We would like to thank the three reviewers for their constructive reviews of the previous version of our paper. We have revised the manuscript accordingly and provide our reply to the reviewers' comments below.

We hope that the revised manuscript is now acceptable for publication in *Biogeosciences*.

Yours sincerely,

Lukas Jonkers
on behalf of all authors

In the following we respond to and provide our views on the comments made by the reviewers. First we list their point and then, in italic printing, we directly respond to them.

Response to reviewer 1

Major comments

Reviewer comment, definition of crust: The authors define lower Mg and Mn layers as crust parts. But what is a criterion of low Mg and Mn layers (statistic criteria)? For example, Mg/Ca of N. dutertrei of LGM from SE African margin (Fig. 6 upper panel) shows gradual increases with ablation time and the boundary of inner calcite and outer calcite is not so obvious. Since all discussion is based on the difference between the inner wall and the outer calcite, this point should be clearly defined in the manuscript. On the other hand, the difference of crystal size is mentioned as a feature of crust using SEM image (Fig. 2) but this is very surface information. Further, crystal size is different between trapped material from the Mozambique Channel and the sedimentary tests from SE African coast. Is the relationship between crystal size and low Mg/Ca layer robust?
Author response: *The rough crystalline texture of the crust is characteristic for this type of calcite in an number of planktonic foraminiferal species (e.g. Bé, 1980; Hemleben et al., 1977) and various authors have shown the compositional differences between crustal and lamellar calcite (e.g. Duckworth, 1977; Sautter, 1998). In N. dutertrei, the presence of a crust has been extensively documented using various analytical methods (Sautter, 1998; Eggins et al., 2003; Hemleben et al., 1985). Here we use this correlation between crystal morphology and differences in Mg/Ca and Mn/Ca in the profiles to distinguish between the two layers (this discussion is integrated in section 5, page 8, lines 5-13). The transition between the crust and the inner layer n the LA-ICP-MS profiles may not be razor sharp due to a) surface structures onto the non-crust calcite and b) bottom of the laser pit being not 100% perpendicular to the surface of the chamber (and plane of the crust-non-crust boundary), both leading to a slight mixing of Mg- and Mn-signals of these layers (page 8, lines 13-17).*

Regional crust variability: We observe a difference in crust surface morphology between the two areas, but not in composition. We have clarified this in the revised manuscript on page 11, lines 31-33 and page 12, lines 1-6.

Reviewer comment, comparison sediment trap and sedimentary material: In the introduction, the authors propose a very attractive approach about the comparison between trapped material and sedimentary tests to infer the diagenetic influence on test chemistry. However, the trapped and sedimentary samples came from very different oceanic regions that we discover in the section of "Regional hydrology" (the authors should present a map of studied sites with WOA09 data position). Does this combination of the trapped and sedimentary samples allow estimating diagenetic influence on foraminiferal test chemistry? Why the authors analysed only F-2 chamber of the trapped samples? The information on other chambers of trapped samples will be helpful and be shown in Figs. 4 and 5 (together with core-top).

The sedimentary foraminiferal tests were extracted from 5 cores of which the depth in water column varies from 1626 m to 3076 m (Table 1). Examining the test chemistry in relation to water depth could be an alternative way to deduce test preservation state. Is there any difference of foraminiferal test chemistry among the studied cores? Since the results from different cores and distinct periods are combined for “statistically robust estimate”, detailed information is not accessible in the present manuscript. It would be interesting to compare not only average values but also the size of dispersion.

Author response*: Since we have no core top material and sediment trap material from the same location, we relied on comparing material collected sites relatively close to each other. The hydrographic differences between the two sites are only minor and the waters in the Agulhas Current (i.e. on the South African margin) are for a very large extent sourced in the Mozambique Channel. We are therefore confident that the Mozambique Channel time-series can be used for comparison to samples from the SE South African Margin. We have added this information on page 3, lines 28-31.*

Moreover comparison between foraminifera from the sediment trap and core tops provide us the possibility to assess the potential influence of diagenesis. Small differences in hydrology may slightly affect Mg, Mn, etc incorporation, but such effects are not focussed on in our manuscript. Primarily, the overall patterns in through-wall element distribution are discussed.

Combining the sediment cores. First of all we would like to stress that we did not combine samples of different periods. Unfortunately, for a statistically robust comparison of the dispersion of the data, we have not been able to analyse enough specimen from every location

Reviewer comment, representativeness of the results: It is not clear for me whether presented results represent the whole data sets or only limited data were acquired. The typical example of this confusion is seen in Fig. 9 (comparison of Mg/Ca between two tests from the same sediment trap). The Mg/Ca values of the inner calcite are similar between the two tests whereas the Mg/Ca ratios of the outer calcite are different (Fig. 9). From this observation, the authors suggest that outer Mg/Ca does not reflect sounding seawater temperature. Is the analysis of two tests enough to support this hypothesis? What we observe in Fig. 9 could simply be inter-test variability.

Are the results shown in Figs. 4 and 5 typical examples of intra-test variability or only limited samples were ablated to examine the variability among different chambers? How many specimens are shown in these figures? Why for the core-top data, the results of thickness and elemental ratios are from different samples?

Author response: *The profiles in Figure 9 are examples of inner-wall similarity and crust dissimilarity and act as an illustration of the discussion of section 5.1. This is now added to the text (page 10, lines 30-32) as well as to the figure caption. The conclusion of compositional heterogeneity in crustal calcite is based on more than 2 profiles through two tests (page 5, lines 22-24 and page 6, lines 20-31).*

We have now added how many analyses were performed from all groups of specimens in section 3 page 5, lines 22-24 and the captions of figures 4 and 5. Note that even if the observed patterns are due to inter-test variability, the fact that such patterns are present has implications for understanding of crust formation as well as for temperature reconstruction using crust-forming species.

The intra-test variability reported here (Figs 4 and 5) is based on 50 laser ablation measurements on 3 specimens. The figures show differences between crust and non-crust that are representative for the acquired profiles. This is added to the revised version of the manuscript (page 5, lines 22-24 and figure caption).

Reviewer comment, depth of calcification: I think that the crust thickness variable with chamber is an interesting feature. This discovery is somehow contradictory with the conventional idea that N. dutertrei forms the inner calcite close to the surface and adds crust at deeper depths. The crust observed in this study is rather similar to the part formed by typical lamellar calcification. Then, what is the authors' hypothesis about calcification depth for the inner and outer calcites? Are the assumed calcification depths consistent with WOA09 data once foraminiferal Mg/Ca is converted into seawater temperature?
Author response: *We agree with the reviewer that the depth of calcification of N. dutertrei needs to be constrained, yet any estimate of depth of crust formation (and of the inner layer) based on Mg/Ca temperature depends on the calibration used. However, to verify such an estimate of calcification depth, independent evidence on the depth habitat is necessary. Additionally, we doubt the applicability of the existing calibrations to individual layers and/or chambers and therefore refrain from comparison of calculated temperatures to water column data (see also the new section in the discussion (page 10, lines 9-28) and our response to other comments). Instead, we argue that inferences of the calcification depth based on whole test chemistry overestimate the calcification depth due to the presence of a crust. The pattern in crust thickness does appear similar to the pattern expected for lamellar calcification, but based on the trace element profiles alone it is not possible to draw strong conclusions on the formation mechanism. There could be several reasons for this pattern (such as continued growth at depth discussed on page 11, lines 17-30), but further studies are required to resolve this matter.*

Minor comments
Reviewer comment: When crust thickness trend is compared with Mg/Ca (Figs. 4 and 5 LGM sample), I see some anti-correlation between them. Does this trend exist for other samples? If yes, what is the possible explanation for this correlation?
Author response: *This trend does not appear when taking into account all measured specimens. However, it might be that crust formation continues as the test sinks and therefore that the thickest part of the crust is on average formed deeper (and at lower temperatures) than thinner parts of the crust. We have therefore taken this comment to discuss this possibility in section 5.2 (page 11, lines 25-30).*

Reviewer comment: The implication to other crust-forming species is mentioned but not enough discussed. It will be nice to develop this aspect.
Author response: *We agree and have included additional information on page 9, lines 2-7.*

Reviewer comment: Table 1. What is the depth of trapped material? When there is no notation "a, b, c", the cores were used for which periods and which dating method was used for them?
Table 2. The meaning of numbers of samples is unclear. Do they correspond to specimen numbers or ablation hole numbers?
Author response: *The captions have been clarified. The depth of the sediment trap is 2000 m, which is also indicated in the table.*

Reviewer comment: Comparison with independent whole test analysis (Fallet et al., 2010; Fallet et al., 2012 DSR) will be interesting.

Author response: *We analysed specimens from two collecting cups of the sediment trap; one cup cannot be used for comparison as only 3 shells were measured for this study. For the other cup Fallet et al. (2011) report an Mg/Ca of 3.09 mmol/mol, whereas we observe a whole wall average of 2.88 ± 0.72 mmol/mol (1 σ, F-2 only). Crust and inner wall values are 1.45 ± 0.48 and 3.48 ± 1.06 mmol/mol, respectively. Statistically the two measurement techniques yield indistinguishable ratios, but note that the sample sizes for both types of analysis are unequal and that a different size fraction was used for the present study. We added this on page 8,lines 29-33.*

Reviewer comment: Scale bar in Figure 2
Author response: *We have improved the visibility of the scale bar in Fig. 2C.*

Reviewer comment: Fig. 3. Ca profile might be interesting to show since this element can present CaCO3 density change, thus crust and inner wall distinction.
Author response: *The Ca-count profiles do not show variability that is consistent with crust/non-crust in any of the profiles. Since both the crust and inner layer are made of calcite, only very small changes in density may be expected.*

Reviewer comment: Fig. 9. It will be interesting to show Mn/Ca of the same tests to examine whether Mn/Ca of the inner tests are close to each other and Mn/Ca of the outer calcite is decoupled, as observed for Mg/Ca.
Author response: *We have mentioned the difference in the Mn/Ca profiles between the tests in the figure caption, but we focus on showing Mg/Ca since the figure serves as an illustration of the effect of encrustation (see page 10 lines 30-32) on temperature estimates*

For the "grey" test Mn is very low, close below the detection limit for the crust and often near the detection limit for the inner layer, resulting in an irregular Mn-signal. This means that although there is a difference between the two layers, the signal for the grey test is noisy and hampers comparison. This information has been added to the figure caption.

Response to reviewer 2

Major comments

Reviewer comment: Although the Anand et al., (2003) calibration is applied to the Mg/Ca ratios, it would be more useful to estimate the temperature and convert this to a depth range at which the foraminifera could be calcifying and compare this to the known depth range of the samples. For example, the sediment trap foraminifera should have the seasons constrained, so the Mg/Ca ratios could be used to estimate the depth at a particular month. How do these depths compare to the known calcification depth range of this species. Do they fall within the thermocline?
Author response: *We agree with the reviewer that the variability in Mg/Ca may have a large effect on Mg/Ca-T calibrations for species forming a calcite crust. We have therefore added a new section on Mg/Ca temperature conversion to the discussion (page 10, lines 9-28). See also our response to a similar question of reviewer 3.*

Calculation of seawater temperature from Mg/Ca could easily be achieved. However, due to the intra-test variability in Mg/Ca it is uncertain whether existing calibration based on whole test measurements can be applied to single layer measurements and hence if such temperature estimates are meaningful. Slopes of existing calibrations for N. dutertrei overlap, indicating a similar sensitivity of Mg/Ca to temperature. The pre-exponential constants however, differ and call for independent appraisal of the depth habitat of this species to justify the use of a particular equation (assuming that it is applicable). Also, Bolton *et al. (2011) have shown that the Mg/Ca temperature relationship varies between chambers, particularly the pre-exponential constant, i.e. the absolute temperature values. So whereas the temperature differences between the crust and inner layer calcite might be estimated relatively accurately, the absolute temperatures depend on the calibration used. We have added a discussion on using single-chamber calibrations vs those based on whole test chemistry (page 10, lines 9-28) and its relation to our results.*

Reviewer comment: Is it possible to define what is considered an encrusted sample? Is it the presence of blocky crystals? Can you also determine encrusted forms based on the presence (or absence) of pore pits, (size-normalised) weights, infilling of sutures?
Author response: *We agree with the reviewer and have added a description of morphological characteristics (page 8, lines 23-28). However, encrustation is variable between and within specimens is most easily recognised by the presence of double layering in Mg/Ca.*

Minor comments
All minor comments have been addressed in the revised text. Some require additional clarification, which we provide below.

Reviewer comment: Could the high counting rates at the beginning of the LA profiles be the TE-enriched veneer mentioned in other papers using LA-ICPMS e.g. Eggins et al., (2003)?
Author response: *These could indeed be due to the presence of a layer of veneer as suggested by several authors and have therefore added references accordingly (page 5, lines 15-17).*

Reviewer comment: Also, salinity (line 12 & 13) has no units
Author response: *The calculation of salinity is based on a ratio and the practical salinity scale (PSS) is therefore dimensionless.*

Reviewer comment: Why do the authors think that the Mg/Ca and Mn/Ca layering is absent in some specimens? Is it because they do not have a crust (or minimal crust)? If this is the reason then you need to state this more clearly in the manuscript. If they were identified as crust bearing but this is not resolved in the laser profiles, could this be related to the methodology e.g. could the high power be ablating a potentially thin but low Mg/Ca (and Mn/Ca) crust?
Author response: *Variable encrustation was observed both on SEM pictures and the in the Mg/Ca and Mn/Ca profiles. The exact reason for absence/presence of a crust eludes us, but since we have observed the absence or patchy distribution of encrustation on SEM pictures (Figure 2A), presence of layering in the ablation profiles is not due to an analytical artefact, but reflects real compositional differences within the test walls.*

Response to reviewer 3

Major comments

Reviewer comment: The laser ablation data: Figure 3 details an example laser ablation profile that shows raw data and a 3-pt running mean. The data shown (symbols) in that plot is not very noisy (i.e. the 'spread' of the measurements in Mg/Ca space is small). The other ablation profiles (Figure 6) are quite noisy and the actual LA data points (the symbols) span 10 mmol/mol for some of the profiles (e.g. LGM, F-2, red spots span 1 to 10 mmol/mol) and the data are smoothed using a 25-pt running mean. I think the authors should comment on

why the ablation profiles are so noisy, necessitating a 25-pt mean to smooth the data. The standard error of the measurements on those profiles is likely large. Are the average TE/Ca ratios of the different chambers statistically significantly different given such a large standard error?
Author response: *The reviewer rightly points out that there seems to be considerable variability in Mg/Ca within the layers (Fig. 6). The variability in Fig. 6 appears higher than the overall compositional variability shown in Fig. 3, but the scales of the two figures are very different, giving the erroneous impression of a larger spread in the the Mg/Ca values in Fig. 6. The layering is however present in >70% of the shells and in both Mg/Ca and Mn/Ca, but not in Sr/Ca, suggesting that it is not an analytical artefact. Furthermore, other studies have clearly demonstrated the compositional difference between crustal and inner layer calcite, lending support to our observations.*

The significance of compositional difference between crust and inner layer calcite was tested using a student t-test (two-tailed) on the mean Mg/Ca values. We conclude that, within a 95 % confidence interval, the crust and the inner layer on all chambers, except F-1 in 0101G_D23, are different. This has been added to section 5, page 7-8, lines 29-31 and 1-2 and the caption of Fig. 6.

Reviewer comment: The author's state in the methods section that they chose to analyze the F-2 chamber for all of the samples because 'previous studies suggest the F-2 chamber contains valuable information on the compositional variability of the whole test'. This is true for species like G. ruber and G. sacculifer because there are only 3 chambers in the final whorl (F, F-1, and F-2). If, as a chamber grows, calcite is added to the previously formed chambers, then yes, F-2 would contain a calcite layer formed with the F-1 chamber and the F chamber and therefore the F-2 chamber includes the growth history of the entire adult whorl. However this is not similar to N. dutertrei, which has variable (5+) chambers in the adult whorl. In fact, it is stated in section 4.1 (Crust and element/Ca layering) that sometimes F-2 also didn't include a crust. Additionally, figure 5 shows variable Mg/Ca ratios in the F-0 – F-2 chambers, which appear to asymptote to similar Mg/Ca values in chambers F-3, F-4, and F-5. While this isn't detrimental to the study and results presented here, the results suggest older chambers may be better to focus on for future studies and chamber specific calibrations because the older chambers in N. dutertrei contain more information about the compositional variability of the whole test and the Mg/Ca ratios are similar in the older chambers, especially when comparing the inner calcite layers.
Author response: *The reviewer's suggestion that in future studies the older chambers might be good targets for analysis is valuable. We have added a recommendation as to which chamber(s) paleoceanographic studies may focus on (page 12, lines 14-17).*

Reviewer comment: The author's do not specify which Anand et al, 2003 equation is used and what the implied temperatures. Von Langen, 2005 has a calibration for N. pachyderma and several calibration points for N. dutertrei (which overlap the N. pachyderma calibration curve). The Von Langen, 2005 equation may be more suitable for the Neogloboquadrinids. Additionally, the authors present sediment trap data but do not compare the Mg/Ca-derived temperatures to regional hydrographic data. The Mg/Ca ratios, especially the sediment trap samples, be converted to Mg/Ca-derived SSTs and any offset from the actual SST should be discussed.
Author response: *Reviewer 2 raised a similar comment; please also refer to our response above. We added a paragraph to the discussion to address this issue (page 10, lines 9-28).*

Calibrations using whole shells are unlikely to be applicable to Mg/Ca derived from individual layers within single chambers and we therefore refrain from suggesting that these equations can be applied to our data. Crystallographic (and indeed element) variability *illustrates the fact that the inner layer and crust calcite are precipitated differently, likely resulting in different temperature sensitivities. Moreover, comparison of Mg/Ca derived temperatures to sea surface temperature, as suggested by the reviewer, may not be useful since calcification depth of N. dutertrei is not within the surface mixed layer, but in the thermocline (Field, 2004). Any offset from SST is dependent on the calibration used. All temperature calibrations for this species (e.g. Anand et al., 2003; Dekens et al., 2002; Von Langen et al., 2005) are based on whole shell measurements and thus integrate over a large depth zone. The actual depth(zone) at which the different layers are formed most likely differs from this integrated depth. Therefore, justification for use of a particular calibration depends quantification of the exact depth of formation of the individual layers. Clearly, independent evidence is needed to establish these depths before Mg/Ca values can be compared to ambient water temperatures.*

Furthermore, the sediment trap samples were analysed to check if the layering observed in sediment samples was due to diagenesis (as stated in section 3, page 4, lines 25-28). Sample numbers do not allow a statistically robust estimate of the population mean and hence preclude meaningful comparison.

Minor comments

All minor comments have been addressed in the revised version of the manuscript. Additional response is provided below.

Reviewer comment: Location of the samples

Author response: *The naming of the locations is clarified and a map with the two areas has been added to Fig. 2. As indicated in the table no sediment samples from the Mozambique Channel were presented in the manuscript. All details are presented in table 1.*

Reviewer comment: Aluminum was taken as an indicator of detrital contamination. What Al/Ca ratio did you use to suggest a sample was contaminated? Why can't aluminum/Ca be quantified (see Fig. 3 caption)? Other researchers report Al/Ca ratios (cf. Bolton et al, 2011).

Author response: *Conversion of element concentrations from the raw mass spectrometer counts is usually achieved using ^{43}Ca as an internal standard. The relative abundance of Ca in the contaminant phase may not be equal to 40 % (as in $CaCO_3$) and quantification of the Al concentration is therefore not possible. It is important to note, however, that this does not affect our data (the contaminant part of the profiles are not selected). Even though the absolute El/Ca may be unknown, the anomalous counts for Al still allow recognition of other-than-calcite phases.*

Reviewer comment: Why report median values instead of an average? Comparing the median value in the Holocene vs. LGM samples is also quite meaningless. All you are saying is the middle value hasn't changed. But has the distribution changed? The box and whiskers plots are really useful in showing the change in the distribution of the TE/Ca ratios, but it would be more useful to report the average TE/Ca ratio and not the median value. The average value would be used to calculate SSTs, not the median.

Author response: *We have included the mean values in the revised manuscript. Note however that the differences are small since the data are close to normally distributed and that the median value is less sensitive to outliers and therefore provides a better characterisation of the population.*

Reviewer comment: Sediment trap Mg/Ca. Why was only F-2 analyzed and compared to the core-top data? More data from the sediment trap specimens should be discussed. Is the

chamber-to-chamber and/or inner vs. outer crust TE/Ca variability similar to the core top specimens?
Author response: *As stated before and in the original manuscript, the purpose of analysis of the sediment trap samples was to provide a diagenetically unaltered reference and these samples unequivocally prove that the layering is not due to post-depositional processes. For this reason only F-2 was analysed. The limited amount of specimens analysed does not warrant statistical discussion of these data, but we acknowledge the suggestion for further work.*

Reviewer comment: Evidently crust formation does not occur simultaneously on all chambers. If it did, then the F-2, F-1, and final chambers in N. dutertrei would also always be encrusted and the crust would be uniform in thickness all over. This is not the case and we can infer from this that crust formation in N. dutertrei does not form simultaneously and on all chambers.
Author response: *We have rephrased the sentence (page 9, lines 15-18). However, based on the chemical data we cannot exclude the possibility that the crust formed, albeit not with the same thickness, rapidly and at the same time on the chambers. Literature states that crust formation occurs rapidly, but we agree with the reviewer that our data suggest progressive crust formation (see also page 9, lines 13-24).*

Reviewer comment: Average Mg/Ca ratios of inner calcite, outer calcite, entire chamber, and whole shell values are not converted to Mg/Ca derived temperatures. Are the temperatures accurate for this location using the calibration chosen to calculate temperatures? What is the implied depth range of crust formation, inner calcite formation, etc?
Author response: *For reasons explained above we are hesitant with applying whole-test calibrations to individual layers, since we argue that the exact temperature at which the crust formed cannot be inferred from Mg/Ca. Knowledge of the mechanisms of crust formation is extremely limited, but some evidence exists that Neogloboquadrinids are able to survive at very low temperatures. The inferred temperature from the low Mg/Ca crust can thus not be discarded. Since all temperature calibrations for N. dutertrei indicate the same temperature sensitivity the difference between the crust and inner layer temperature is independent of the calibration. With the assumption that this sensitivity is correct we have included an estimate of the formation depth difference between the two layers (page 11, lines 3-6).*

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Encrustation and trace element composition of *Neogloboquadrina dutertrei* assessed from single chamber analyses, implications for paleotemperature estimates

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Abstract

Crust formation is a common phenomenon in planktonic foraminifera. Because of their different formation mechanism and hence composition, crusts affect the overall test composition and therefore complicate the use of crust-bearing foraminifera in paleoceanography. Such species are often used to estimate subsurface paleotemperatures and although the influence of the crust on the trace element/Ca ratio is recognised, it has not been systematically explored between and within tests. Here we use laser ablation ICP-MS to assess the variability in trace element composition of the crust of *Neogloboquadrina dutertrei* within individual chambers, as well as the effect of compositional heterogeneity of the crust on whole test chemistry. Compositionally, the outer crust differs from inner layer by lower Mg/Ca and Mn/Ca, but is indistinguishable in Sr/Ca. Crust thickness decreases towards the younger chambers and it may be entirely absent from the last chamber. In contrast to Mn/Ca and Sr/Ca, crustal Mg/Ca ratios show a tendency towards higher values on the younger chambers. These patterns in crust thickness and in crust Mg/Ca indicate that temperature is not the dominant factor controlling crust composition. Temperature estimates based on *N. dutertrei*, and presumably other crust-forming species too, are therefore biased towards too low values. Through comparison of modern and glacial tests we show that this bias is not constant and that changes in crust thickness and/or in Mg/Ca values can spuriously suggest temperature changes.

1 Introduction

Many planktonic foraminifera form an outer crust that may account for a significant part of their total test mass (Bé, 1979; Bé, 1980; Duckworth, 1977; Hemleben et al., 1977). Such crusts are formed during the later stages of the foraminiferal life cycle. In some species crust formation is associated with gametogenesis, whereas in others it appears to be triggered by temperature decrease upon sinking of the test (Hemleben et al., 1989; Hemleben and Spindler, 1983). Crust formation occurs rapidly, in less than ~20 hours in culture conditions (Bé, 1980). Since the crust is known to be compositionally different from the inner test wall (e.g. Duckworth, 1977; Eggins et al., 2003), and its thickness varies within and between specimens, it is necessary to understand and quantify the effects of encrustation to reliably use whole test chemistry for paleoenvironmental reconstructions.

Here we assess the inter- and intra-test trace element variability of *Neogloboquadrina dutertrei* (*N. dutertrei*) from modern and glacial samples from the SW Indian Ocean to determine the effects of encrustation on the species' Mg/Ca, Mn/Ca and Sr/Ca ratios. *N. dutertrei* is an subtropical thermocline dweller (Bé and Tolderlund, 1971; Field, 2004), is non-spinose and bears symbionts (Gastrich, 1987). Intra-annual whole-test Mg/Ca and stable oxygen isotopes show considerable variability, but on average were suggested to reflect a depth habitat around 60 m depth, just below the surface mixed layer (Fallet et al., 2011; Bé and Hutson, 1977; Dekens et al., 2002). The species is often used to infer past thermocline conditions (Leduc et al., 2009; Kiefer et al., 2006). However, *N. dutertrei* often develops an outer calcite crust consisting of well-formed blocky calcite crystals (Sautter, 1998; Hemleben and Spindler, 1983). This crust has lower Mg/Ca values than the remaining part of the test and is known to significantly enrich the whole-test $\delta^{18}O$, both suggesting that the crust is formed at lower temperatures deeper in the water column (Sautter, 1998; Fehrenbacher and Martin, 2010; Pena et al., 2005; Eggins et al., 2003; Sadekov et al., 2005). Anecdotal evidence suggests that crust formation in *N. dutertrei* occurs when temperatures drop below 15° C, but very little remains known about crust formation in this species (Hemleben and Spindler, 1983). In order to quantify trace element variability within *N. dutertrei* tests and the influence of encrustation on bulk test composition we present detailed test wall element/Ca profiles of a large number of tests from sediment samples of recent and glacial age from the SW Indian Ocean. This allows for a robust estimate of the effects of intra- and intertest el/Ca variability in *N. dutertrei*.

2 Regional hydrography

Samples were taken from two locations in the SW Indian Ocean: the SE South African continental margin off Durban, and from the narrowest part of the Mozambique Channel. Both areas are characterised by large intra-annual variability in water mass properties due to meso-scale eddy activity and connected since most water that flows out of the Mozambique Channel ends up in the Agulhas current flowing along the SE South African margin (De Ruijter et al., 1999; De Ruijter et al., 2002). Sea surface temperatures off Durban are lower than in the Mozambique Channel by on average ~4°C (Fig. 1A). The mean seasonal amplitude is ~6°C, 1.5°C more than in the Mozambique Channel (Fig. 1A). While the sea surface temperature variability shows a clear seasonal pattern, temperature below the seasonal thermocline shows higher frequency (70-90 days) eddy-related variability (Fallet et al., 2011), which is not resolved in the monthly averaged values (Fig. 1A).

Monthly mean sea surface salinity in the SE South African margin varies slightly around ~35.6, and while average salinities in the Mozambique Channel are lower (35.0), they show a more pronounced influence of the seasonal variation in net precipitation (Fig. 1B). Both locations show a subsurface salinity maximum around 200 m depth, but values are considerably lower in the Mozambique Channel, except below ~750 m (Fig. 1B). Subsurface intra-annual salinity variability is more pronounced in the SE South African margin area, particularly at ~500 m depth (Fig. 1B).

3 Materials and methods

Tests of *N. dutertrei* (355-500 μm) were picked from core-top and last glacial maximum (LGM) age sediments from kasten cores from the SE South African margin (Table 1). Depth of the LGM in the cores was determined from the non crust-forming *G. ruber* stable oxygen isotopes, magnetic susceptibility and lightness data. Modern ages for the core tops were inferred from the same planktonic $\delta^{18}O$ records and substantiated by high ^{210}Pb activities, indicative of ^{210}Pb excess and thus ages <~150 years. ^{210}Pb was analysed through its granddaughter ^{210}Po using α-spectrometry (Table 1). In order to provide a statistically robust estimate of the TE/Ca variability in the Holocene and glacial populations, all sediment samples of the core top and LGM age have been lumped together. Comparison of the two populations is uncompromised since combining the samples affects both populations equally. In addition to the sediment samples, a limited number of tests from a sediment trap from the Mozambique Channel was analysed to provide a reference of diagenetically unaltered material (Table 1; for details on the moored trap see Fallet et al., 2010).

All samples were repeatedly cleaned using short sonication with de-ionised water and methanol prior to analysis (cf. Eggins et al., 2003; Wit et al., 2010). Foraminiferal test minor element and trace element composition was determined using laser ablation inductively coupled plasma-mass spectrometry (LA-ICP-MS) at Utrecht University. The system consists of a 193 nm laser (GeoLas 200Q Excimer) coupled to a sector field ICP-MS (Element2, Thermo Scientific). Laser spots were 40-80 μm in diameter; repetition rate was set to 7 Hz and laser power density to 1 J/cm^2. The mass spectrometer was run in low-resolution mode and masses monitored were ^{24}Mg, ^{26}Mg, ^{27}Al, ^{43}Ca, ^{44}Ca, ^{55}Mn and ^{88}Sr. Measurement routine took 0.34 seconds to cycle through the masses. Calibration was performed against a SRM NIST610 glass standard ablated at a higher energy density (~5 J/cm^2) between every 12 foraminiferal samples. Simultaneous ablation of a calcite crystal (Iceland spar) at the lower energy density settings allowed for calculation of the analytical uncertainties of the ICP-MS, which are 0.06, 0.04 and 0.07 mmol/mol (1 σ) for Mg/Ca, Mn/Ca and Sr/Ca respectively (Dueñas-Bohorquez et al., 2011).

Data reduction was done using the SILLS (Signal Integration for Laboratory Laser Systems) software package (Guillong et al., 2008). Raw counts of Mg, Mn, Sr and Ca were converted to concentrations using ^{43}Ca as internal standard. Aluminium was taken as an indicator of detrital contamination. Measurements were performed on intact tests from the outside inwards. High counting rates at the start of ablation through the test wall are not included in the integrated ablation profile (Eggins et al., 2003; Hathorne et al., 2009). To provide an indication of the effect of intra-test Mg/Ca variability, calcification temperature estimates were obtained using the following equation: $T\ (°C) = 11.11 \times \ln\ (Mg/Ca \times 2.92)$ (Anand et al., 2003). Applicability of a whole test calibration to single chamber data presented here is discussed in section 5.1.

To assess intra-test variability (multiple) measurements were conducted per chamber on a limited number of specimens (n = 9, 97 profiles) from the SE South African margin. Previous work has shown that antepenultimate chamber (F-2) may yield valuable information on the compositional variability of the whole test (Eggins et al., 2003). The inter-test variability was therefore obtained from measurements of this chamber of >100 individual tests from both core-top and LGM samples (Table 2). The sediment trap specimens (n = 15) were analysed on F-2 only. Since ablation time and pit depth have been shown to be linearly related (Hathorne et al., 2003), test wall thickness was estimated from ablation time.

4 Results

4.1 Crust and trace element/Ca layering

The presence or absence of a crust consisting of blocky calcite crystals is not readily visible by conventional binocular light microscopy. However, scanning electron microscope (SEM) pictures clearly show the rough crystalline texture of the crust (Fig. 2). The size of the crystals and/or the presence of the crust vary between the chambers, with the final and penultimate chambers (F0 and F-1) often having a relatively thin crust or lacking one at all (Fig. 2A). The distribution and thickness of the crust over the chambers is heterogeneous. Sediment trap and sediment samples from the Mozambique Channel show a crust consisting of smaller crystals than those from the SE South African margin (Fig. 2c).

The ablation profiles of many tests show a pronounced two-layered structure of the test wall in most chambers (Fig. 3). Mg/Ca and Mn/Ca are often lower in the outer layer (in F-2 the mean difference between the two layers in core-top samples is 1.6 mmol/mol and 7.5 μmol/mol respectively), whereas Sr/Ca are relatively constant throughout the chamber wall (Fig. 3). The changes from low to high Mg/Ca and Mn/Ca coincide with each other. This layering is observed in the F-2 chambers of 81 % of the sediment trap specimens, 72 % of the core top and 85 % of the LGM tests.

4.2 Intra-test variability

The thickness of both the inner and outer layer shows a consistent pattern over the chambers. Typical shells have walls that are thickest on F-4 and thin progressively towards younger (F-3 to F0) and older (F-5) chambers (Fig. 4). The outer layer is always thinner than the inner one, but the thinning pattern in both layers is similar (Fig. 4). Double layering is not observed on all chambers; it is often absent from F0 and F-1 and occasionally also from F-2.

In the outer layer Mg/Ca values show an increase towards F0 that is clearest in the last two to three chambers (Fig. 5A and 6). This pattern is not always present in the inner layer (Fig. 5A). Mn/Ca ratios in the outer layer are always lower than in the inner layer (Fig. 5B). Although Mn/Ca values are variable in both layers, no systematic inter-chamber trend is detectable. Sr/Ca shows very little variability throughout the test wall and between different chambers (Fig. 5C).

4.3 Inter-test variability and Holocene-LGM changes

4.3.1 Composition

Mg/Ca in the inner layer of core top specimens varies between 1.2 and 5.0 mmol/mol around a mean value of 2.7 mmol/mol (Fig. 7A). The median Mg/Ca in this layer does not differ from those of specimens from the LGM, but the variability increases considerably (interquartile range, IQR, increase: 0.6 mmol/mol; Fig. 7A). Core top mean Mg/Ca in the outer layer is 1.1 mmol/mol and shows less variability (IQR: 0.4 mmol/mol) than the inner layer (Fig. 7A). In contrast to the inner layer, the outer layer shows a 0.3 mmol/mol lower mean and median Mg/Ca in the LGM, while variability in Mg/Ca of the outer layer is similar in recent and the LGM specimens (Fig. 7A).
Core top Mn/Ca mean values of the inner and outer layers are 12.7 and 5.2 μmol/mol, respectively (Fig. 7B). Both variability and mean values in the inner layer were higher during the LGM, but this pattern is less clear in the outer layer (Fig. 7B).
The Sr/Ca in both layers in the core tops differ very little and both layers have higher Sr/Ca in the LGM samples (Fig. 7C). However, the increase in Sr/Ca in the LGM samples is slightly larger in the outer layer (0.07 mmol/mol on average; Fig. 7C).

4.3.2 Outer layer thickness

Core top average outer layer ablation time (~wall thickness) varies between 20 and 25 seconds. In only very few tests (~ 1%) the outer layer makes up more than 55 % of the test wall thickness (Fig. 8A). LGM tests have thicker outer layers that make up a small, but significantly higher proportion of the test walls (student t-test on the means, 99 % confidence level; Fig. 8B).

5 Discussion

SEM pictures of *N. dutertrei* from the SW Indian Ocean show that many tests are (partly) covered by blocky calcite crystals and Mg/Ca and Mn/Ca profiles through these tests with a blocky calcite surface reveal a pronounced double-layered structure of the shell walls (Fig. 2, 3 and 6). The outer layer is characterised by lower Mg/Ca and Mn/Ca ratios when compared to the inner layer. This outer low Mg and Mn layer is not distributed uniformly over the chambers and several profiles show variability in the composition of both layers. Whether this variability reflects finer scale additional layering remains to be investigated, but it does not affect the net compositional difference between crust and inner layer calcite (Fig. 6). The heterogeneous distribution of the low Mg/Ca and Mn/Ca on the individual chambers is similar to the presence of blocky crystals observed on the outside of the test (Fig. 2).

The rough crystalline texture of the outer layer is characteristic for crustal calcite in a number of planktonic foraminiferal species (e.g. Bé, 1980; Hemleben et al., 1977) and various authors have shown compositional differences between crustal and lamellar calcite (e.g. Duckworth, 1977; Sautter, 1998). In *N. dutertrei*, the presence of a crust has been extensively documented using various analytical methods (Sautter, 1998; Eggins et al., 2003; Hemleben et al., 1985). Here we use this correlation between crystal morphology and differences in Mg/Ca and Mn/Ca in the profiles to distinguish between the two layers (cf. Sautter, 1998; Fehrenbacher and Martin, 2010; Kozdon et al., 2009; Sadekov et al., 2005). The transition between the crust and the inner layer n the LA-ICP-MS profiles may not be razor sharp due to a) surface structures onto the non-crust calcite and b) bottom of the laser pit being not exactly perpendicular to the surface of the chamber (and plane of the crust-non-crust boundary), both leading to a slight mixing of Mg- and Mn-signals of these layers. Others have described the layering in foraminiferal test walls as primary or lamellar ontogenetic calcite on the inside and a gametogenic calcite layer, including a crust, on the outside (e.g. Erez, 2003; Hemleben et al., 1977). Here we define the layers based on compositional differences and to avoid alluding to their formation process we refer to the outer, low Mg and Mn layer as the crust.

Recognition of encrusted specimens using normal light microscopy is not straightforward, although Sautter (1998) mentions that presence of clearly visible micropores is a good criterion to distinguish non-encrusted specimens. However, light microscopy (and SEM imagery) only offers information on the presence, but not on the degree of encrustation. Micro-scale analysis thus remains the most reliable way to determine the extent of, and correcting for encrustation.

The micro-scale results presented here are indistinguishable from conventional ICP-MS data on the same sediment trap sample (Fallet et al., 2011). For the same collecting cup Fallet et al. (2011) report a Mg/Ca value of 3.09 mmol/mol; we determine a whole-wall mean of 2.88 ± 0.72 mmol/mol and crust and inner layer Mg/Ca values of 1.45 ± 0.48 and 3.48 ± 1.06 mmol/mol, respectively. This presence of double-layered tests in samples from the sediment trap excludes the possibility that the element patterns in the test walls are caused by diagenesis (Pena et al., 2005). Moreover, similar variability in crust Mg/Ca and in crust thickness between chambers has been observed in specimens of *N. dutertrei* from the SE Indian and Atlantic Oceans (Eggins et al., 2003; Fehrenbacher and Martin, 2010), but also in other deep-dwelling species such as *G. truncatulinoides* and *G. inflata* (Duckworth, 1977; Hathorne et al., 2009), indicating that the observed pattern is not exclusive to our samples nor to *N. dutertrei* alone.

The decrease in thickness of the inner layer with younger chambers (Fig. 4) agrees well with typical lamellar calcification in which a layer of calcite is precipitated onto existing chambers with every chamber added (Erez, 2003 and references therein). Surprisingly, crust thickness follows a similar pattern, being thinnest on chambers formed later (Fig. 4), thereby resembling variability in wall thickness due to lamellar calcite growth. The observed increase in Mg/Ca of the crust towards the younger chambers would then suggest upward migration of this species during crust formation. This suggestion of gradual crust formation is in contradiction with laboratory studies that showed that crust formation occurs rapidly and simultaneously for all chambers (where crust formation takes place) when the foraminifer descends to colder waters near the end of its life cycle (Hemleben and Spindler, 1983; Bé et al., 1979; Hemleben et al., 1985). Furthermore, *N. dutertrei* is thought to descend in the water column as it ages (e.g. Hemleben et al., 1989), and therefore the inter-chamber increase in crust Mg/Ca towards the final chamber is unlikely to be related to decreasing seawater temperatures under which those chambers form. This has important implications for the application of the species' Mg/Ca to reconstruct past seawater temperature.

Although limited data is available, seawater manganese concentrations in the Indian Ocean distant from the oxygen minimum zones in the Arabian Sea, generally show a maximum in the upper ~200 m and very little change below these depths (Morley et al., 1993). The low Mn/Ca in the crust therefore confirms its precipitation deeper in the water column than the inner layer. The relatively uniform Mn/Ca of the crust across chambers indicates that crust formation took place entirely below the surface Mn maximum. This too renders it unlikely that variability in crust Mg/Ca reflects upward migration through the water column.

5.1 Implication for paleotemperature estimates

Since the crust makes up a considerable part of the total calcite mass, temperature reconstructions based on whole test Mg/Ca values of crust-forming foraminifera only partly reflect temperature. The potential error associated with this variability in Mg/Ca may be enhanced through preferential preservation of the low Mg/Ca crust in the sediments, but importantly also during conventional reductive cleaning. When Mg/Ca values are converted to temperature the bias towards low temperature will be enhanced by the logarithmic nature of the calibrations.

Several calibrations exist for *N. dutertrei* (Anand et al., 2003; Von Langen et al., 2005; Dekens et al., 2002). These calibrations are based on whole test measurements and have overlapping exponential constants, i.e. an equal sensitivity to temperature. Whether these can be applied to single layers from individual chambers has not been tested, but some evidence suggests that this is unlikely to be the case. The nearly identical range in Mg/Ca in both layers (Fig. 7) would suggest that both layers were formed under a similar temperature variability regime. This is unlikely if the crust is formed deeper in the water column where temperature variability is lower than in the thermocline (Fig. 1). A crust-only calibration may therefore have a steeper slope. If, on the other hand, the crust were formed at a depth similar to the inner layer, the different composition of the crust would require a calibration with a higher pre-exponential constant to account for the difference in Mg/Ca values. In both cases a different calibration is needed to convert individual layer Mg/Ca to temperature. Additionally, Bolton et al. (2011) have shown that for *G. ruber* Mg/Ca temperature calibrations differ for individual chambers. The choice of a particular calibration can thus not be justified in the absence of independent determination of the depth of formation of both layers. For this reason we refrain from comparing inferred temperatures to seawater values and although the inferred temperatures differences may be more robust than absolute temperatures, the values presented here should be taken as indicative only.

Both thickness and Mg/Ca of the crust influence whole shell Mg/Ca value and consequently bias temperature estimates from whole-test measurements. Figure 9 depicts the Mg/Ca values converted to temperature through the test wall of two specimens from the same sediment trap sample that clearly show both effects. Both tests settled within the same 3-week period and therefore calcified under nearly identical conditions. This is reflected in the identical Mg/Ca values of the inner layer.

The Mg/Ca of the crust and the ratio of crust to inner layer calcite is however very different for both tests, causing the temperature estimates based on the entire profile to differ by over 2° C (Fig. 9). If the temperature sensitivity of the calibration(s) holds for the individual layers, the Mg/Ca contrast between the crust and the inner layer would amount to>10 °C, indicating a formation depth difference of several hundreds of meters.

The effect of encrustation on whole-test Mg/Ca is also clear in the sediment samples from the SE South African margin. The crust is considerably lower in Mg/Ca, the equivalent of >3° C, in LGM specimens (Fig. 7A). Together with the slight increase in the relative proportion of the crust (Fig. 8B) this lowers the average composition of the entire wall and - by extension - of the entire test (Fig. 7A). The lower LGM whole-test Mg/Ca ratios suggest a lowering of thermocline temperatures, but only through detailed (LA-ICP-MS) analysis it becomes obvious that this cooling is entirely due to a Mg/Ca decrease of a slightly thicker crust and hence is unrelated to actual changes in temperature.

5.2 Potential controls on crust heterogeneity

The pattern of progressive increase in thickness and Mg/Ca ratios of the crust of *N. dutertrei* could suggest that crust formation occurs gradually and over a prolonged period. It appears that thicker crusts have lower Mg/Ca, perhaps as a result of formation deeper in the water column (Fig. 4 and 5). Laboratory studies have shown that species of the genus *Neogloboquadrina* are capable of adopting a benthic life style and surviving for a prolonged period in cold and dark conditions (Hemleben et al., 1985). Hemleben et al. (1977) also mention gradual crust formation in *G. menardii*, but information on the duration of the crust formation process is not available. The thicker crusts with larger crystals on chambers F-4 to F-2 would indeed be consistent with continued growth deeper in the water column and potentially at the sea floor. Yet the Mg/Ca data do not show a simultaneous Mg/Ca increase through the crust that one would expect from continued growth during descent through the water column. On the contrary, crustal Mg/Ca is often lowest at the boundary between the outer and inner layer (Fig. 6).

The difference in crystal size of the crust of samples collected in the Mozambique Channel and those on the SE South African margin could suggest that there are regional differences and thus environmental parameters influencing crust formation

(Fig. 2B and C). Temperature is lower and salinity higher in the upper 100 m of the water column in SE South African margin area, but it is not clear if and how this could account for the different crust morphology. Importantly, samples from both locations show the same layering in Mg/Ca and Mn/Ca, so while there may be hydrographic controls on the crust morphology the composition appears independent of water column conditions.

5.3 Holocene-LGM changes

The inner and outer layer median Mg/Ca in modern *N. dutertrei* tests differ by ~1.7 mmol/mol (Fig. 7A), which translates in a temperature difference of ~10° C. However, to obtain a reasonable estimate of paleotemperature based on the Mg/Ca of the inner layer of *N. dutertrei* a new calibration is needed. Such an inner wall Mg/Ca calibration, perhaps chamber specific, will have to take into account the significant (and apparently random) intra-test variability. Future studies should preferably target the older chambers, as these appear to integrate most of the (life) history of the test. However, rigorous (re)calibration of the species' Mg/Ca-temperature relationship would be the only way to avoid the bias from encrustation.

The Mg/Ca of the inner layer likely provides a more reliable paleothermometer than whole-test Mg/Ca. The similarity between the core top and LGM inner wall Mg/Ca (Fig. 7A) is striking since nearby temperature records based on surface dwelling foraminifera and alkenones do show lower LGM temperatures, particularly in winter (Levi et al., 2007; Bard and Rickaby, 2009). An explanation for the near constant Mg/Ca ratios is that (sub)thermocline temperatures did not change or, that *N. dutertrei* adjusted its depth or seasonal habitat to remain in a certain temperature range.

Both crust and inner layer show elevated Mn/Ca ratios in the LGM samples; the inner layer, supposedly formed higher in the water column, more so than the crust (Fig. 7B). These Mn/Ca may be used to infer changes in seawater Mn concentration due to variable terrigeneous input (Klinkhammer et al., 2009). Culture studies with benthic foraminifera have shown that Mn uptake linearly relates to Mn concentration in the seawater (Munsel et al., 2010). Therefore, the observed (near) doubling in the median Mn/Ca in the inner layer thus may suggest a twofold increase in Mn concentration in the ambient seawater, likely related to enhanced terrigeneous input via dust or lower sea level. The smaller increase in crustal Mn/Ca indicates that the dissolved Mn increase during the LGM was most pronounced in the surface waters (Fig. 7B).

The nearly constant Sr/Ca in through the entire test walls contrasts with Mn/Ca and Mg/Ca. Hathorne et al. (2009) and Eggins et al. (2003) report similar behaviour of strontium, also in other crust-bearing species. The similarity of the Sr/Ca in both layers, which are precipitated under different environmental (temperature, salinity, pH etc.) conditions, suggests that Sr uptake in *N. dutertrei* does not depend on those factors and is biomineralised in a unique way (Lea et al., 1999). The approximately 10 % increase in Sr/Ca in the LGM shells (Fig. 7C) is in accordance with other reconstructions based on the genus *Neogloboquadrina*, and probably too large to be caused by an increase in the oceans' Sr inventory alone (Stoll et al., 1999; Elderfield et al., 2000).

6 Conclusions

Detailed trace element profiling using LA-ICP-MS of tests of *N. dutertrei* demonstrated that the outer crust of blocky calcite crystals is compositionally different from the inner test wall. In the antepenultimate chamber of core top and LGM samples median Mg/Ca and Mn/Ca are 1.7-2.0 mmol/mol and 8-19 µmol/mol lower in the crust than in the inner layer. Sr/Ca ratios are invariant through the entire wall.

Over 70 % of the analysed tests have such a crust on chamber F-2, but the crust is not homogeneously distributed over the individual chambers. There is a clear thinning of the crust towards younger chambers and a crust is often absent from the ultimate and penultimate chambers. Mg/Ca ratios also show a pattern over the different chambers and increase in the last formed chambers. Both patterns in thickness and composition point at biological control on crust formation and composition, impacting the use of the species' Mg/Ca to reconstruct past seawater temperature.

We have illustrated this temperature bias using modern and LGM samples. Modern tests grown under very similar conditions show clearly different crust composition and crust to inner layer ratio, causing considerable whole test Mg/Ca differences. Tests from LGM samples tend to have more and slightly thicker crusts, yielding the Mg/Ca bias due to encrustation greater. In the SW Indian Ocean decreased bulk test

Mg/Ca in LGM age samples is caused by changes in the crust alone. This highlights the need for detailed analyses, where it is possible to separate the crustal from the inner layer and calls for better understanding of the mechanisms of crust formation in order to improve paleotemperature estimates based on the Mg/Ca of encrusted foraminifera.

Acknowledgements

We thank Helen de Waard for assistance with LA-ICP-MS and Nuria Casacuberta and Pere Masque for the ^{210}Pb analyses. The work described in this paper and the research leading to these results has received funding from the European Community's Seventh Framework Programme FP7/2007-2013 – Marie-Curie ITN, under grant agreement n° 238512, GATEWAYS project and the Netherlands NWO programme Inatex. The constructive comments of three anonymous reviewers are thankfully acknowledged.

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Table 1. Location of cores and moored sediment trap

Name	Lat (°S)	Lon (°E)	Depth (m)
CD154-01-01K	29.29	33.14	1997
CD154-02-03K[1]	29.06	32.77	1626
CD154-03-05K	29.12	32.89	1747
CD154-05-07K[1]	29.93	33.82	1850
CD154-10-10K/06P[1,2]	31.17	32.15	3076
MOZ-2 A-06/08[3]	16.42	40.85	2000

[1] ^{210}Pb excess; [2] core top: 10K/LGM: 06P, other cores were used for both modern and LGM; [3] sediment trap

Table 2. Numbers of specimens analysed

	Analysed	Low Al/Ca	Double layered	% double layered
Core top	205	121	87	72
LGM	218	178	152	85

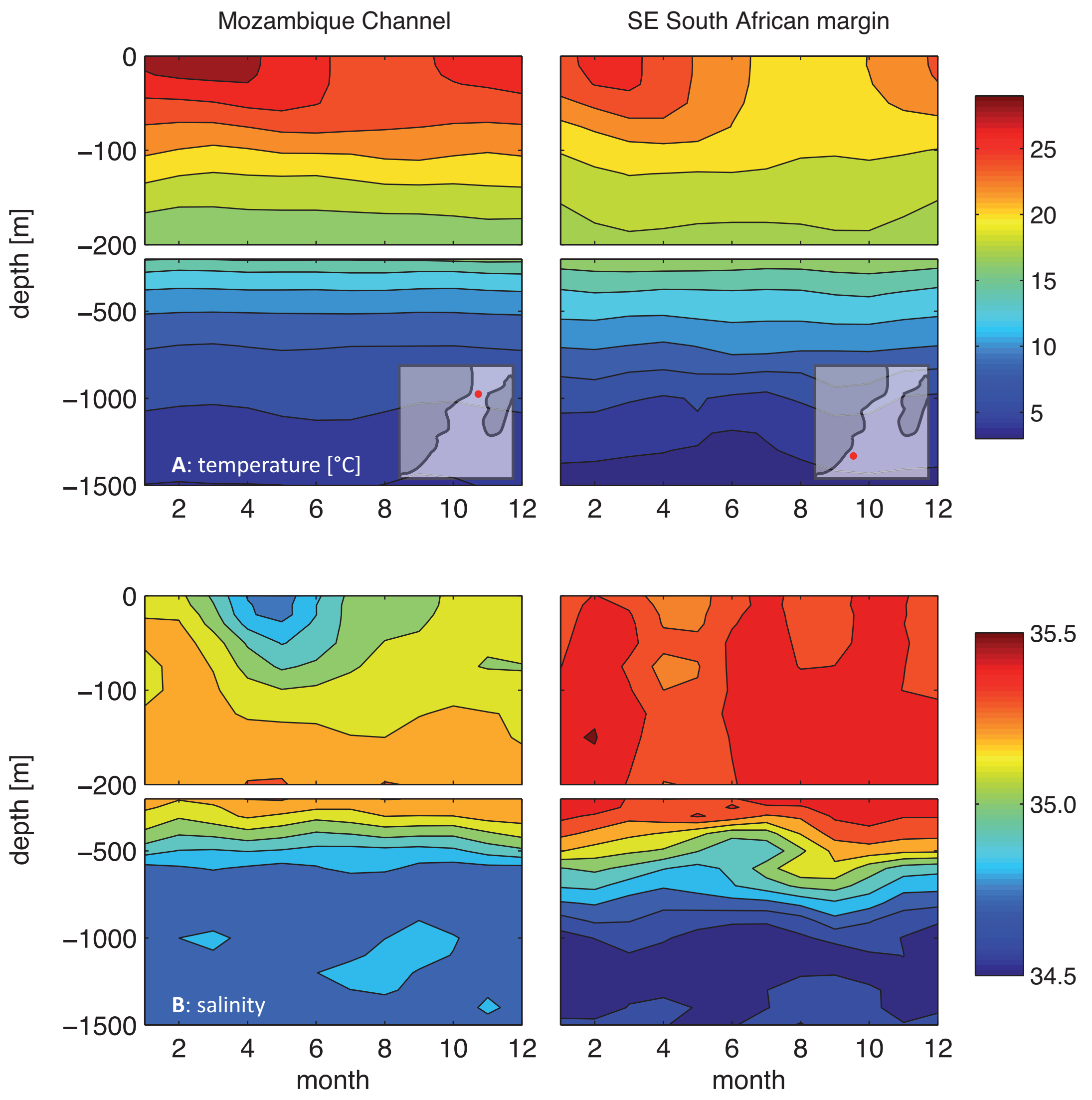

Fig. 1: Monthly averaged temperature (A) and salinity (B) for the Mozambique Channel and SE South African margin sites. Map insets show location of the sites (see also table 1). Note the difference in scale for the upper 200 m. Data from World Ocean Atlas 2009 from the ¼ degree grid cells closest to the sites; the SE South African Margin panels show averaged values for all sites (Antonov et al., 2010; Locarnini et al., 2010).

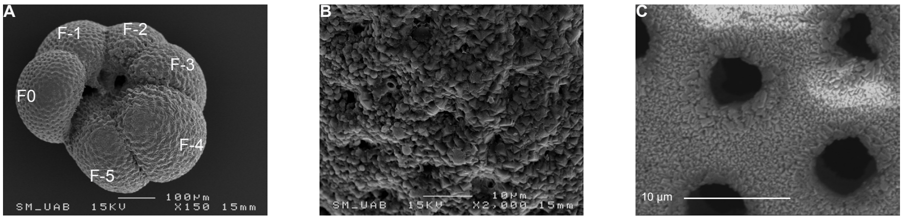

Fig. 2: SEM pictures of *N. dutertrei*. A: overview of entire test from LGM sediment with the naming of the chambers indicated. Note the large crystals on F-2 to F-5. B: close up of the crust morphology of a LGM test. C: close up of the crust morphology of a test from the sediment trap. No cleaning was applied prior to SEM photography.

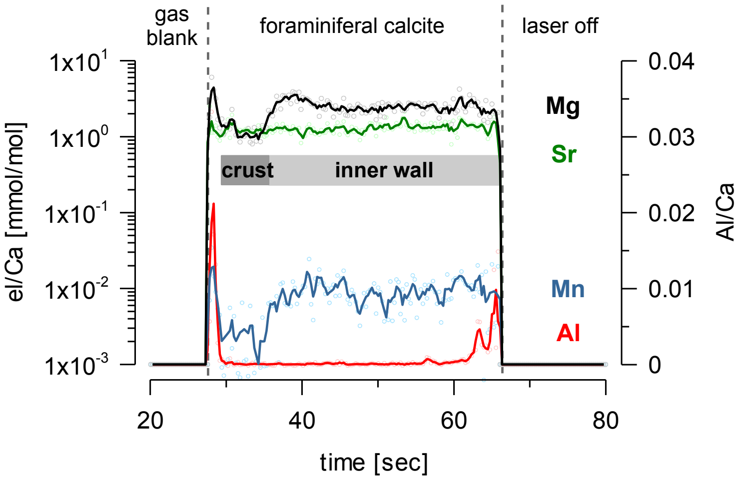

Fig. 3: element/Ca ratios through chamber F-2 of *N. dutertrei* from the Mozambique Channel sediment trap. Thick lines represent 3-point running means. The double layering is highlighted with grey bars. The high values at the beginning of the ablation profiles are ignored in the analyses. The Al/Ca shows presence of a sediment infill of the test, this part of the profile is therefore not taken into account when calculating the average values (despite the limited effect of the infill on other el/Ca ratios). As Al cannot be quantified absolutely, Al/Ca ratios are expressed as raw counts and therefore unit less.

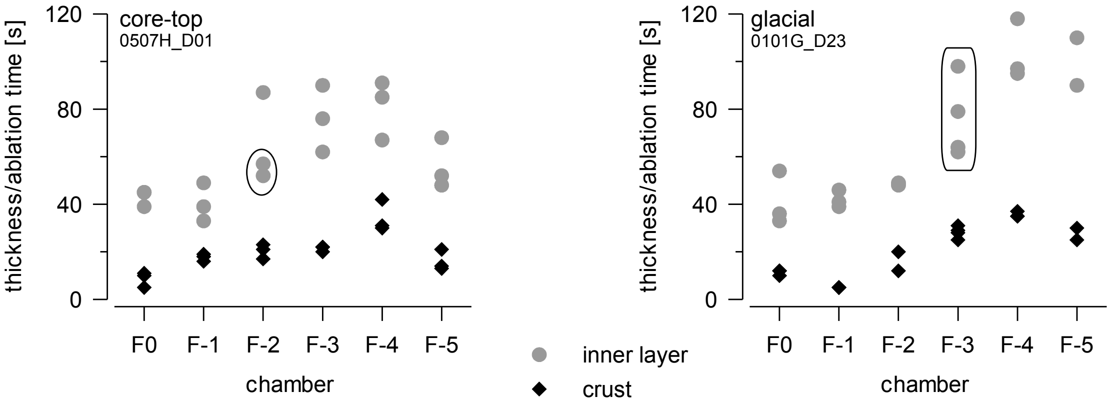

Fig. 4: Intra-test layers thickness pattern in *N. dutertrei*, examples from a core top (left) and an LGM (right) specimen from sediments from the SE South African margin. Each pair of a grey dot and a black diamond represents a single measurement. Encircled dots are minimum values only, in cases where the wall was not completely ablated. For chamber coding see Fig. 2.

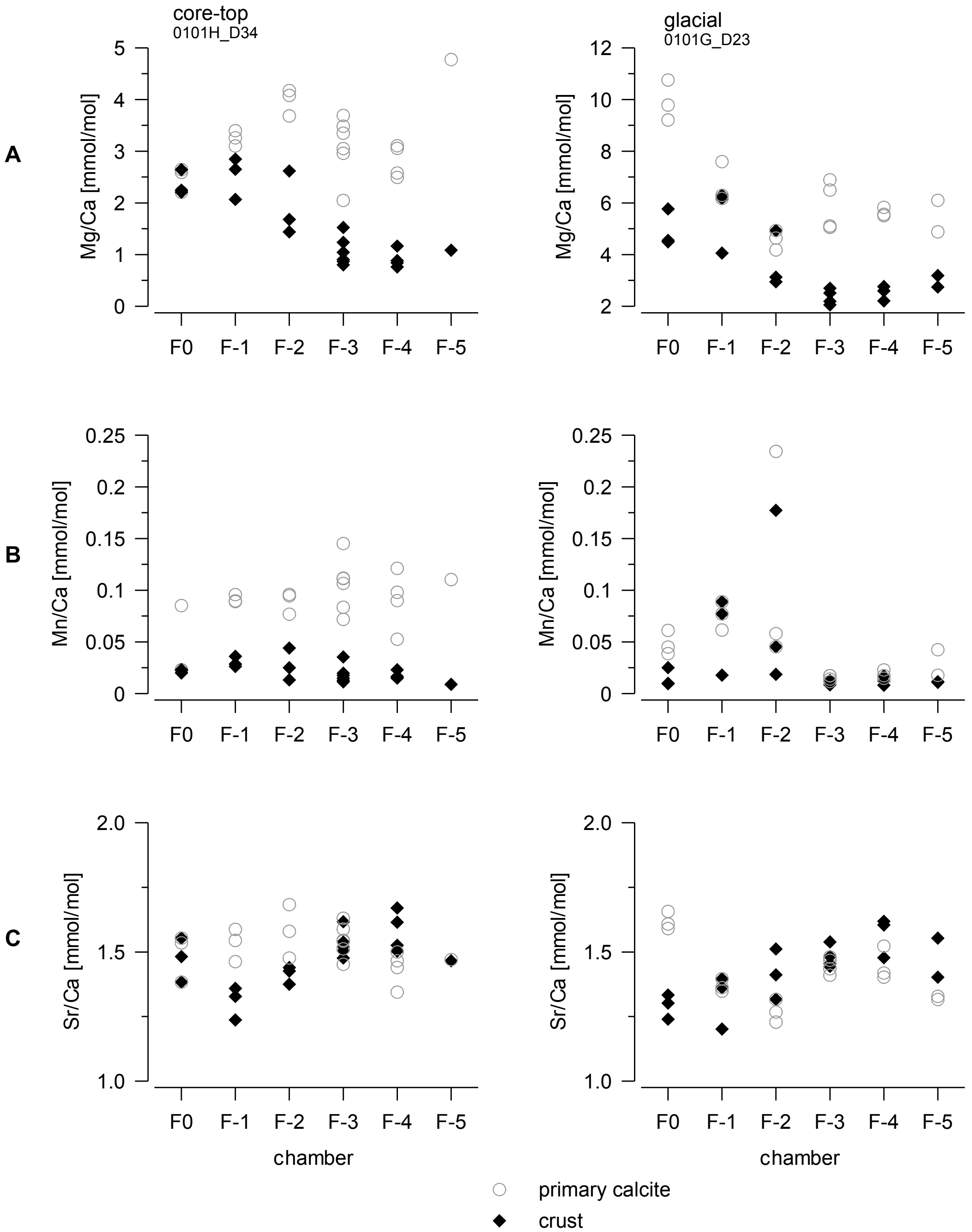

Fig. 5: Intra-test trace element variability in *N. dutertrei*, examples from a core top (left) and an LGM (right) specimen from sediments from the SE South African margin. A: Mg/Ca; B: Mn/Ca and C: Sr/Ca ratios for individual chambers. Symbols as in Fig. 4.

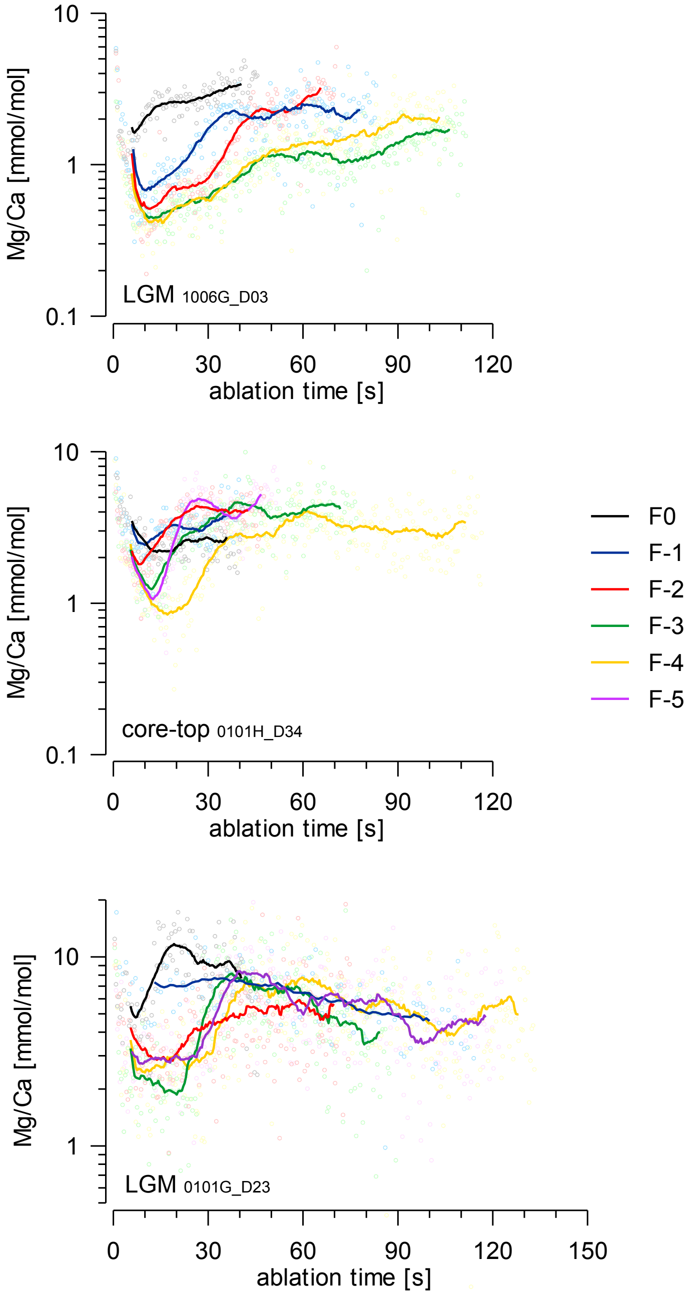

Fig. 6: Mg/Ca profiles through individual *N. dutertrei* chambers from SE South African margin sediments, both core-top and LGM age. Dots represent raw data, thick lines 25-point smoothed averages that highlight the double-layered structure of the test walls. Note differences in the scale of the y-axes. The difference between the layers is statistically robust at the 95 % confidence interval (student *t*-test on means).

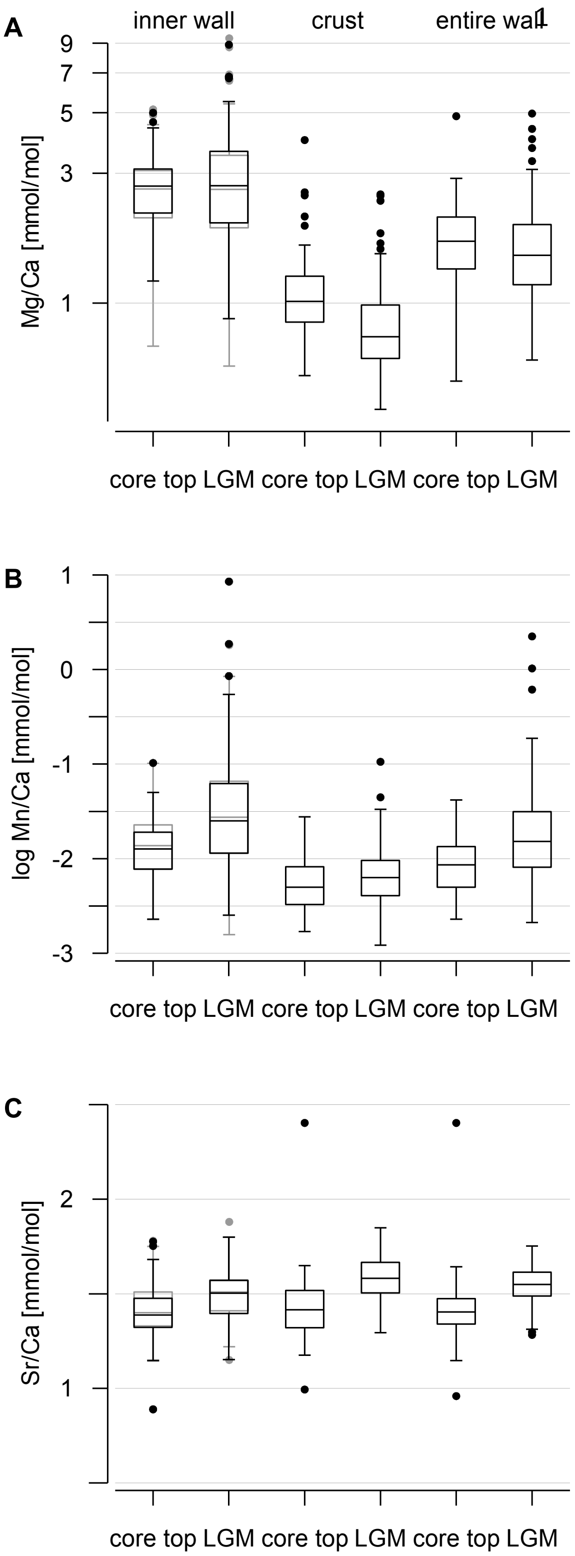

Fig. 7: Core top-LGM element/Ca changes in F-2 of *N. dutertrei* in crust and non-crust calcite. A: Mg/Ca; B: Mn/Ca and C: Sr/Ca. Horizontal lines show median values, boxes interquartile ranges and bars the interquartile ranges multiplied by 1.5. Values outside these ranges are considered as outliers (dots). The grey boxes in the

inner wall panel show the dispersion in all samples, i.e. including those without double layering.

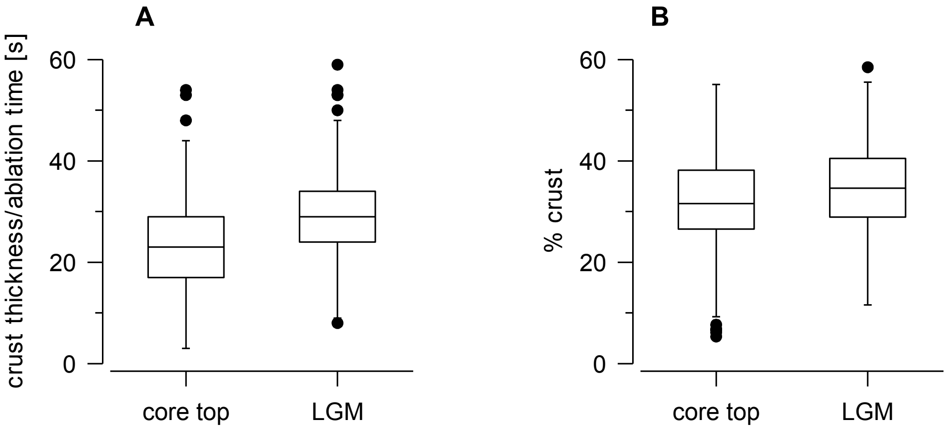

Fig. 8: Core top-LGM crust thickness (A) and crust proportion (B) changes in F-2 of *N. dutertrei*. Symbols as in Fig. 7.

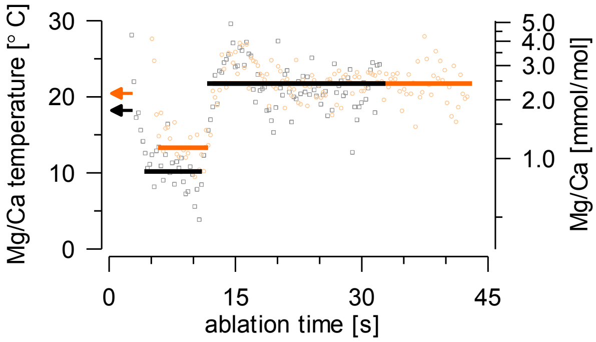

Fig. 9: Illustration of the influence of crust heterogeneity on temperature estimates. Calculated Mg/Ca-temperatures through the walls of two tests from the same sediment trap bottle (colours denote different tests). Thick lines show average crust and inner layer values and arrows the whole wall mean. Mn/Ca profiles through the two tests are close to the detection limit, but appear different.