber mean and sample mean Mg/Ca compositions for station 2 as an example, is estimated as follows (i.e., $SD_{total} = \sqrt{((SD_{intertest})^2 + (SD_{intratest})^2)}$ $((0.89)^2 + (0.17)^2)^{1/2} = 0.91$ (value from Table 2). We are then able to propose that the null hypothesis H_0 is $\mu = \mu_0$, where at the 95% confidence level our sample mean will be equal to or less than the upper error limit. Using a single-mean *t* test, H_0 will be rejected up to a sample size of 57 individuals, given a type 1 error $\alpha = 0.05$ and a type 2 error $\beta = 0.80$. This indicates there is only a 5% probability that the true population mean lies above the error value μ_0 and there is an 80% probability that we have rejected H_0 correctly.

[27] A summary of power analysis results calculated for different specified temperature uncertainties is shown on Figure 9, based on the variance characteristics of station 2 which are representative of the samples measured in this study. These results suggest that the errors of reconstructed SSTs were thought to be too small in previous studies, particularly for samples from temperate regions where the ability to discern a 1°C difference in SST with 95% confidence, is estimated to require the analysis of ~250 tests at a SST of 18°C and ~60 tests at 28°C.

[28] Our power analysis results are broadly consistent with those able to be drawn from standard error estimates of SST. Again using station 2 as the example, it can be seen

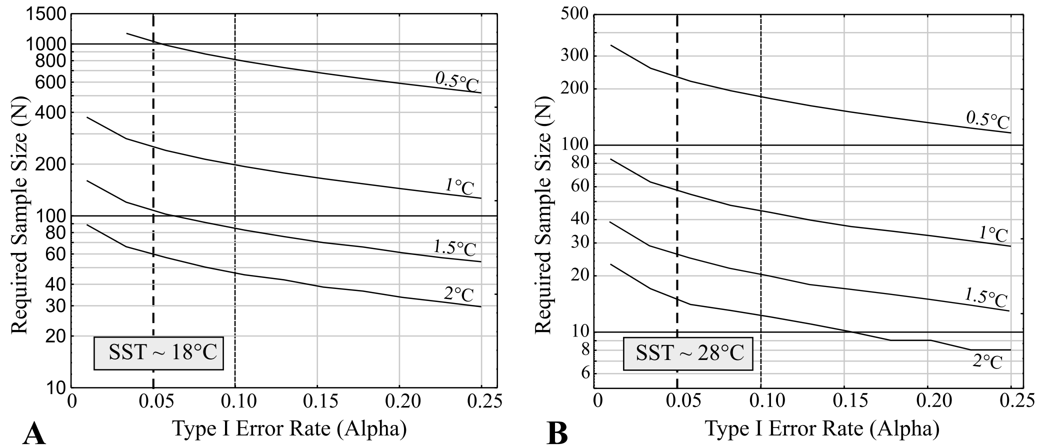

Figure 9. Calculated sample size (number of tests) required to achieve a target temperature uncertainty using Mg/Ca thermometry of *G. ruber* based on power analysis. Results for SST values of (a) 18°C and (b) 28°C based on the typical intratest and intertest Mg/Ca sample variance (for details see text). Calculations have been performed for four different target levels of SST uncertainty (0.5°C, 1°C, 1.5°C, and 2°C) using intratest and intertest of Mg/Ca variance values equal to those observed for station 2.

that, at the 95% confidence interval (i.e., ±1.96 SE assuming a normal distribution), a sample size of 15 tests corresponds to an uncertainty of 2°C, which is almost identical to the sample size predicted by power analysis for an SST = 28°C (i.e., that appropriate for station 2) and a target uncertainty of 2°C (Figure 9).

[29] Power analysis provides a simple approach to estimate the precision of Mg-paleoseawater thermometry, and offers some advantages over previously employed methods [*Anand and Elderfield*, 2005] in that it incorporates both type I and type II errors and only requires knowledge of the standard deviation of the sample mean Mg/Ca value. The variance and standard deviation can be readily estimated from a few measurements (e.g., 5–10) of individual tests and an appropriate statistical method depending on the nature of the distribution (e.g., normal, skewed, unknown) of Mg/Ca values. For example, the ‘‘jackknife’’ method, which has been previously used to assess the variability of δ^{18}O of planktonic foraminiferal populations [*Schiffelbein and Hills*, 1984], can be applied to nonnormal distributions. The RMLM (residual maximum likelihood method) can be used to estimate the variance of hierarchical data sets such as the profile, chamber and sample mean values developed in this study. We have employed both these methods to calculate the variance and then the target sample size for Mg/Ca core top samples using power analysis. In both cases, we have found the target sample size to be consistent with that calculated assuming a normal Mg/Ca value distribution. Accordingly, the use of these more sophisticated methods might not be required for Mg/Ca–thermometry except where evidence exists for a nonnormal Mg/Ca value distribution.

5. Conclusions

[30] Laser ablation ICP-MS microanalysis reveals large variability of individual foraminifer test Mg/Ca compositions in both core top and plankton pump samples from the eastern Indian Ocean. Despite this variability the mean composition of analyzed tests from each core top sample correlates strongly with average annual SST, and is described by an exponential fit of Mg/Ca to sea surface temperature that is consistent with previously published Mg/Ca–thermometer calibrations for *G. ruber* [*Lea et al.*, 2000; *Dekens et al.*, 2002; *Anand et al.*, 2003; *McConnell and Thunell*, 2005]. Our results demonstrate that the microanalysis of Mg/Ca in individual foraminifera by laser ablation ICP-MS is a valid approach to reconstructing paleo-SST and that results are commensurate with conventional bulk analysis methods.

[31] The large variance of test Mg/Ca compositions tests observed within individual sediment core samples can be partly accounted for by calcification temperature variations that are linked to seasonal (and interannual) changes in sea surface temperature, and also the presence of *G. ruber* morphotypes that possess distinct Mg/Ca compositions because of differing seasonal growth preferences (or habitat depths). The total variance, however, cannot be attributed to calcification temperature fluctuations alone and requires an additional significant and stochastic ‘‘vital effect’’ influence on the Mg/Ca composition into foraminiferal calcite. We suggest this vital effect may be related to the differential development of Mg/Ca banding and possible symbiont activity in *G. ruber*.

[32] Significant care is required when estimating the uncertainty of the mean test and hence also bulk test Mg/Ca compositions for both fossil and live-collected samples. The variance of mean test Mg/Ca compositions is sample specific, and shows: (1) a significant positive correlation with the seasonal range in sea surface temperature, and (2) influenced by *G. ruber* morphotypes where present, which are characterized by consistent differences in Mg/Ca composition. We demonstrate the application of a simple

statistical approach (power analysis), which requires a measure of Mg/Ca intertest variance, to determine the number of tests needed to obtain a prescribed level of uncertainty for SST reconstruction.

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