Response to Referees and short comments and Revised Manuscript

We provide herein a detailed point-by-point response to all referee comments and short comments and we specify the changes made in the revised manuscript. The response to the comments is structured as: (1) comments from Referees, (2) author's response, and changes in manuscript. We include a marked-up manuscript version showing the changes made (using track changes in Word).

Change to MS: we modified the previous title as follow "*Microfossil evidence for trophic changes during the Eocene-Oligocene transition in the South Atlantic (ODP Site 1263, Walvis Ridge)*" because it better reflects the conclusions. We used "microfossil" since we studied both planktonic and benthic foraminifers and nannoplankton.

Response to Referees and short comments

Short comments by P. Pearson

Comment 1) The first issue relates to the isotope shift and the term 'Oi1'. […] In my opinion, the several ambiguities associated with the term Oi1 are not conducive for clarity in discussion hence I recommend abandoning the term Oi-1 by which I mean no disrespect to the work of Miller et al. (1991) which is one of the seminal papers in the field.

Response: It is true that some confusion and misuse of the term Oi-1 occurred in the text. This was also related to a graphical mistake in the figures: the placement of the term "Oi-1" was intended to highlight a 400-kyr interval (rather than an "event"), including the positive peak in δ^{18}O. We recognize that this use of "Oi-1" in the figure leads to misunderstanding and that it does not add more information to the figure. For this reason we omitted it from the revised figures. We adopted the use of Step 1 (first step of the cooling) and Step 2 (the second and more marked step of the cooling), as used also in Pearson et al. (2008) and Bohaty et al. (2012).

Change to MS (with reference to the lines in the marked-up version): We clarified this definition in the text as follow *"To avoid confusion with previous definitions of these two steps, here we follow Pearson et al. (2008) and Bohaty et al. (2012): thus, Step 1 which is the first δ^{18}O step related to a global cooling with a modest ice growth component, and Step 2 which is the second step representing the major ice growth leading to a continental scale ice sheet over Antarctica (Miller et al., 2009)."* (**Lines 93-97**).

Comment 2a) Correlating the EOB. […] Given the fact that the *Hantkenina* / *Cribrohantkenina* extinction occurs clearly and sharply between the steps in Tanzania (Pearson et al., 2008), my suspicion is the EOB is best correlated to Site 1263 using a combination of the Tanzania constraint and the isotope stratigraphy to about 93.5 mcd. The lack of *Hantkenina* fragments in the few metres below this could be sampling. A formal correlation to Site 522 would be an interesting exercise.

Response: We agree that the position of the boundary based only on the Top of *Hantkenina* may not be correct in this case. Indeed, *Hantkenina* has been affected by dissolution, and placing the top just on the basis of the spines presence may not be reliable. On the other hand, we cannot rely on the isotope stratigraphy because at this site the $\delta^{18}O$ data available record clearly just Step 2. We can say that the boundary should be placed in between these two datums: above the Top of *Hantkenina* and below the peak in $\delta^{18}O$ (Step 2; 93.4 mcd). An indication that the boundary could be placed in between these two depths comes also from the acme of *Cl. subdistichus*, which occurs just above the boundary (Agnini et al., 2014), and at Site 1263 at 96.92 mcd (well below the Step 1 defined by Peck et al. 2010 at 93.8 mcd).

Change to MS: The comparison with Site 1263 and Site 522 oxygen isotope records allowed to confirm that the placement of Step 1 made by Peck et al. (2010) is not correct. Indeed, the two sites are very close and with similar sedimentation rates. The complete $\delta^{18}O$ curve at Site 522 records the two steps of cooling within 4 meters. We can conclude that also Site 1263 might have recorded a similar $\delta^{18}O$ signal, and thus, Step 1 may fall in between 97.5 and 98.5 mcd. (**Lines 529-549**). This position is also confirmed by the additional planktonic foraminiferal analyses performed, which confirmed the position of the EOB at 97.5 mcd (see reply to comment #3 below).

We also revised **Figure 2** as follow: we added the $\delta^{18}O$ data from Site 522, the absolute and relative abundance of *Cl. subdistichus* together with the smoothed curves on 5 five samples to help the identification of the acme, and the biozones of Agnini et al. (2014). All the figures have been modified with the new position of the EOB.

Comment 2b) However there are other ways to improve the planktonic foram biostratigraphy, which is to find the level of the *Pseudohastigerina* size reduction and the *T. cerroazulensis* group extinctions in Site 1263, not done in this study. I would like to know where these are in the core before confirming correlation of the boundary. My prediction is they would be found at a higher level than the top *Hantkenina* so far found. […].

Response: We performed the suggested analysis to solve this issue of the placement of the EOB. The new analyses on planktonic foraminifers show that the Top of *Hantkenina* and *T. cerroazulensis*, and the dwarfing of *Pseudohastigerina* occur between 97.14-97.91 mcd at Site 1263. A spacing of theses bioevents in between 80 cm is possible, but the resolution of our sampling does not allow to identify it. Nevertheless, we can now place with certainty the position of EOB in between 97.14-97.91 mcd. This also confirmed by the isotope comparison with Site 522 (see reply above to comment #2).

Change to MS: we describe the new planktonic foraminifer data and discuss the position of the EOB (**Lines 506-528**).

Comment 3) the Tanzania cores in Fig 1 seem to have been transposed to Somalia.

Change to MS: Correct. **Figure 1** has been modified as suggested.

Referee T. Dunkley Jones

Comment 1) My primary concern is the placement of the Eocene/Oligocene boundary within this section (see also the comment of Pearson and other reviewers). […] In fact, the pattern they observe is actually consistent with the calcareous nannofossil assemblage record from Tanzania, with major assemblage changes actually preceding the first oxygen isotope step and the major planktonic foraminifera extinctions (Dunkley Jones et al. 2008).

Reply: See reply #2 and 3 to P. Pearson for the placement of EOB at 97.5 mcd. The nannoplankton assemblage changes occur above the EOB and before Step 2, as in Dunkley Jones et al. (2008) where they recorded two changes in the nannoplankton assemblage: one before Step 1 and one (more marked) between EOB and Step 2.

Change to MS: we changed the text on the nannofossil assemblage variations according to the new evidence for the position of the EOB (**Section 4.1, and Discussion, in particular Section 5.1**).

Comment 2) I would reinforce the comment of Paul Pearson - the authors need to be careful about their use of “Oi-1”.

Reply and change to MS: See response #1 to P. Pearson.

Comment 3) I’m intrigued as to why two independent samples sets were worked on by two different nannofossil workers. […] I think this shows a positive willingness to collaborate that shouldn’t be “covered up” or reengineered into an a priori experimental test. It has proved to be a very informative test in its own right, and I strongly support its publication, however it came about. To me it demonstrates that, although there are some minor differences, the primary signals are consistent and recovered. This is reassuring.

Reply: The two groups/researchers collected the two datasets independently. The collaboration came about when all the analyses were performed. We agree that it is very interesting to show how, independently from the operator or the sample preparation or the (taxonomic) criteria adopted during the counts, the primary signals were recorded by both researchers.

Change to MS: We clarify this point in the text (**Lines 205-208**).

Comment 4) P1619 – increase in ∂13C benthic as a change in storage of organic carbon in the lithosphere through increased organic carbon burial – maybe, but check other mechanisms of Merico et al (2008). Simple driver of this shift by carbon burial alone appears hard to reconcile with carbon cycle box models.

Reply and change to MS: Correct. We mentioned the effects of the glacioeustatic sea-level fall and sea water carbonate chemistry dynamics citing Merico et al. (2008) (**Line 64**).

Comment 5) the explanation of H diversity could be clearer: really a combination of evenness and diversity rather than “taking into account the relative abundances”.

Change to MS: We changed the text following the suggestion of the referee (**Lines 389-391**).

Comment 6) the placement of Oi-1; as noted above the base of this should be placed at the maximum δ18O value in the basal Oligocene. This seems clear in the Riesselman et al. 2007 paper, but my impression is that the current authors are sliding into a usage for Oi-1 that includes the isotope shift itself.

Reply and change to MS: See response #1-2 to P. Pearson.

Comment 7) Why are the authors using Okada and Bukry nannofossil zonations? Given that they are citing the new Agnini et al. 2014 zonations, and this zonation scheme seems to give better resolution around the E/OB, I would suggest either they use this scheme or justify why it is better not to. (Or at least show both).

Reply and change to MS: We followed the suggestion of the referee and adopted the zonation of Agnini et al. (2014) both in the text (**Paragraph 3.1**) and figures.

Comment 8) does the softness of the sediment really control the presence and / or preservation of palaeomagnetic signals?

Reply: In the Shipboard report (Zachos et al., 2004) this issue with the paleomagnetic data was mentioned. The softness of sediments may affect the magnetic signal because the grains tend to re-orient with the polarity shift. Moreover, the high carbonate content (thus, low % of clay material) led to poor paleomagnetic resolution.

Comment 9) I don't like these references to the calibrated ages. I would much rather the authors use the properly compiled calcareous nannofossil bioevents and calibrations given in Agnini et al. (2014). The authors would then need to make it explicitly clear which timescale they are using and why, and insure that all nannofossil datums are consistently calibrated with the chosen timescale.

Reply: We adopted now the zonations of Agnini et al. (2014), and the calibrated ages from Pälike et al. (2006) for the nannofossil bioevent available. We also clearly state that the timescale used is the one in Agnini et al. (2014) for the nannofossil bioevents and Gradstein et al (2012) for the foraminifer bioevents. We adopted the age from Lyle et al (2002) for the Top of *I. recurvus* because it is not available in Pälike et al. (2006).

Change to MS: See the modified text (**Lines 412-418**) and **Table 1**.

Comment 10) Table 1 – typo in "Massignano".

Change to MS: The sites references have been removed from **Table 1**.

Comment 11) use of abbreviations "B" and "T" for base and top within the text. I am happy with the use of Base and Top, […].

Reply and change to MS: We agree and changed the text accordingly using Top and Base (**Section 3**).

Comment 12) using top *D. saipanensis* to approximate the EOB, when this is clearly some way below the EOB (Dunkley Jones et al. 2008; Agnini et al. 2014). And in the figures, (e.g. Fig. 2) they clearly haven't used this event to approximate the EOB, but place the EOB 6 meters above it! If they haven't used this (wrongly) to approximate the EOB, why say they have? […].

Reply and change to MS: The reviewer is right and we made the necessary corrections in the text (**Lines 450-453**).

Comment 13) identification of *Sph. tribulosus* – the figured specimen in the supplementary information (Fig. S1, 8) is not *Sph. tribulosus*, but looks like *Sph. predistentus* with somewhat overgrown upper spines. […].

Reply and change to MS: We agree that the photograph in itself in the supplement is not enough to justify the classification as *S. tribulosus*. Thus, we added in the Supplement a photo to show the same specimens oriented parallel to the crossed nicols (**Fig. S1 photos 9-10**). With the two orientations of the specimens, the identity should now be indisputable.

Comment 14) I also agree with Guiliana Villa – Fig. S1, Fig15 isn't a dissolved *Dictyococcites* but a (slightly overgrown?) grill-bearing reticulofenestrid.

Reply and change to MS: Correct. We removed the misleading photo from **Fig. S1** in the supplements. See also reply #6 to G. Villa.

Comment 15) use of *Clausicoccus obrutus*. I would like a little more detail on the species concept here and on the differentiation (if any) between this species and *Cl. subdistichus* and *Cl. obrutus*. Do the authors differentiate between these two species at a size of 5.7 *μ*m? Or by number of plates visible in the central area? Based on their distinction, what is the difference between the acme events in *Cl. obrutus* and *Cl. subdistichus*? At ODP 1263, is this increase in abundance more marked in the larger forms, for example?

Reply: We divided *C. obrutus* and *C. subdistichus* on the base of size (*C. obrutus* >5.7 µm). There is a difference between the acmes of the two species: in Fig. S2 it is clear that *S. subdistichus* has an acme slightly before *C. obrutus*. The specimens of *C. subdistichus* are usually very small, so its abundance can be also affected by dissolution. In the end, we used the definition of Agnini et al. (2014) for *C. subdistichus*, by combining the two species in one group. It is also important to notice that the % of *C. subdistichus* is always very low (see Fig. S2) – average 1% - with a maximum peak of 6%.

Change to MS: we specified the use of C. subdistichus of Agnini et al., (2014) (**Lines 454-456**). We changed **Fig. 2** plotting the curve of *C. subdistichus* (sum of *C. subdistichus* and *C. obrutus*). We also added the smoothed curves on 5 five samples to help the identification of the acme.

Comment 16) Also be careful with previous zonal schemes – Okada & Bukry (1980) (based on Bukry 1975) – the base of the zone is defined by *Cl. subdistichus* not *Cl. obrutus*. […].

Reply and change to MS: We use the definition of Agnini et al. (2014) for *Cl. subdistichus*, by combining the two species in one group (**Lines 454-456**).

Comment 17) consistent presence of hantkeninid spines below 96.41 mcd. Linked to discussions above - were these in observed in absolutely every sample studied below the last occurrence in this section? This is important, and if there are samples without spines below this level, they should also be plotted in Figure 2 along with the crosses identifying the presence of spines. Unless of course all samples truly did show spines, in which case I'd like clear confirmation of this from the authors in the text.

Reply: We do not see spines above 97.14 mcd, but they are present in all the samples below.

Change to MS: We now made it clearer in the text (**Lines 512-515**) and added in **Table S1** in Supplement.

Comment 18) "dissolution may be intense"; I think this is over-estimating the dissolution; with "intense" dissolution, I'd expect to see nothing but some robust placolith rims and heavily calcified nannoliths. […].

Reply and change to MS: Correct. We modified in the text referring to a "dissolution slightly increased". (**Lines 598-609**).

Comment 19) I have significant concerns about the discussion of nannofossil abundance (and assemblage) changes relative to the EOB. […].

Reply and change to MS: Now that the EOB position is well constrain, the major events recorded by the nannofossil assemblage occurred after the EOB (and also Step 1). The discussion (**Section 5**) on nannofossil abundance and variations also of PC1 and PC2 were revised to make clear that the changes occurred above the EOB and pre-dated the major cooling (Step 2).

Comment 20) Section 5.3. As above the placement of nannofossil assemblages changes in association with the EOB. For the reasons outlined above, I think the nannofossil assemblage changes significantly precede the EOB, as evidence by their relationship to the oxygen isotope stratigraphy in this section.

Reply and change to MS: **Section 5.3** has been revised considering the new evidences for the placement of the boundary. The nannoplankton response occur after EOB and before Step 2.

Referee G. Villa

Comment 1) I suggest to use the Eocene –Oligocene transition (EOT) whenever it is not strictly referred to the E/O boundary. Many of the changes reported occur across the boundary, not exactly at the boundary.

Reply and change to MS: We revised the text using the term EOT where possible (e.g. **Line 90**). See reply to P. Pearson for the placement of the EOB at 97.5 mcd.

Comment 2a) Biostratigraphy - Line 292 ...Riesselman et al. (2007) placed Oi-1 on the basis of an increase in the benthic δ18 O records from ∼1.5‰ (94.49 mcd, uppermost Eocene) to ~2.6‰ (93.14 mcd, lowermost Oligocene). The Oi-1 according to Reisselman (2007) is instead between 93 and 89 mcd. Also in Peck et al. 2010 is placed between 94 and 93 (fig.6).

Reply and change to MS: We agree that in the previous version of the text the depth of the increase in δ^{18}O and of the Oi-1 were not clear. We changed the text to make it clearer (**Lines 531-533**).

Comment 2b) In your Fig. 2 steps 1, 2 and Oi-1 are indicated as 3 separated events. Step 2 is reported at the same depth as Peck et al., 2010 and therefore Oi-1 should coincide with step 2 (eg. Ladant et al. 2014 Paleoceanography). I cannot understand if it a graph error or if you consider Oi-1 as a third event. Please verify or discuss.

Reply: In Fig. 2 (also other figures) there was indeed a graphical mistake: with the term "Oi-1" we intended to indicate the entire duration of the cooling from above the Oi-1 (=Step 2) and following 400 kyr, but we realize that, as was, the figures might have been somewhat misleading.

Change to MS: We revised **all the figures**, reporting the depth of Step 2, as placed by Peck et al (2010), and we erased the term "Oi-1".

Comment 3) Line 412 the dissolution index shows more intense dissolution from 87 mcd.

Reply and change to MS: Correct. The values of the coccolith dissolution index are lower above 87 mcd, but there is also an important dissolution episode at 90.5 mcd. We changed the text (**Lines 598-599**) and **figs 3 and 6** (grey bar of intense dissolution).

Comment 4) *R. daviesii* is here considered a large species, while it is a medium sized species (5-8 microns). This should be changed.

Reply and change to MS: We agree that *R. daviesii* is a medium sized species, and changed in the text and figures (i.e. **Fig. 5**, V:SA for "large species" re-calculated without *R. daviesii*). Despite this, it has to be noted that the abundance of *R. daviesii* is not so high as to significantly affect the dotted green curve in Fig. 5, which remained very similar. (**Line 637**).

Comment 5) *R. daviesii* is here reported as decreasing at the EOT, while other researchers evidenced a neat increase at the boundary, in particular in the Southern Ocean Sites and at Site 1090, which is quite close to 1263. In Fig S1-15 a specimen classified as *Dictyococcites* with signs of dissolution does not look like a Dictyococcites, and could be a slightly dissolved *R. daviesii*. If this is the case, *R. daviesii* could have been over looked. […].

Reply: We agree that the photo in Fig. S1-15 is ambiguous. Nevertheless, the specimens classified as "dissolved *Dictyococcites*" are very few (1-3 specimens on 300 counted) and very sporadic along the sequence. Even if we would combine the dissolved *Dictyococcites* with *R. daviesii*, the trend of the curve would not change. Thus, we cannot say that *R. daviesii* was overlooked, also because similar trends of this species were independently obtained by two of the authors.

Change to MS: we erased the photo Fig. S1-15. We mentioned in the text the different results reported at other sites for this species (**Lines 638-641**).

Comment 6) In the dataset B the presence of *R. circus* is indicated from about 98 mcd. The specimen illustrated in Fig S 1-20 looks like a *R. circus*. The graphs of dataset A (Fig. 3) of Ret sp.1 shows a very similar pattern of *R. circus* of data set B, […].

Reply: We agree with this suggestion. The specimens in **Fig S1-20** is *R. circus* var. *lata* (Marino and Flores, 2002), according to nannotax3 definition. In database B some *R. circus* (medium size) were detected, but also in Tori (2008) this bioevent was considered unreliable because its presence is discontinuous and it is associated with similar specimens of *Cyclicargolithus* and *Reticulofenestra* (Tori, 2008). For the complexity and very low reliability of this datum it was not discussed in the biostratigraphy section. The graphs of dataset A (Fig. 3) of Ret sp.1 shows a very similar pattern of *R. circus* of data set B, except that it occurs 2 meters below, but it could be the effect of more resolution sampling. It is very likely that R.sp.1 is a *R. circus* and it could be demonstrated that it has an older first appearance. Marino and Flores (2002), at Site 1090, report of a circular Reticulofenestra sp. before the FO of *R. circus* that they considered related to the taxon *R. circus*.

Change to MS: we changed *Reticulofenestra* sp 1 for *R. circus* group in text (**Lines 636 and 799**), **Fig. S1** (**photo 20**), and **taxonomical remarks** (we explained the difference between taxonomical criteria in datasets A and B for this nannofossil group, mainly based on size ranges).

Comment 7) line 494: PC1 is better mirrored more by the red line than that of all placoliths bearing....

Reply: The two curves in Fig. 5 (red and green area) are very similar in terms of trends. The correlation coefficients between the PC1 and V:SA of *Cyclicargolithus* (red curve Fig. 5; r= 0.799) and between PC1 and all placolith-bearing taxa (green area Fig. 5; r=0.79) are also very similar. Thus, we can say that both curves correspond very well to PC1.

Change to MS: we changed the colour of the placolith-bearing taxa curve from green area to a blue line in **Fig. 5**. In this way it is more evident also graphically that they are similar.

Comment 8) Fig caption 3 add if these data are form dataset A only.

Reply and change to MS: Yes, they are just from dataset A. Added in the caption.

Comment 9) Fig .5 there is not the graph of the total abundance (mentioned in Fig caption). The legend of black and white circles are inverted. TDP must be changed in TDP 17/12.

Reply and change to MS: We changed the caption.

Anonymous referee

We edited the text accordingly, for the comments #4-5-7-12-13-15-17-19-21-23-24-25-26-28-30-35-38-39-40-42-43-46-47-48-50-53-55-59-60-66-70.

A) The Oi1/EOB issues
I totally agree with Paul Person (reviwerer #1) on the Oi1/EOB issue. The position of EOB at Site 1263 is quite suspicious and the Top of *Hankkenina* and *Cribroantkenina*, the authors use to mark the boundary, is more likely anticipated because of dissolution and/or ecological factors. In addition, the use of different nomenclatures makes the reading very difficult and confusing.

Reply and change to MS: This was clearly an issue picked up by all reviewers. See reply 1-2-3 to P. Pearson for EOB placement and Oi-1 issue.

B) The biostratigraphic issues
There are many comments on taxonomy, reliability and positioning of biohorizons, misleading use of biostratigraphic concept, mistaken use of biozone definition, age model, etc

Reply and change to MS: We followed the suggestions of the reviewer, paying attention to the different concepts used for the biostratigraphy (changes have been done in Section 3).

1) Pag. 1617, line 14. Do you have evidences for that?

Reply: It is possible that smaller, less calcified taxa have lower PIC/POC ratios on a cellular level than larger, heavily calcified taxa (compare for example modern species *Gephyrocapsa oceanica* vs. *Coccolithus pelagicus*, e.g. Zondervan et al. 2001; Langer et al., 2006; Rickaby et al. 2010; Müller et al. 2010). Smaller individual nannofossils/coccoliths relates to smaller carbonate mass (per individual) and may also relate to lower carbonate production per cell, but the net amount of carbonate produced is not only linked to the coccolith's size but also to the overall productivity of coccolithophores.

Change to MS: Since we don't undertake any modelling in this paper to deepen this aspect, we will erase this statement here to avoid any over-interpretation (**Lines 33-34**).

2) Pag. 1617, lines 16-23 E/I is sensitive to carbonate saturation and O2 not just food supply. How can you disentangle the role played by these three parameters?

Reply: We should not address this question in the abstract, but in the text.
Change to MS: We changed section 4.3 (**Lines 751-762**) to make this concept clearer.

3) Pag. 1617, line24.~~planktonic~~ calcareous nannofossil. Too much general, in principal includes planktonic forams, but the authors do not present any new planktonic foram assemblage result, […].

Reply and change to MS: Correct. We changed for "calcareous nannofossil" (**Line 43**).

6) Pag. 1619, lines 5-8. I would agree with Paul Person. Though the formal definition of Rupelian GGSP includes the Top of *Hantkenina* and *Cribrohankenina.* These biohorizons better play the role of primary markers, which denote rather define the boundary. The recognition of the Top of *Hantkenina* and *Cribrohankenina* could be problematic in some cases but the use of alternative markers could serve to better constrain the position of the EOB.

Reply: We agree that by using only the T of *Hantkenina* we cannot accurately assess the position of the boundary, so we performed more analyses. See replies 2-3 to P. Pearson.

Change to MS: we moved the explanation on the planktonic marker species in **Section 3.2 (Lines 494-505)**, to make this paragraph more effective and shorten the introduction. See also reply #2-3 to P. Pearson.

8) Pag. 1619, lines 15-16. This concept in non-intuitive and should be explained by the authors. The increase of C org/Cinorg ratio can surely be the result of enhanced export productivity but can also be related to the increase in C org preservation. […].

Reply: Coxall and Wilson (2011) documented how the increase in C org/C inorg at the EOB is related to increased surface biological production and/or increased efficiency of organic carbon export by comparing the δ^{13}C signal and the BFAR results which "appears to correlate predictably with export production in a variety of modern locations [Herguera, 1992; Loubere, 1994; Jorissen et al., 2007] and the method has been used widely as a qualitative/ semiquantitative proxy of paleoproductivity […] and early Cenozoic […]".

Change to MS: We changed the text to make it clearer: "*an increased ratio in the burial of organic versus inorganic carbon (calcite) due to enhanced marine export production and/or increased preservation of organic matter"* (**Lines 99-104**).

9) Pag. 1619, line 20. The CCD deepening is a consequence more than a cause, as inferred by the authors. The sentence should be probably re-write in order to make this clear […].

Reply and change to MS: We re-phrased the sentence (**Lines 105-109**).

10) Pag. 1619, line 21. The use of term response sounds strange.

Reply: The word "response" is commonly used to indicate how a biotic community is behaving/changing to environmental/climatic variations. So the term is correct in this sentence. See also the use of "response" in e.g. Flores et al. (1995), Villa et al. (2008; 2014), Dunkley Jones et al., (2008).

11) Pag. 1619, lines 22-23. Extinctions always occur in the geological time. What the authors might mean is that rate of this extinctions either increases in its absolute number or increase if compared the speciation rate of the same interval.

Reply and change to MS: Correct. The extinctions across the EOT increased if compared to the speciation rate. Changed in the text (**Line 112**).

14) Pag. 1620, lines 9-14. I'm not aware of any comprehensive species diversity study. This would include a measure of both species number and 'equitability' (or 'evenness') (e.g., Simpson Index, Fisher's alpha).

Reply: Several studies on nannofossil assemblages displayed the Shannon Weaver Index – i.e., H index (e.g., Persico and Villa, 2004; Dunkley Jones et al., 2008; Pearson et al., 2008), so we report here what is available in the literature for this time interval. It is possible to show also other indices, but they do not add different/ or new information to the H index.

16) Pag. 1620, line 15. I'm not sure they were more abundant but for sure they dominated the marine phytoplankton and show a higher species diversity with a maximum recorded in the early-middle Eocene (Bown et al., 2004).

Reply and change to MS: We followed the suggestion and modified the text (see **Lines 132-137**).

18) Pag. 1620, line 17. The increase in abundance and species richness of diatoms started well before the EOB and coincide with the general decline displayed by calcareous nannoplankton since the early/middle Eocene (e.g Bown et al., 2004; Spencer-Cervato, 1998).

Reply and change to MS: Correct; now this is better specified in the text (see **Lines 145-148**).

20) Pag. 1620, line 26. What do you mean with "driven"? A macroevolutionary trend observed in group can not be "driven" by a part of the that group. Rather, it could be the result of something affecting selectively a part of the group. That is a complete different concept. Not sure what the authors mean.

Reply and change to MS: In this context "driven" means that the trend is linked or mainly due to the decrease in abundance of large reticulofenestrids. We changed "driven" to "caused by" in the text (**Line 157**).

22) Pag. 1621, line 1. The authors should strengthen their point using modern ocean analogues that are easily findable in literature.

Reply and change to MS: We changed the text adding also analogues from recent culture experiments, in particular referring to the relationships between pCO_2 and cell size variations. (**Lines 161-169**).

27) Pag. 1621, lines 1 6-17. This is crucial. The number of forms per gram does not provide an estimate of fluxes, it is rather an evaluation of absolute abundance. The definition of paleofluxes are very different since it implies to put absolute abundance in a time tuned series (g10-6mm-2 y-1).

Reply: The number of nannofossils per gram does not provide an estimate of fluxes, and in the text we always refer to these data as absolute abundances, not as fluxes. The calculation of paleo-fluxes in this context is not possible in our opinion, because it requires a very detailed and well-constrained age model. Unfortunately, this is not the case at Site 1263. We identified several bioevents, but the magnetostratigraphy is not available (Zachos et al., 2004). Thus, by adding an estimated sedimentation rate that is below the resolution of the data, we would introduce a substantial error in the evaluation of paleo-fluxes. For these reasons we decided to present only absolute abundances.

29) Pag. 1622, lines 6-15. The authors should explain why they follow this approach, which is the bonus of having two datasets of "virtually" the same material? This is non-intuitive.

Reply and change to MS: We now explained better in the text why we compared two different datasets (see also reply #3 to T. Dunkley Jones). Two groups worked independently at the same time interval for this site, although the amount of samples and the temporal resolution are different. This approach shows how, independently from the sample preparation and operator, the primary signals are documented and consistent. (**Lines 205-208**).

31) Pag. 1622, line 22. How the authors avoid the selective settling effect? This step should be described in much more detail.

Reply: For sample preparation we followed the protocol described in Bordiga et al. (2015), where the technique was accurately tested. Any further description is not required. The settling is not an issue in this technique as we start from bulk sediment and all the suspension (sediment and buffered water, well mixed by short sonification intervals) is evenly placed on the cover slip.

Change to MS: we changed the text referring to Bordiga et al. (2015) protocol. (**Lines 220-235**).

32) Pag. 1623, lines 3-4. It is quite significative considering the relative abundance changes observed for most of the taxa (see Fig. 3). The author should comment on this issue. CV => Please in full (coefficient of variation), at least the first time.

Reply: We will define "CV" as coefficient of variation in the text. The CV of 11% refers to the absolute abundances, not to relative abundances which are highly comparable on replicates and when compared with different techniques (see Bordiga et al., 2015). The variation among abs. ab. replicates of 11% is comparable to results obtained in previous studies (Bollmann et al., 1999; Geisen et al., 1999; summary table in Bordiga et al., 2015). This explanation in no longer necessary in the text, as we referred to Bordiga et al. (2015).

33) Pag. 1623, lines 4-6. This is not correct. The number of specimens per gram counted in a prefixed area is an absolute abundance but this index does not take into account the time. What I mean it is that we know the number of forms per gram (absolute abundance) but we do not know the flux. i.e. number of forms x g10-6mm-2 y-1. This value could have could changed substantially if the mass accumulation rates change. In other word, if the authors has not a good highly-resolved age model for their study profile then they can say very little about paleofluxes /paleoproductivity. […].

Reply: It is correct that the number per gram does not take into account the time. We did not use the absolute abundances with this connotation in the paper, and we cannot calculate paleofluxes for the low resolution of the age model (see the reply #27).

On the other hand, we were able to discuss paleoproductivity using the assemblage data (treated with the PCA) because we compared them with the independent proxies - the isotopes ($\Delta\delta^{13}$Cp-b) and benthic assemblage data - to reinforce our observations.

34) Pag. 1623, lines 6-10. I would agree with the authors but this issue is rather more complicated than explained here. See for instance (discussion paper of Gibbs et al. 2012 http://www.biogeosciencesdiscuss.net/9/C618/2012/bgd-9-C618-2012.pdf). In the following the authors eventually decide to use relative abundance data quite heavily (though with some transformations), so why to destroy this kind of data. This seems incoherent. Please comment.

Reply: We made this point more clear. The reviewer is right that the wording we used before could be construed as to "destroy" the relative abundances. Here we used both absolute and relative abundances and by comparing them we can investigate the influence of dilution and sedimentation rate. Indeed, as pointed by the reviewer, Gibbs et al. (2012) correctly argue that "Relative abundance provides 'biological' information, i.e. relative abundances of taxa within the populations, which is independent of modifying sedimentological effects, such as varying accumulation rate." But we disagree that "changing the relative abundances into a more 'absolute' abundance form, such as numbers per gram does not provide any additional information and is actually adding a level of degradation. For example, to convert to numbers per gram would mean we introduce a dilution signal to our data that isn't necessarily anything to do with a biotic response." It is true that absolute abundances (N g-1) are affected by dilution and sedimentation rates, but by comparing the absolute with the relative abundances (see results section and supplement Fig. S2) we documented that their trends are very similar. This suggests that the dilution/sed. rates at this particular Site were constant over time, thus we can refer to the absolute abundances as linked to biological processes. (The constant sedimentation rate is also documented by the age model which, although not highly-resolved, provides a relatively constant sed. rate). In our case, the absolute abundances provide additional information about the total coccolith absolute abundance, which it is not possible to derive from other type of data (such as the relative abundances), and it is an important part to define the calcareous nannoplankton productivity. Indeed, if the trends of absolute and relative abundances of single species are similar, we can infer that the total absolute abundance is linked to biological processes.

Change to MS: we better specify in the text (both in the methods and results sections) the benefits of using absolute and relative abundances. (**Lines 236-244 and Lines 627-630**).

36) Pag. 1623, lines 22-23. I would suggest that this information (number of fields of view (FOV) observed) would be added to the supplementary material.

Reply and change to MS: The number of FOVs and its relative mm2 area observed are added in the text, not in the supplementary material (**Lines 261-263**).

37) Pag. 1623, lines 22-23. This is not clear to me. The authors first claimed that relative abundance data are problematic and now they decide to use these data to describe the composition of nannofossil assemblages. This is awkward. Why they do not use their absolute abundance data? This point should be better explained and justified.

Reply and change to MS: We did not want to claim that relative abundances are problematic, but we agree that the sentence might have led to this misunderstanding. Now we rephrased it. See reply to #34 above for more details on the use of relative and absolute abundances. We used the relative abundances mainly for PCA: that was necessary because we wanted to compare two different datasets, and the absolute abundances were available only for one of these datasets. (See changes at **Lines 236-244 and Lines 662-666**).

41) Pag. 1625, lines 2-6. I think this is a very nice approach but then, again, the authors should rethink about their statements on the poor validity of relative abundance data. Either they are a good proxy of what is going on or they possibly lead to loss of information and misinterpretation of the results (as stated above). You cannot have your cake and eat it, you have to reformulated your sentence…

Reply: See comment #34 and 37, we do support the validity of relative abundances. Considering the explanation now given in **Paragraph 2.2.1**, the sentence does not need to be reformulated.

44) Pag. 1628, lines 11-13. I do not get the point here, which kind of bias do you mean? In addition, Set A and Set B area quite different one to each other and even if consistent results finally popped out, these should be discussed properly. Just as a note, if you think something can bias your data, as it is generally written in your sentence, then, in principal, you have to be worried about the possible misleading alteration due to "the two operators effect". Again, I do not see the real point in following this approach (duplicating datasets).

Reply and change to MS: We erased that sentence as it could lead misunderstanding (**Lines 405-406**). We meant that we wanted to compare the occurrence (detection) of marker species, especially the rare ones. The use of the two datasets is also important to compare the results of PCA, and to observe if the primary signals are documented by both datasets. About the use of two datasets see reply #29.

45) Pag. 1628, lines 20-26. The age model as constructed by the authors to compare the two dataset followed a quite circular reasoning, without any independent correlation tool (e.g., magnetostratigraphy, isotope stratigraphy,…) in support of their chronological framework. Now, it is quite clear that this will not going to affect the correlation between set A and set B too much because the two series are recovered by different holes of the same Site and they hopefully recorded the same geohistory, but what could instead happen if these datasets (without any independent age constrains, but derivated nanno biochronology) will be correlated using the same rationale? I would suggest the authors to add a sentence about this issue.

Reply and change to MS: The correlation with the oxygen isotope curve is inserted in the text (**Lines 529-549**) and figures (fig. 2). The datums from planktonic forams and nannofossils well constrain the age model. Unfortunately, no magnetic data are available for Site 1263.

49) Pag. 1629, lines 8-9. The stratigraphic range is related to a species not to a bioevent. The position of bioevent (e.g., B *S. trilobosus*) is the consequence of the stratigraphic range of *S.*

trilobusus. In other word, a biohorizon has not a stratigraphic range is rather a stratigraphic level, in this case the stratigraphic level where *S. trilobosus* first occurred.

Reply and change to MS: Correct. Modified in the text (**Line 437**).

52) Pag. 1629, lines 12-14. abundant and it's the poor preservation *of the study material* is commonly compromiseing the identification at the species level and thus possibly, its B. Again, this sentence sounds strange. Abundant? This is an euphemism. I would say that this species is rare to very rare and sporadic.

Reply and change to MS: Yes, the species is rare and sporadic. Modified (**Lines 440-442**).

54) Pag. 1629, lines 18-19. Looking at the abundance pattern, I would say that the Top of *D. saipanensis* should be positioned at ca. 104 mcd, where this species goes to 0. […].

Reply: The T of *D. saipanensis* had been placed at 102.27 mcd because specimens of *D. saipanensis* had been continuously found until 102.52 mcd, although outside the count of 300 specimens.

Change to MS: We added the presence of *D. saipanensis* between 104 and 102.27 mcd (green crosses) in **Fig. 2** and specified in the text (**Lines 448-449**).

56) Pag. 1629, line 24. Please consider to use *C. subdistichus* in place of *C. obrutus*.

Reply and change to MS: Modified in the text (**Lines 454-456**): according to Agnini et al. (2014) we combined *C. obrutus* and *C. subdistichus* because *Clausicoccus obrutus* is considered to be a junior synonym of *C. subdistichus* (www.nannotax.org).

57) Pag. 1630, lines 2-3. This is not correct. The base of Zone CP16b is defined by the T of *C. obrutus*. Recently, Agnini et al (2014) proposed to use the B of *C. subdisticus* (whose definition include also *C. obrutus*) to define their Zone CNO1. Backman (1987) never emended the original definition of the base of CP16b. In his key paper, he emphasized the potential of the Bacme of *C. subdistichus* and suggested that this biohorizon could be used to subdivide Zone NP21. He did not mentioned about the base of CP16b, whose application is for sure difficult since the Tacme of *C. subdistichus*. All this issue should be managed.

Reply and change to MS: We modified the text (**Lines 454-456**) and **Fig. 2** according to the definition of Agnini et al., (2014).

58) Pag. 1630, line 10. The B of *C. altus* can be *is tentatively* placed with certainty at 89.4 mcd. […], I would say that the use of "with certainty" should be avoided.

Reply and change to MS: Correct. Modified in the text (**Line 473**).

61) Pag. 1630, lines 16-17. B and Bc were identifiable I would agree for Bc of *Sphenolithus akropodus*but B of *Sphenolithus akropodus* is very very tentative. Do you really think this is a reproducibible event?

Reply and change to MS: Just few and sporadic species of *S. akropodus* are present below its acme. We think that it is important to also report the first occurrence of this species, but we specified in the text that this is tentative due to the rarity of this species (**Lines 478-480**).

62) Pag. 1630, lines 20-22. This is tricky. The abundance plot stops exactly where *E. formosa* goes to 0. My point is how can you be sure that 85.15 mcd actually corresponds to the Top of the species. Is this just because of the shipboard data. I cannot see any other independent evidence for this statement. Please comment.

Reply and change to MS: Considering both datasets A and B, six samples above 85.15 (up to 83 mcd) where analysed to detect the disappearance of *C. formosus*. We consider this good evidence for the position of this datum. Moreover, this depth is very close to the one detected by the Shipboard Party (at 86 mcd). We modified **Fig. 2** plotting also the six samples where *C. formosus* is not present.

63) Pag. 1630, lines 23-27. See comment above.

Reply: For the case of *I. recurvus* we cannot be sure that the depth of 83.19 mcd truly corresponds to its disappearance because just one sample above the last observed specimen was analysed. This is why we stated in the text that this depth is “an approximation”.

64) Pag. 1631, lines 11 -13. I totally agree with Paul Pearson. The Top of *Cribohantkenina* and *Hantkenina* are in fact a marker of the EOB but they should be used with extreme caution and, if possible, integrated with other additional biohorizons that would strengthen the datum.

Reply: We agree with this observation. See also reply #2-3 to P. Pearson for the placement of the EOB and **Lines 506-528** for the explanation of the new data.

65) Pag. 1632, lines 5-7. This inconsistency could be related to a change in carbonate source (more forams) but this is not the only possible explanation. The authors should take into account the different amount of carbonate produced by different taxa. […].

Reply and change to MS: We agree with the reviewer. We modified the text specifying that we don’t know the amount of carbonate produced by foraminifers and calcareous nannoplankton, so that the real contribution of each group cannot be unravelled in this study (**Lines 576-581**).

67) Pag. 1632, lines 17-18. How can you say that? During the late Eocene - early Oligocene, small placoliths are by far the dominant taxa in the ocean, the total absence of small placoliths would have an (important) impact on the paleoecological interpretation of CN data. This is the endless debate pristine signal vs dissolution. Please comment.

Reply: The small placoliths were not abundant in the studied interval, particularly during the late Eocene. It is true that the absence of small placoliths might be indicative of dissolution, and clearly dissolution is present at this site. For comparison we calculated also the coccolith dissolution index which shows intervals of increased levels of dissolution (although overall still “moderate to good preservation” Zachos et al., 2004), but not always corresponding to lower absolute abundances. Of course such hypothetical/possible signal from small placoliths is not detectable anymore, but by calculating the coccolith dissolution we provide an alternative and valid index for comparison between samples and with other data. Nevertheless, the lack of small placoliths does not prevent the identification of the main features of the medium-large sized taxa

Change to MS: Text has been changed following the suggestion (**Lines 593-597**).

68) Pag. 1632, line 20. (Fig. 3) => I would add the isotope curve…

Reply and change to MS: Due to the new evidence on the position of the EOB and the comparison with Site 522 for the $\delta^{18}O$, it is not relevant to insert the isotope curve in Fig. 3. Nevertheless, we modified **Fig. 3** plotting the position of the Step 2.

69) Pag. 1632, lines 24-26. The absolute abundance of CN is not preserved. As you stated just few lines above, many small placoliths were dissolved. What you can try is to support the idea that the relative abundance of the different taxa remained the same, but this is obviously not true because, as you said, dissolution is selective, which in turn implies it does not affect different taxa in the same way.

Reply and change to MS: It is correct that the absolute abundance may have been affected by dissolution, because we cannot reconstruct the story of small placoliths. But we can say that moments of intensified dissolution do not correspond to moments of lower abundances: thus, we can still discuss about the main features of the assemblage. See also comment #18 from T. Dunkley Jones: the dissolution was not so intense, otherwise “we would expect to find just some robust placoliths and heavily calcified nannoliths”. Instead, we find abundant small-medium sized *Cyclicargolithus* and also holococcoliths (which are recognized to be very prone to dissolution; Blaj et al., 2009; Bown et al., 2008; Young et al., 2005) in our samples. (**Lines 588-597**).

71) Pag. 1632, lines 25-28. This is actually not clear to me. At the EOB, the large placoliths increase, this is crystalline, but if I look at the total absolute abundance the decrease is much less marked, may be because of the increase of *C. pelagicus* that, at least in part, counterbalances the trend of large placoliths. It is likely, however, that the export carbonate productivity decreases because larger coccoliths produce more carbonate. So again, coccolith absolute abundance and carbonate export productivity are different concept.

Reply and change to MS: It is correct that *C. pelagicus* slightly increase when the large reticulofenestrids decrease (above the boundary), and this is why the decrease of total abundance is less marked than the abundance curves of large reticulofenestrids. We better explained this point in the text, being careful to refer to the decrease of large “reticulofenestrids” and not decrease of large species in general. (**Lines 650-655**).

72) Pag. 1636, lines 8-9. I would say that *Cycligarlorithus* mean cell size drives the high correspondence between in V:SA and PC1.

Reply: We do not show "*Cyclicargolithus* mean cell size" as a separate curve, but we agree with the reviewer that the abundance and medium size of *Cyclicargolithus* influences the red curve most (since it is the most abundant species in the mix). However, in similar fashion one could argue that the contribution (abundance) of large-sized reticulofenestrids (which, as shown in the green dashed curve in Fig. 5, do not drastically vary in size, but rather, decrease in abundance (as shown in Fig. 3) is "driving" the curve.

The comment prompted us to clarify the point that the mean V:SA values are derived from the relative abundances of differently sized morphotypes, so that both changes in relative abundance and potential size shifts within morphotypes may influence the results (although the latter could also mean jumping into a different size class and thus be seen as a relative abundance shift). For example, the large retics actually do not "decrease in size" above the EOT, but they contribute less to the mean V:SA value above the EOT.

Change to MS: the text is now clearer (**Lines 717-720**). We changed the colour of the placolith-bearing taxa V:SA in **Fig. 5** to make more evident the similarity with the red curve.

73) Pag. 1636, lines 15-18. I would reiterate my point. You do not have any information of dominant taxa, just because they are not in the assemblage anymore. It might be the case that smaller placoliths show a particular trend. For instance, if they would be very abundant, where larger coccolith are very rare then your hypothesis is collapsing like a house of cards.

Reply and change to MS: The information about the very small placoliths is not available, either because they indeed were rare or, more probably, because of dissolution. But we think that it is encouraging that the independent proxy for dissolution, i.e. the coccolith dissolution index, does not correspond to increases in mean size (which could be seen as the result of dissolving away the smaller sizes of the assemblage).

Moreover, the loss of large reticulofenestrids cannot be linked to a dissolution bias – since they are more robust and resistant to dissolution than smaller (medium) forms such as *Cyclicargolithus* spp. The trend of mean V:SA under discussion is not affected by the presence or absence of small placoliths, that are irrelevant in this consideration of mean size variation within the medium to large reticulofenestrids. (**Lines 730-732**). See also our response above, #72.

Other authors did not either document a substantial or consistent presence of small (<3µm) specimens in the same time interval (late Eocene-early Oligocene) at various latitudes (Persico and Villa (2004) and Villa et al. (2008) in the Southern Ocean, Fioroni et al. (2015) in the equatorial Indian Ocean). (**Lines 594-595**).

74) Pag. 1636, line 9. This paragraph is a long dissertation on what is going on in the placolith world where CO2 values are decreasing. This is really fascinating but I would like that the authors look at the entire assemblage. Are the changes observed in non-placolith taxa (e.g, *Sphenolithus, Discoaster, Z. bijugatus*) confirming their interpretation? This is would be really interesting to see. The authors may claimed that these taxa represent a minor

component but they are ca. 20% (on average) of entire assemblage and, even more importantly, they produced much more carbonate than a small placolith. Finally…

What about *C. pelagicus?* It is a placolith (a major component of the assemblage) but it does not seem to follow the same trend observed for reticulofenestrids (Fig.3). How can you explain that? Why V:SA ratio of *C. pelagicus* increase when reticulofenestrids decrease in their cell size?

Reply and change to MS: Other species, such as *Sphenolithus* spp. (all species grouped together), do not show any particular abundance variations at the same depth as the large reticulofenestrids. *Z. bijugatus* shows a slight decrease before the decrease of large reticulofenestrids and in correspondence of the decrease in abundance of *Discoaster* spp. (Fig. S3). Considering the paleoecological significance of *Sphenolithus* and *Discoaster*, as reported in the literature, their higher abundances may relate to the more oligotrophic conditions during the late Eocene. A relationship with the reconstructed trends in paleo-CO_2 cannot be seen. These species are better discussed in the **section 5.2** (**Lines 846-862**), and in the text we refer to the species (reticulofenestrids) that have a clear relationship with the CO_2.

We re-iterate that mean V:SA values are derived from the relative abundances of differently sized morphotypes – whether for (groups of) reticulofenestrids or *C. pelagicus*. *Coccolithus pelagicus* shows little variation in abundance and "size composition" (differently sized morphotypes), and thus not either in mean V:SA estimates. Why this is the case, from a paleo-biological perspective, is of course a very interesting question. We don't know why this taxon would be less affected across the EOT, but it seems evident that *Coccolithus* vs reticulofenestrids tend to respond in different ways, likely linked to differences in physiology between the two families (e.g. Rickaby et al. 2010; Krug et al. 2011; Lohbeck et al. 2012; Gibbs et al., 2013).

75) Pag. 1637, lines 10-12. I would stress this point because this really supports the authors' scenario. Larger forms are proved to be less prone to dissolution. A general decrease in size would not be caused by dissolution, which works the other way around.

Reply and change to MS: Correct. We modified the text as follows, making this point clearer (**Lines 767-771**): "The distinct variation in nannoplankton abundance and average size of medium to large placoliths above the EOB at Site 1263 cannot be explained by dissolution – which would affect smaller coccoliths preferentially and lead to an increase of the mean size of the whole assemblage, opposite to what is observed.".

76) Pag. 1640, lines 8-13. a) The positive loading of PC2 is bizarre. How can you explain the fact that the major component of the assemblage (even considering that clr is applied to the dataset), the reticulofenestrids, show very little load capacity?

b) How can you explain that *Sphenolithus* and *Discoaster*, two warm oligostrophic taxa, have a positive component loading in PC2. If your interpretation is correct, PC2 represents paleoproductivity, then I would expect the opposite behavior. This result points for a strong eutrophic affinity for sphenoliths and discoasters.

c) How can you explain that *D. stavensis* and *D. bisectus* show an opposite behaviour if compare with that of *R. scrippsae?* The reason why I ask this question is that if you accept the taxonomic validity of genus *Dictyococcites*, you should consequently ascribed *scrippsae* to *Dictyococcites* not to *Reticulofenestra*. As a note, *D./R. scrippsae* (Fig. 4) is possibly considered a junior synonymous of *D. hesslandii,* so please consider to revise its taxonomy.

d) Are you sure that PC2 (by the way, PC2 could account just for the 14% of the variance of the entire assemblage) could be correlated with paleoproductivity so straightfully?

Reply a) The reticulofenestrids show little loadings with PC2 because two components retained through the PCA are, by definition, uncorrelated variables (Quinn and Keough, 2002 – Data analysis for biologists). Thus, the species strongly loading the PC1 will not necessarily load also the other components (as it is the case here). It is not always the case that the most abundant species have to load both (or several) components of the PCA. Moreover, we cannot really say that PC2 is loaded by “not abundant” species, because *C. pelagicus*, *L. minutus*, *Sphenolithus* together represent the 20-40% of the assemblage.

Reply and change to MS b) Recently *Sphenolithus* has been related to more nutrient conditions than to temperature control (**Lines 857-862**). We can infer that probably *Sphenolithus* was an opportunistic taxon, as also documented by Wade and Pälike (2004) and Dunkley Jones et al. (2008) across the EOT. This interpretation is well in agreement with the loading of *Sphenolithus*, opposite to the oligotrophic *L. minutus* (**Lines 860-862**). The positive loading of *Discoaster* is not strong enough to be meaningful for explaining the PC2.

Reply and change to MS c) We agree that from a taxonomical point of view it is better to ascribe *R. scrippsae* as *Dictyococcites scrippsae*, we changed in **Fig. 4**. We cannot fully define the different behaviours of the different loadings in PC2, especially because the loading of *D. scrippsae* is very small, thus it does not have to be taken into account for the interpretation of PC2. We maintained the name species “*scrippsae*” because our specimens can be described as ”coccoliths with a solid central plug” (nannotax3 website), while *D. hesslandi* is defined as “Coccoliths small to medium sized (3-7µm) with central area nearly closed by inner tube” cycle (nannotax3 website). We did not observe a nearly closed inner tube but a solid central plug.

Reply d) We correlate PC2 to paleoproductivity not only on the basis of the species loadings, but also from the comparison with carbon isotope data and data assemblage from the benthic foraminifers (**Lines 864-882**). Indeed, PC2 is not the primary factor, so it is not mainly driving the assemblage.

77) Pag. 1640, lines 19-20. …Or just because this correlation doesn’t work. I do not mean that the final interpretation is incorrect but it shoudn’t be based on so weak an argument. The authors have a stronger potential defense for their interpretation.

Reply and change to MS: The low correlation between the two curves is due to the low number of samples in common between PC2 and carbon isotopic gradient (indeed the use of the same samples is required to make a correct correlation analysis). But from a visual comparison between the two curves we can see that both recorded an evident decrease around 92 mcd. We have a good comparison with the benthic foraminifer assemblage, which confirms the interpretation of PC2 as paleoproductivity or nutrient supply. Indeed, the increase of phytodetritus species corresponds very well to the positive peak of the PC2. So, we have two independent proxies to strengthen the interpretation of PC2 derived from the nannofossil data (**Paragraph. 5.2**).

78) Pag. 1640, lines 24-25. This is counter-intuitive. Looking at the Pacific record (Coxall et al., 2005) as many others, I would expect an increase in productivity. If the authors claimed for the opposite, they should provide an explanation for this inconsistency. Is this a local effect? And, if this is the case, Can they provide a global paleoproductivity model in which their dataset could be included? Is there any chance that their results could be interpreted in a different way?

Reply: We reported in the text several previous studies where the paleoproductivity has a similar trend as we recorded (**Lines 924-931**). This confirms that similar conditions occurred also at other tropical and equatorial latitudes. Of course we cannot completely exclude the influence of some local effects. The text had been changed to make this point more clear. About providing a global paleoproductivity model: this would imply another level of analysis and it is not the main aim of this paper. We think that our interpretation is well supported given the two fossil groups and isotope data.

79) Pag. 1641, line 3. This data set could nicely account for what is going on in bottom waters not in sea surface waters. It often happens that these two domains are “disconnected”, especially during dramatic changes in paleoenviromental conditions, as the EOB.

Reply: Although the two domains might be disconnected, the benthic foraminifers have been used as indicator of paleoproductivity at the surface also at the EOB (e.g. Coxall and Wilson, 2011).
We previously used “nutrient supply” which suggests supply in N and P to photosynthesizers, but here we are talking about food supply to the benthos, which is not ‘nutrient supply’. To make this point clearer we changed **Lines 883-885** to: “The benthic foraminifer assemblage confirms the interpretation of the PC2, adding information on the nature of supply of organic matter to the seafloor (Fig. 6).” We specify better and separate nutrient supply to the nannoplankton from food supply to the benthos (**Lines 899-910**).

80) Pag. 1641, lines 12-14. If I have understood correctly, the decrease in coccolith size is driven by decreasing CO2 values, Am I wrong? Which is the driving forcing for this change? The CO2 or the paleoproductivity? And, in case they are both responsible for this change, which is the factor commanding the decrease in coccolith size?

Reply and change to MS: We postulate that the decrease in mean cell size of placolith-bearing coccolithophores could have been driven both by varying levels of CO2 and/or paleoproductivity (or, rather, nutrient supply). We divided the discussion of the two factors in two paragraphs, but it is probably better to state again the influence of pCO2 (**Lines 894-896**).

In the previous section 5.1 we discuss that other abiotic factors might have affected the decrease in cell size, and that they are deemed to be dominant.

81) Pag. 1641, lines 16-20. I do not get the point here. The decrease in coccolith size occurred at ca. 96 mcd and coincides with high seasonal productivity in benthic communities (Fig. 6 and Fig.7=> phytodetritus abundance). Is there a possibility that buliminids show a relative decrease in abundance because they are temporary overcompeted by phytodetritus species, in a different but still high productivity regime? Did I miss something?

Reply: As explained in the reply to comment #79, benthic forams cannot be directly linked to nutrients (N, P) since they are not autotrophs.

Change to MS: we made this point clearer in the **Lines 883-910**.

82) Pag. 1641, lines 21 -24. What about the missing part of the story, the smaller placoliths. If I follow your reasoning, I would imagine that smaller placoliths (3-4 µm), which are absent from the fossil record, should have been very abundant at that time, may be increasing in number as their larger counterpart (4-7 µm) did.

Reply: With “small placoliths” we meant specimens smaller than 3 µm. Specimens with a size of 3-4 µm are present and well-represented in the assemblage. It is possible that during moments of higher productivity the small nannoplankton could have flourished or increased its abundance, and it is true that we don’t have this information at the Site 1263. But, also other sites, with a good preservation, high abundances of small placoliths have not been recorded during the same time interval. See reply #73.

83) Pag. 1642, lines 0-13. As I commented above, you need to synthesize all these data in more global perspective.

Reply: In the text we present general synthesis of the data available also from other sites (**Lines 920-932**), inferring a “meridional gradient in biotic response”. A wider synthesize, which would include also a modelling (as suggested in comment #78), is outside of the scope of this paper.

84) Pag. 1644, lines 12-15. See comments above on the same issues.

Reply: See reply #83. We have a good comparison with previous studies (**Lines 965-976 and 1006-1019**).

85) Pag. 1645, line 13. I commented above on each of the main results reported in the conclusions. Some should be revised.

Reply and change to MS: The **Conclusions** have been revised/clarified following all discussions in this open online forum. In particular, we will clarify the position of the biotic changes relative to the EOB (updated with new plaktonic foram data) and Step 2, as discuss the paleoproductivity issues raised by this referee.

~~The Eocene-Oligocene transition at ODP Site 1263, Atlantic Ocean: decreases in nannoplankton size and abundance and correlation with benthic foraminiferal assemblages~~

Microfossil evidence for trophic changes during the Eocene-Oligocene transition in the South Atlantic (ODP Site 1263, Walvis Ridge)

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Abstract

The biotic response of calcareous nannoplankton to environmental and climatic changes during the Eocene-Oligocene transition ~~(~34.8-32.7 Ma)~~ was investigated at high resolution at Ocean Drilling Program (ODP) Site 1263 (Walvis Ridge, South East Atlantic Ocean), and compared with a lower resolution benthic foraminiferal record. During this time interval~~, the~~ global climate, which had been warm ~~during the Eocene,~~ under high levels of atmospheric CO_2 (pCO_2) during the Eocene, transitioned into the cooler climate of the Oligocene, ~~with~~ at overall lower pCO_2. At Site 1263, the absolute nannofossil abundance (coccoliths per gram of sediment; N g^{-1}) and the mean coccolith size decreased distinctly ~~across~~ after the E-O boundary (EOB; 33.89 Ma), mainly due to a sharp decline in abundance of large-sized *Reticulofenestra* and *Dictyococcites*, occurring within a time-span ~~~53~~ 47 kyr. ~~Since c~~Carbonate dissolution did not vary much across the EOB, thus the decrease in abundance and size of nannofossils may ~~highlight~~ reflect an overall decrease in their export production, which could have led to ~~an increased ratio of organic to inorganic carbon (calcite) burial, as well as~~ variations in the food availability for benthic foraminifers.

The benthic foraminiferal assemblage data ~~show~~ are consistent with a~~the~~ global decline in abundance of rectilinear species with complex apertures in the latest Eocene (~34.5 Ma), potentially reflecting changes in the food source, thus phytoplankton~~,~~. This was followed by transient increased abundance of species indicative of seasonal delivery of food to the sea floor (*Epistominella* spp.; ~33.9-33.4 ~~34.04-33.54~~ Ma), with a short peak in overall food delivery at the EOB (buliminid taxa; ~33.~~9~~ 8 Ma). ~~After Oi-1 (starting at ~33.4 Ma), a high~~Increased abundance of *Nuttallides umbonifera* (at ~33.3 Ma) indicates the presence of more corrosive bottom waters, possibly combined ~~with~~ arrival of less food ~~arriving~~ at the sea floor after the second step of cooling~~,~~ (Step 2).

The most important ~~signals~~ changes in the calcareous nannofossil and benthic communities~~, i.e. the marked decrease of large reticulofenestrids, extinctions of planktonic foraminifer species and more pronounced seasonal influx of organic matter, preceded~~ occurred ~120 kyr after the EOB. ~~the major expansion of the Antarctic ice sheet (Oi-1) by ~440 kyr. During Oi-1~~After ~~, our data show t~~There was no major change in nannofossil abundance or assemblage composition at Site 1263 ~~occurred at Site 1263~~ after Step 2, although benthic foraminifera indicate more corrosive bottom waters ~~following~~ during this ~~event~~time. During the onset of latest Eocene-earliest Oligocene climate change, m~~M~~arine phytoplankton thus showed high sensitivity to fast-changing conditions, as well as to possibly enhanced, ~~but~~ pulsed nutrient supply, ~~during the early onset of latest Eocene-earliest Oligocene climate change,~~ or to the crossing ~~a~~ of a climatic threshold ~~in these changes~~ (e.g. pCO_2 decline, high-latitude cooling and changes in ocean circulation).

1 Introduction

The late Eocene-early Oligocene was marked by an important change in global climate and in oceanic environments, reflected in significant biotic turnover~~s in marine and terrestrial biota~~. ~~The~~ Earth's climate was driven from a warm "greenhouse" with high pCO_2 during the middle Eocene through a transitional period in the late Eocene to a cold "icehouse" ~~with~~ at low pCO_2 in the earliest Oligocene (e.g. Zachos et al., 2001; DeConto and Pollard, 2003; Pearson et al., 2009; Pagani et al., 2011; Zhang et al., 2013). During this climate shift, Antarctic ice sheets first reached sea level, sea level dropped, and changes occurred in ocean chemistry and plankton communities, while the calcite compensation depth (CCD) deepened rapidly, at least in the Pacific Ocean (e.g. Zachos et al., 2001; Coxall et al., 2005; Pälike e~~a~~t al., 2006; Coxall and Pearson, 2007; Merico et al 2008). There is ongoing debate whether the overall cooling, starting at high latitudes in the middle Eocene while the low latitudes remained persistently warm until the end of the Eocene (Pearson et al., 2007), was mainly caused by changes in oceanic gateways (opening of Drake Passage and the Tasman gateway) leading to initiation of the Antarctic Circumpolar Current ~~as proposed by (~~e.g. Kennett, ~~(~~1977), or by declining atmospheric CO_2 levels that favored ice sheet growth ~~as proposed by (~~e.g. DeConto and Pollard, ~~(~~2003~~)~~;~~,~~ Barker and Thomas, ~~(~~2004~~)~~;~~,~~ Katz et al., ~~(~~2008;~~)~~ ~~and~~ Goldner et al., ~~(~~2014)~~,~~ in combin~~ed~~ation with specific orbital configurations (Coxall et al., 2005), or by some combination of ~~both~~ these factors (Sijp et al., 2014). Recently, it has been proposed that the glaciation itself caused further oceanic circulation changes (Goldner et al., 2014; Ladant et al., 2014; Rugenstein et al., 2014).

~~The Eocene-Oligocene boundary (EOB; ~33.89 Ma, Gradstein et al., 2012) is formally defined denoted? by the extinction of two planktonic foraminiferal generas (specifically, the genus *Hantkenina* and *Cribrohantkenina*; Premoli Silva and Jenkins, 1993)), and falls within this climate revolution, followed after ~450 kyr by a positive peak in~~ ~~$\delta^{18}O$~~~~, referred to as the Oi-1 event (Miller et al., 1991) which lasted for ~400 kyr and reflects intensified Antarctic glaciation (Zachos et al., 1996; Coxall et al., 2005), probably associated with cooling (e.g. Liu et al., 2009; Bohaty et al., 2012). Pearson et al. (2008), however, recorded the extinction of Hantkeninidae, thus by definition the EOB, in the plateau between the two main steps in the stable isotope records (i.e. within Oi-1) at Tanzania Drilling Project (TDP) Sites 11, 12 and 17. An*Hantkenina* spp. and *Cribrohantkenina* spp. fragmentation and At several well-studied, for example Site ,In such cases, additionalcouldconsidered in order to identify and correlate the EOB between sites thepossible explanation of this inconsistency might be that theThe highest occurrence of *Hantkenina* spp. and *Cribrohantkenina* spp. may be may be influencedaffected by preservation, since the taxon is sensitive to dissolution.~~

Comment [m1]: Moved in section 3.2

~~Recently, several h~~High-resolution~~,~~ benthic foraminiferal δ^{18}O records ~~foraminifera-based geochemical studies~~ across the Eocene-Oligocene transition (~~EOB~~EOT; ~~~33.89~~34-33.5 Ma, Pearson et al., 2008)~~, at different latitudes,~~ have ~~provided~~ shown a two-step cooling at several latitudes ~~detailed information on the stepwise cooling~~ (e.g. Coxall et al., 2005; Katz et al., 2008; Lear et al, 2008; ~~Riesselman et al., 2007; Peck et al., 2010;~~ Coxall and Wilson, 2011; Bohaty et al., 2012). To avoid confusion with previous definitions of these two steps, we follow Pearson et al. (2008) and Bohaty et al. (2012): Step 1 is the first δ^{18}O increase related to global cooling with a modest ice growth component, and Step 2 is the second increase in δ^{18}O representing~~s~~ the major ice growth leading to a continental-scale ice sheet over Antarctica (Miller et al., 2009). Foraminifer-based geochemical studies documented the dynamics of the oceanic carbon cycle during the EOT, with ~~A~~an increase in benthic foraminiferal δ^{13}C ~~δ^{13}C. This δ^{13}C increase~~which, on kyr-time scales, could relate to ~~is a major indication of changes in the carbon cycle, e.g. storage of organic matter in the lithosphere, through~~ an increased ratio in the burial of organic *versus* inorganic carbon (calcite)~~an increased ratio of organic to inorganic carbon (calcite) burial~~ due to enhanced marine export production and/or increased preservation of organic matter- (e.g. Diester-Haass, 1995; Zachos et al., 1996; Coxall and Wilson, 2011). ~~There is, however, evidence that e~~Enhanced export production, however, may not have been~~was not~~ global (e.g. Griffith et al., 2010; Moore et al., 2014). The δ^{13}C shift and oceanic carbon cycle reorganization, linked to increased biological production and deepening of the CCD, have also been related to a rapid drop in pCO$_2$ ~~again linkedwhichthat would have been lead to higher increased biological production and CCD deepening~~ (Zachos and Kump, 2005).

There is a strong link between late Eocene-early Oligocene climate change and the response of ~~the~~ marine and ~~land biota~~terrestrial -biota~~during the late Eocene-early Oligocene~~. The global cooling, with ~~This was a time of~~ high ~~substantial~~ extinction rates ~~extinction~~ and ecological reorganization, ~~in~~ affect~~ing~~ed many biological groups, including: calcifying phytoplankton (coccolithophores; e.g. Aubry, 1992; Persico and Villa, 2004; Dunkley Jones et al., 2008; ~~Tori, 2008;~~ Villa et al., 2008), siliceous plankton (diatoms and radiolarians; e.g. Keller~~ et al.~~, 1986; Falkowski et al., 2004), planktonic and benthic foraminifers (e.g. Coccioni et al., 1988; Thomas, 1990, 1992; Thomas and Gooday, 1996; Thomas, 2007; Pearson et al., 2008; Hayward et al., 2012), large foraminifers (~~*nummulites*~~*Nummulites*; e.g. Adams et al., 1986), ostracods (e.g. Benson, 1975), marine invertebrates (e.g. Dockery, 1986), and mammals (e.g. Meng and McKenna, 1998). Among the marine biota, the planktonic foraminifers experienced a synchronous extinction of five species in the Family Hantkeninidae (e.g. Coccioni et al., 1988; Coxall and Pearson, 2006), the extinction of ~~-~~ *Turborotalia cerroazulensis* group and the reduction in size of the *Pseudohastigerina* lineage

(Wade and Pearson, 2008 and references therein). Benthic ~~foraminiferal~~ foraminifers ~~assemblages recorded~~ experienced a gradual turnover, marked by an overall decline in diversity, largely due to the decline in ~~the relative~~ abundance of cylindrical taxa with a complex aperture (Thomas, 2007; Hayward et al., 2012), and an increase of species which preferentially use fresh phytodetritus delivered to the seafloor in strongly seasonal pulses (e.g. Thomas, 1992; Thomas and Gooday, 1996; Pearson et al., 2008).

~~The calcareous~~ Calcareous nannoplankton assemblages ~~community~~ underwent significant global restructuring ~~changes at~~ during the ~~EOB~~EOT,~~.~~ a~~A~~lthough the group did not suffer extinctions ~~right exaclty~~exactly at the Eocene-Oligocene boundary (EOB) in contrast with~~as the~~ planktonic foraminifers~~-~~.~~, the structure of the assemblages underwent global reorganization.~~ Calcareous nannoplankton -flourished and diversified during the warm-oligotrophic Eocene, with species diversity at maximum during the early-middle Eocene, decreasing during the cold-eutrophic early Oligocene (Bown et al., 2004). Furthermore, coccolithophores were globally more common and widespread in the Eocene, distinctly declining in (common) occurrence since the early Oligocene (Hannisdal et al., 2012). Species diversity decreased ~~through the loss~~at the expenses of ~~K-selective,~~ specialist taxas, ~~taxa and the~~ favoring ~~the increase of abundance of~~ opportunistic species that were~~,~~ more adapted to the new ~~climate/~~environmental~~,~~ ~~increased~~ conditions (e.g. Persico and Villa, 2004; Dunkley Jones et al., 2008; Tori, 2008). ~~Calcareous nannoplankton, overall, flourished and diversified during the warm-oligotrophic Eocene with a maximum in species diversity during the early-middle Eocene rather than during the cold-eutrophic early Oligocene (Bown et al., 2004). Furthermore, coccolithophores were globally more common and widespread during the Eocene, distinctly declining in (common) occurrence since the early Oligocene (Hannisdal et al., 2012). In the late Eocene and Oligocene T~~The decline in diversity ~~displayed by~~inof nannoplankton since the middle Eocene coincided with~~,~~ a diversity ~~when~~increase in~~ the~~ ~~siliceous~~ diatoms, which ~~s become more abundant~~eventually ~~over~~outcompeted the nannoplankton ~~group~~as the dominant phytoplankton group (e.g. Spencer-Cervato, 1999; Bown et al., 2004; Falkowski et al., 2004).~~ Time series analysis (Hannisdal et al., 2012) has confirmed that coccolithophores were globally more common and widespread during the Eocene, declining since the early Oligocene. On million-year time scales, atmospheric CO_2 levels appear to have influenced coccolithophore macroevolution more than related long-term changes in temperature, sea level, ocean circulation or global carbon cycling (Hannisdal et al., 2012).~~

In addition, the late Eocene to early Oligocene decrease in the average cell size of reticulofenestrids (~~presumed~~ ancestors of modern-day alkenone producing coccolithophores *Emiliania huxleyi* and *Gephyrocapsa oceanica*) corresponds to a decline in pCO_2 (Henderiks and Pagani, 2008; Pagani et al., 2011). This macroevolutionary trend appears to have been global and primarily ~~driven~~ ~~realtedby~~ caused by ~~from to~~ the ecological decline of large reticulofenestrid species. Henderiks and Pagani (2008) hypothesized that large-celled coccolithophores were adapted to high pCO_2 and $CO_{2(aq)}$ conditions (late Eocene), whereas small-sized species ~~became~~ are more competitive at lower pCO_2 (early Oligocene). ~~However, t~~This hypothesis has not yet been tested in detail in the fossil record. Culture experiments, however, provide evidence that elevated levels of CO_2 alleviate carbon-limitation in *E. huxleyi* and *G. oceanica*, and that even these small-celled, bloom-forming coccolithophores operate carbon concentrating mechanisms (CCMs) under today's natural conditions (e.g. Rost et al., 2003; Moolna and Rickaby, 2012). The adaptations in algal carbon acquisition due to lower pCO_2 may have occurred as late as during the late Miocene (about 7-5 million years ago; Bolton and Stoll, 2013), suggesting that Paleogene coccolithophores did not (yet) operate CCMs and that diffusive uptake of CO_2 and growth rates were mainly determined by the volume-to-surface area of the cells.

~~Only~~ To date, only few high-resolution studies ~~have~~ describe~~d~~ the response of coccolithophores to environmental change ~~across~~ along the ~~EOB~~ EOT at high- (Southern Ocean; Persico and Villa, 2004; Villa et al., 2008, 2014~~; Fioroni et al., 2015~~) and low latitudes (Tanzania; Dunkley Jones et al., 2008; Fioroni et al., 2015). These studies have highlighted distinct ~~compositional~~ shifts in the composition of the assemblages and ~~changes~~ decreasings in species diversity at or close to the boundary. ~~Here, we present a new high-resolution record (<10,000 kyr across the EOB) from Ocean Drilling Program (ODP) Site 1263, at mid-latitudes in the southeast Atlantic Ocean.~~

Here, we~~We~~ report on calcareous nannofossil and foraminiferal biotic events between 34.~~76~~8-32.7 Ma at Ocean Drilling Program (ODP) Site 1263, recovered in the southeast Atlantic Ocean. In particular, we~~, to~~ refine the shipboard biostratigraphy published in Zachos et al. (2004), including new data on planktonic foraminifers, and describe the ecological response of calcareous nannoplankton and benthic foraminifers to environmental change during the EOT. The ~~calcareous nannofossil assemblages~~ reveal distinct fluctuations in total abundance and ~~species~~ taxonomic composition of the calcareous nannofossil assemblages are~~, which we~~ compared to stable isotope data (Riesselman et al., 2007; Peck et al., 2010), and to benthic foraminiferal assemblage data ~~from the same site~~. For the first time, estimates of the number of nannofossils per gram of dry sediment ~~were~~ are calculated for the Eocene-Oligocene time interval to ~~investigate~~ evaluate how paleo-export fluxes and food supply to the benthic community were affected. This ~~record~~ is also the first high-resolution (<10,000 yr) record ~~to~~ of ~~investigate~~ coccolith size variations (and related changes in mean cell size, cf. Henderiks and Pagani, 2007) across the ~~EOB~~EOT. ~~in greater detail.~~

2 Material and methods

2.1 ODP Site 1263

ODP Leg 208 Site 1263 (28°31.97'S and 2°46.77'E, Atlantic Ocean~~; Fig. 1~~) was drilled at a water depth of 2717 m on the southern flank of Walvis Ridge, an aseismic ridge west of the African coast (Fig. 1). This site provides one of the most continuous sediment sequences of the ~~early~~ lower Cenozoic in the Atlantic Ocean, and was at least 1 km above the lysocline prior to the lowering of the CCD during the E-OT ~~transition~~ (Zachos et al., 2004). Foraminifer-bearing nannofossil ooze and nannofossil ooze are the dominant lithologies in the studied interval (Zachos et al., 2004).

The Eocene-Oligocene sediments of ODP Site 1263 generally have a ~~high~~ high-carbonate content ($CaCO_3$ wt%), ranging from 88 to 96% through 84.2-100.8 mcd (~~Fig. 2;~~ Riesselman et al., 2007). Only a few samples with lower values ~~in~~ of $CaCO_3$ (~87%) ~~have been recorded~~occur ~~prior to the EOB, below the Oi-1 δ^{18}O excursion~~ at 99.19 and 99.49 mcd ~~(Fig. 2; Riesselman et al., 2007)~~ (Riesselman et al., 2007).

A total of 190 samples was used for nannofossil analyses across the EOB ~~in Holes 1263A and 1263B~~. Two datasets, A and B, were independently produced at two laboratories and are here combined in a collaborative effort to also demonstrate whether, and how, the primary nannofossil signals are consistently detected from the same sediment cores independent from sample spacing, microscopy slide preparation and operator. ~~These samples were studied in two sets, A and B.~~ Dataset~~Set~~ A includes 114 samples from 83.19 to 101.13 meters composite depth (mcd). The sampling resolution is high across the EOB (5-10 cm), and decreases above and below it: 20-90 cm between 83.19-89.6 mcd, and 20-50 cm between 97.44-101.13 mcd. Dataset B includes ~~An an additional total of~~ 76 samples ~~were analysed in set B~~ (83.59-105.02 mcd, sampling resolution of 10-50 cm). For analyses on benthic foraminiferal~~al~~ assemblages~~s~~, 27 samples ~~from Hole 1263A~~ between 80.89 mcd to 109.79 mcd were used, while for planktonic foraminiferal analysi~~e~~s 16 samples between 93.42 and 107.29 mcd were ~~analysed~~studied ~~from 1263A-9H-1-32-34cm (80.89 mcd) to 1263A-11H-CC (109.79 mcd)~~(see Table S1, Supplement).

2.2 Microfossil preparation and assemblage counts

2.2.1 Nannofossils

Sample set A was prepared by weighing 5 mg of dried bulk sediment and diluting with 50 mL of buffered water. Then, 1.5 mL of well-mixed suspension was placed on a cover slip with a high-precision pipette, and the sample was dried on a hotplate at 60°C. This technique (called the "drop technique" by Bordiga et al., 2015; modified after Koch and Young, 2007) ~~assures an even distribution of particles, and~~ avoids selective settling effects ~~as~~because ~~all~~ the ~~1.5 mLselected of well-mixed~~ suspension volume ~~was~~is ~~evenly~~ placed evenly on a cover slip and left to settle and dry under low heat (see Bordiga et al., 2015 for details ~~on sample preparation~~). Besides assuring slides with an even particle distribution, ~~I~~this preparation technique also~~, and~~ allows calculation of the absolute coccolith abundances per gram of dry sediment (N g^{-1}). Repeated sample preparation and counting revealed a coefficient of variation (CV) of 6-10% for absolute abundances (Bordiga et al., 2015), which is comparable to other techniques (e.g. Bollmann et al., 1999; Geisen et al., 1999). The drop method also provides a good reproducibility for the relative species abundances (Bordiga et al., 2015). ~~Five samples along the studied sequence were also prepared with the filtration technique (Andruleit, 1996) and spiked with microbeads to investigate the reproducibility of absolute abundances obtained with our technique. This resulted in similar temporal trends between the techniques (mean CV=11%).~~

~~Here~~In this study we ~~used~~report ~~and~~on ~~displayed~~ both absolute (N g^{-1}) and relative species abundances (%). ~~Indeed, the r~~Relative abundances are independent from sedimentological effects and estimates of sedimentation rate (e.g. Gibbs et al., 2012), but in contrast ~~with~~to ~~the~~ absolute abundances %-values~~they are related to the~~represent a closed-sum, as each percentage value refers to how common or rare a species is relative to other species without knowing whether a species truly increased or decreased in absolute abundance. For these reasons a comparison of both ~~absolute and relative abundances allow us to~~~~may help us identify~~is helpful to evaluate the influence of dilution and sedimentation rate variations, and ~~The estimates of absolute abundances (N g^{-1}) allow us to better identif~~identify the real ~~real~~ fluctuations in ~~absolute~~ abundance of single species. ~~within the sediment. In contrast, the use of the relative abundances (%) could lead to loss of information and misinterpretation of the results through are related to the closed-sum problem, as each percentage value refers to how common or rare a species is relative to other species without knowing whether a species truly increased or decreased in abundance.~~. Sample set B was prepared with the standard smear slide technique (Bown and Young, 1998), and the results are given as relative species abundances (%) only.

In both datasets A and B, calcareous nannofossils were examined under ~~crossed~~ polarized light microscopy (LM) at 1000X magnification. ~~Quantitative analyses were performed by counting a~~At least 300 specimens were counted in each slide. Additional observations were performed on the slide to detect the occurrence of rare species, especially biostratigraphical markers. All specimens were identified at species or genus level, depending on the coccolith preservation. We used *Cyclicargolithus* spp. to group the specimens with dissolved central area that can be associated to the genus *Cyclicargolithus* but not directly to the species *Cyclicargolithus floridanus* (Fig. S1, ~~in the~~ Supplement). Taxonomy of the calcareous nannofossils follows the references contained in the web-site http://ina.tmsoc.org/Nannotax3 (edited by Young et al., 2014). Additional taxonomical remarks are given in the Supplement. For dataset A, the number of fields of view (FOV) observed were also noted in order to calculate absolute abundances. An average of 26 FOVs (=0.31 mm^2) was observed along the sequence, from a minimum of 18 FOVs (=0.21 mm^2) to a maximum of 44 FOVs (=0.52 mm^2).

Both datasets were used to provide biostratigraphical information: dataset A with a more detailed resolution across the EOB, and dataset B covering a longer interval below the EOB. ~~For quantitative description of the nannofossil assemblage, relative abundances (%) for all the identified species were calculated for both datasets A and B.~~

2.2.2 Foraminifers

The ~~27~~ samples were oven-dried at 60°C, then washed over a 63 µm sieve. The complete > 63 µm size fraction ~~63 µm~~ was used for the study ~~studied for~~of benthic ~~and planktonic~~ foraminifers. Taxa were generally determined at species level (Fenero et al., 2010) and relative abundances were calculated. The benthic foraminiferal studies were on the number of foraminifers in the full sample. All specimens were picked from material spread out in a picking tray, and mounted on microslides for identification, then deposited in the Department of Earth Sciences, University of Zaragoza (Spain).~~.~~ The planktonic foraminiferal assemblages were observed in the >63 µm fraction to determine the presence of biostratigraphical markers, such as the *Turborotalia cerroazulensis* group and species of the Family Hantkeninidae. The presence or absence of tubulospines was noted (Table S1, Supplement). The reduction in size of the *Pseudohastigerina* lineage was observed by counting the number of *Pseudohastigerina micra* and *Pseudohastigerina naguewichiensis* in a total of 300 planktonic foraminifers in the 150-250 µm and 125-150 µm fractions (cf. Wade and Pearson, 2008; Table S1, Supplement).

2.3 Biotic proxies

2.3.1 Nannofossil dissolution index and cell size estimates

Sample set A was ~~also~~ used to characterize nannofossil dissolution across the investigated interval. A coccolith dissolution index was calculated using the ratio between entire coccoliths and fragments (cf. Beaufort et al. 2007; Blaj et al., 2009; Pea, 2010). This index is indicative of the preservation/dissolution state of the nannofossil assemblages: higher values correspond to better preservation. Entire coccoliths and all fragments were counted until at least 300 entire coccoliths had been counted. Only pieces bigger than 3 µm were considered as fragments.

Mean coccolith and cell size estimates (volume-to-surface area ratio, V:SA; cf. Henderiks and Pagani, 2007; Henderiks, 2008) were calculated based on the relative abundance and size range (3-7 µm, 7-11 µm and 11-16 µm for *Coccolithus*; 3-5 µm, 5-7 µm and 7-9 µm for all the other species) of placolith-bearing taxa (*Coccolithus*, *Cyclicargolithus*, *Dictyococcites* and *Reticulofenestra*)~~ and the different size groups within each (3-7 µm, 7-11 µm and 11-16 µm for *Coccolithus*; 3-5 µm, 5-7 µm and 7-9 µm for all the other species)~~.

2.3.2 Calcareous ~~N~~nannofossil~~s~~ ~~proxies~~paleoecology

The distribution of coccolithophores in sea surface waters is controlled by the availability of light, temperature, salinity and nutrient availability (e.g. Winter et al., 1994). ~~Based on s~~Studies of modern and past paleogeographic distributions of coccolithophores, allow determination of (paleo)environmental tolerances of various taxa ~~may be determined~~ (see Table 3 in Villa et al., 2008). However, some paleoecological ~~labels~~ interpretations remain unresolved, or ~~contrasting~~ contradictory between~~in~~ different regions (see Table 3 in Villa et al., 2008). Therefore, we~~, so our analyses~~ aimed to circumvent problems in interpretation ~~such issues~~ by not tagging certain (groups of) species *a priori*, but instead investigating the behaviour~~s~~ within ~~total~~ assemblages (see Section 2.4) and then compare these with independent proxies (i.e. geochemical ~~data~~ and benthic foraminiferal assemblage data).

2.3.3 ~~Foraminifera-based~~The δ^{13}C gradient in foraminiferal tests

The difference between planktonic and benthic foraminiferal carbon isotope values ($\Delta\delta^{13}C_{p-b}$) was proposed ~~by Sarnthein and Winn (1990)~~ as a semi-quantitative proxy of paleoproductivity (Sarnthein and Winn 1990). It provides information about the surface to deep-water ~~δ^{13}C~~ gradient in δ^{13}C in Dissolved Inorganic Carbon (DIC), reflecting a combination of surface paleoproductivity and ocean circulation and stratification (e.g. Zhang et al., 2007; Bordiga et al., 2013). We calculated the $\Delta\delta^{13}C_{p-b}$ using ~~the foraminifer~~ data in Riesselman et al. (2007) and Peck et al. (2010).

2.3.4 Benthic foraminifers~~al~~ as paleoenvironmental proxies

We determined the relative abundances of benthic foraminiferal taxa, and the diversity of the assemblages, ~~was~~ expressed as the Fisher’s alpha index (Hayek and Buzas, 2010). ~~We used~~ ~~e~~Changes in the relative abundances and diversity were used to infer changes in carbonate saturation state, oxygenation and food supply (e.g. Bremer and Lohmann, 1982; Jorissen et al., 1995, 2007; Gooday, 2003; Thomas, 2007; Gooday and Jorissen, 2012).

The relative abundance of infaunal benthic foraminiferal taxa has been linked to a combination of oxygenation and food supply (‘TROX model; Jorissen et al., 1995, 2007; Gooday, 2003), with high relative abundances reflecting a high food supply, extreme low oxygenation levels, or some combination of both. In addition, calcifying infaunal dwellers may gain an advantage over epifaunal dwellers during deep-water acidification (Foster et al., 2013). We have no sedimentological or stable isotope evidence for low oxygen conditions, and $CaCO_3$% remains high over the studied interval (Riesselman et al., 2007). Therefore, we interpret a high relative abundance of the infaunal, ~~taxa (including the~~ triserial buliminids~~)~~ as indicative of a high, year-round food supply (Jorissen et al., 1995, 2007; Gooday, 2003). High relative abundances of phytodetritus-using taxa indicate an overall more moderate, as well as highly fluctuating ~~but highly (~~seasonally or episodically) flux of non-refractory particulate organic matter (e.g. Gooday, 2003; Jorissen et al., 2007). A~~, and a~~ high relative abundance of *Nuttallides umbonifera* indicates waters ~~which are~~ highly corrosive to $CaCO_3$ in generally low-food supply settings (Bremer and Lohmann, 1982; Gooday, 2003).

Comparisons between past and recent benthic assemblages as indicators for features of deep-sea environments need careful evaluation, because Eocene deep-sea benthic foraminiferal assemblages were structured very differently from those living today, and the ecology even of living species is not well known. For instance, in the Paleogene, taxa reflecting highly seasonal or episodic deposition of organic matter (phytodetritus) were generally absent or rare, increasing in relative abundance during the ~~E-O transition~~EOT (e.g. Thomas and Gooday, 1996; Thomas, 2007). At Walvis Ridge, these species did occur at much lower abundances ~~than~~ during the EOT ~~in the interval studied here during~~ ~~than around~~after Eocene hyperthermal event 2 (Jennions et al., 2015), during the transition from early into middle Eocene (Ortiz and Thomas, 2015) and during the middle Eocene climatic~~e~~ ~~maximum~~optimum (MECO; Boscolo-Galazzo et al., 2015). During the time interval from the early-late Eocene through the EOT their abundance thus increased overall, though episodically and with considerable fluctuations.

In contrast, in the Paleogene cylindrically-shaped taxa with complex apertures (called 'Extinction Group'-taxa by Hayward et al., 2012) were common (e.g. Thomas, 2007). These taxa globally declined in abundance during the increased glaciation of the earliest Oligocene and middle Miocene to become extinct during the middle Pleistocene (Hayward et al., 2012). The geographic distribution of these extinct taxa resembles that of buliminids but differs in detail (e.g. Hayward et al., 2012). These taxa~~, and they~~ were probably infaunal, as confirmed by their $\delta^{13}C$ values (Mancin et al., 2013). It is under debate what caused their Pleistocene extinction and decline in abundance across the EOB (Hayward et al., 2012; Mancin et al., 2013). Changes in the composition of phytoplankton, their food source, have been mentioned as a possible cause, as well as declining temperatures, increased oxygenation or viral infections (Hayward et al., 2012; Mancin et al., 2013).

2.4 Statistical treatment of the nanno~~plankton~~fossil data

Relative species abundances are commonly ~~observed as~~ lognormally distributed~~ions~~ (MacArthur, 1960). To generate suitable datasets for statistical analysis, different transformations yielding Gaussian distributions must be applied, such as log transformation (e.g. Persico and Villa, 2004; Saavedra-Pellitero et al., 2010), centered log-ratio (e.g. Kucera and Malmgren, 1998; Buccianti and Esposito, 2004), arcsine (e.g. Auer et al., 2014)~~, etc~~.

The nannofossil species percentages were used in the statistical treatment to compare the datasets A and B. ~~We applied t~~Two transformations were tested:~~to the nannofossil species percentage abundances:~~ i) log-transformation by log(x+~~-~~1), which amplifies the importance of less abundant species, and minimizes the dominance of few abundant species (Mix et al., 1999), and ii) centered log-ratio (clr) transformation (Aitchison, 1986; Hammer and Harper, 2006), which opens a closed data matrix and retains the true covariance structure of compositional data~~ as well~~. The normal distribution of each species before and after the transformations was verified using SYSTAT 13.0 software. Datasets A and B were treated the same, but ~~were~~ analysed independently.

Principal component analysis (PCA) was performed on the transformed data using the statistics software PAST (PAleontological STatistic; Hammer et al., 2001). Species with an abundance <1% in all samples were not included in the PCA. The PCA (Q-mode) was performed to identify the major loading species and to evaluate the main factors affecting the changes on fossil coccolithophore assemblages.

The closed-sum problem, or constant-sum constraint, may obscure true relationships among variables ~~as first noted by~~(Pearson, ~~(~~1896).~~ when performing statistical data analysis of~~

~~compositional data~~. The clr transformation retains a major problem in carrying out the PCA on the covariance matrix, and the goal of keeping the most important data information with only few principal components (PCs) can fail using clr transformation in associations containing many outliers (e.g. Maronna et al., 2006), as ~~is~~ often the case in nannofossil assemblages. To minimize the presence of outliers we worked with abundant species and groups of nannofossils, instead of with single species.

The PAST software was also used to calculate the Shannon Index, H, a diversity index taking into account a combination of evenness and diversity ~~the relative abundances as well as the number of taxa~~. High values indicate high ~~diversity~~evenness and/or high richness.

3 Biostratigraphy

The EOB at Site 1263 was tentatively placed between 83 and 110 mcd by the Leg 208 Shipboard Scientific Party (Zachos et al., 2004). ~~Riesselman et al. (2007) and later Peck et al. (2010) placed "Oi-1"the Step 2 XXin correspondence of the maximum value recorded by the benthic δ^{18}O.on the basis of an increase in the benthic δ^{18}O records from ~1.5‰ (94.49 mcd, uppermost Eocene) to ~2.6‰ (93.14 mcd, lowermost Oligocene). The δ^{18}O values remained high up section, to 88.79 mcd. Steps 1 and 2 in the δ^{18}O increase were identified by (Riesselman et al., 2007; Peck et al., (2010), although these stepsy are not as clearly defined as at Site 1218 in the Pacific Ocean (Coxall et al., 2005) and Site 522 at Walvis Ridge (Zachos et al., 1996; Coxall and Wilson, 2011). (Fig. 2) See section 3.2 for a detailed discussion on EOB and isotope data.~~

Comment [mb2]: Moved and better discussed in foram section (3.2)

Our high-resolution sampling allowed refining the position of the EOB by locating nannofossil and planktonic foraminiferal bioevents (Fig. 2; Table 1), including some ~~nannofossil~~ bioevents not ~~yet~~ reported in Zachos et al. (2004). ~~To avoid bias sample sets A and B were analysed by two different operators for the occurrence of nannofossil marker species, especially the rare ones (Fig. 2).~~

The identified bioevents are delineated as Base (~~B,~~ stratigraphic lowest occurrence of a taxon), Top (~~T,~~ stratigraphic highest occurrence of a taxon), and Base common (Bc, first continuous and relatively common occurrence of a taxon) ~~according~~ following~~to~~ Agnini et al. (2014).~~, and acme beginning (AB, base of the acme of a taxon) according to Raffi et al. (2006).~~ No correlation with magnetochrons was possible because the ~~soft~~ nannofossil oozes ~~at Site 1263 does~~ did not carry a clear signal (Zachos et al., 2004). The depths of all identified nannofossil and planktonic foraminiferal datums, together with the ages assigned to the most reliable datums as defined in

Pälike et al. (2006) and Gradstein et al. (2012) are displayed in Table 1. ~~For~~ Only one bioevent~~s~~ - the Top of *Isthmolithus recurvus* - is ~~which are diachronous or~~ not reported in Pälike et al. (2006): thus, we adopted the age given in Lyle et al. (2002)~~Gradstein et al. (2012), the most recent literature was selected, considering the datums recorded at latitudes as close as possible to the studied site~~ (Table 1).

Based on the identified bioevents (see below for details), ~~T~~we documented that the studied succession spans from 32.7 Ma (Top~~HO~~ of *I.~~sthmolithus~~ recurvus*, Lyle et al., 2002) to 34.~~76~~ 77 Ma (~~HO~~ Top of *Discoaster barbadiensis*, ~~Gradstein et al., 2012~~Pälike et al., 2006). The estimated average sedimentation rate is ~~9.8~~ 12 m/myr, ~~somewhat lower than~~close to the average value of ~10.8 m/myr in Zachos et al. (2004). In dataset A, where the sample distribution is more homogeneous, the sampling resolution is ~~~~~≤10.000 years across the EOT (from 97.29 to 90.02 mcd).

3.1 Calcareous nannofossils

The results from both datasets A (higher-resolution) and B (longer time interval) render similar biostratigraphical evidence and well-constrained bioevents, especially for the rare species. Using the absolute (N g^{-1}) and ~~the~~ relative (%) abundances of both datasets, we identified nine calcareous nannofossil datums (Fig. 2; Table 1). The studied interval spans from CNE20~~P15b~~ (pars) Zone to ~~CP16c~~ CNO2 (pars) Zone, ~~according~~ in~~to~~ the recent biozonation of ~~Okada and Bukry (1980~~Agnini et al. (2014). The bioevents include:

- Base of *Sphenolithus tribulosus*, the lowermost datum identified (at 103.11 mcd, Table 1). We detected ~~The range for of this bioevent index species (Bown and Dunkley Jones, 2006) is from Zones NP21 to NP23 (biozonation of Martini, 1971), corresponding to CP16-18 Zones (). We detected tT~~this ~~event~~ species at the top of ~~CP15b~~ CNE20 Zone (Fig. 2),~~;~~ ~~is~~ slightly below the range reported by Bown and Dunkley Jones (2006), who~~. They~~ documented it ~~the occurrence of *S. tribulosus*~~ between the NP21 and NP23 Zones (biozonation of Martini, 1971) corresponding to the CNE21-CNO4 Zones (Agnini et al., 2014)~~range)Tori, 2008)~~. At Site 1263, this species is ~~not abundant~~rare and sporadic and ~~its~~ poor preservation of the studied material ~~is commonly~~ compromises~~ing~~ the identification at the species level and thus, possibly, its Base.
- Top of *Discoaster barbadiensis* and *Discoaster saipanensis*. The rosette-shaped discoaster~~id~~s at the bottom of the succession are usually well preserved without overgrowth (Fig. S1,~~ in the~~ Supplement). The Top of *D. barbadiensis* was not ~~identified~~ reported by the Shipboard Scientific

Party (Zachos et al., 2004), and we placed ~~it~~ this bioevent one meter below the Top of *D. saipanensis* (Fig. 2), identified by Zachos et al. (2004) two meters below our datum (Table 1). We placed ~~T~~the Top of *D. saipanensis* at 102.27 mcd because specimens of *D. saipanensis* had been continuously found until 102.52 mcd, although outside the count of 300 specimens (Fig. 2). These two bioevents were usually considered concurrent, but high-resolution studies (Berggren et al., 1995; Lyle et al., 2002; Tori, 2008; Blaj et al., 2009; Fioroni et al., 2015) show that they are ~~not coeval~~shortly spaced. The Top of *D. saipanensis* is used ~~to approximate the EOB and~~ to define the ~~CP15b/CP16a~~CNE20/21 zonal boundary.

- ~~AB~~ Base common of *Clausicoccus* ~~*obrutus*~~ *subdistichus*~~(>5.7 μm)~~. We included *Clausicoccus obrutus* in the *C. subdistichus* concept following Agnini et al. (2014), although *C. obrutus* is the most abundant of the two species at Site 1263 (see Fig. S2, Supplement). The absolute abundance variations~~,~~ together with the relative abundance in the more detailed dataset A, identify the ~~AB~~ Bc at 96.92 mcd, ~2~~01c~~ m below the depth reported by the Leg 208 Shipboard Scientific Party (94.77 mcd; Table 1; Fig. 2) and ~~~1~~60 cm~~slightly~~ above the observed Top of *Hantkenina* spp. and reduction in size of *Pseudohastigerina* ~~-~~(Fig. 2; see the foraminifer~~s~~ section)~~ — i.e. it approximates the EOB (Backman, 1987)~~. ~~AB~~ The Bc of *C.* ~~*obrutus*~~ *subdistichus* defines the base of C~~P16b~~NO1 (Agnini et al., 2014), which corresponds to the upper zone NP21 (Martini, 1971). The Bc of *C. subdistichus* (referred to as *C. obruta*) has been observed shortly after the EOB at Deep Sea Drilling Project (DSDP) Sites 522 and 523 in the SE Atlantic (Backman, 1987)~~,~~ - in the vicinity of Site 1263 - as well as ~~. (Okada and Bukry, 1980) as suggested by Backman (1987). This bioevent increase in abundance is has been well recognized shortly above the EOB also~~ in the Tethys Massignano GSSP and Monte Cagnero sections (Tori, 2008; Hyland et al., 2009), ~~and also~~ at ~~the~~ high-~~-~~latitude~~s~~ Site 1090 (Marino and Flores, 2002) and in the NW Atlantic (Norris et al., 2014).
- Base of *Chiasmolithus altus*. The rare and discontinuous presence of *C. altus* creates some bias in the detection of its Base. Moreover, *C. altus* specimens are highly affected by dissolution as their central-area is commonly completely dissolved (Fig. S1~~ in the ~~, Supplement). The Base of *C. altus* ~~can be~~is tentatively placed ~~with certainty~~ at 89.4 mcd where a specimen with whole central crossbars meeting at 90° was observed (Fig. S1,~~ in the~~ Supplement). At Site 1263, the Base of *C. altus*, the youngest representative of the genus, falls inside the lower Oligocene (Zone ~~CP16b~~CNO1; Fig. 2), as also documented NE Atlantic ~~by~~ (de Kaenel and Villa, ~~(~~1996)~~,~~ and at high-latitudes (Persico and Villa, ~~(~~2004~~)~~;~~, and~~ Villa et al., ~~(~~2008).
- Base and Bc of *Sphenolithus akropodus*. ~~The r~~Rare sporadic occurrence and poor preservation affect the recognition of this species, but ~~B and~~ Bc was~~ere~~ identifiable (Fig. 2; Table 1). We

tentatively placed the Base also, but just few and sporadic species were detected (Fig. 2). The Bc is ~~well related~~ consistent with the ~~first occurrence as~~ identified datum reported in de Kaenel and Villa (1996), who used this bioevent to approximate the Zone NP21/22 ~~(or CP16b/CP16c)~~ boundary, and the Top of *Coccolithus formosus*.

- Top of *Coccolithus formosus*. This bioevent was easily detectable, as *C. formosus* is abundant and well preserved. Its Top defines the ~~CP16b/CP16c~~CNO1/CNO2 zonal boundary (Fig. 2), close to the depth suggested on board ship (Table 1).
- Top of *Isthmolithus recurvus*, the highest datum identified (Fig. 2). Its abundance is low, so that its distribution becomes discontinuous towards the top of the studied interval. The 83.19 mcd depth (Table 1), 3 m above that reported by the Shipboard Scientific Party (Zachos et al., 2004), is an approximation because just one sample above the ~~last~~ highest observed specimens of *I. recurvus* was analysed.

3.2 Planktonic foraminifers

The Eocene-Oligocene boundary (EOB; ~33.89 Ma, Gradstein et al., 2012) is denoted at its Global Stratotype Section and Point (GSSP) at Massignano in Italy by the extinction of the Family Hantkeninidae (specifically of species in the genera *Hantkenina* and *Cribrohantkenina*; Premoli Silva and Jenkins, 1993). Unless well-preserved material is available (as for e.g. the Tanzania Drilling Project (TDP) sites; Pearson et al., 2008), the sensitivity of hantkeninids to fragmentation and dissolution may lead to a misplacement of its true highest occurrence. At several well-studied sites, for example ODP Site 744 (Zachos et al., 1996~~4~~) and Site 1218 (Coxall et al., 2005), hantkeninids are not present. In such cases, additional planktonic foraminifer bioevents must be considered to identify and correlate the EOB between sites: i) the extinction of the *Turborotalia cerroazulensis* group which preceded the EOB (Berggren and Pearson, 2005; Pearson et al., 2008), and ii) the reduction in size of the *Pseudohastigerina* lineage which occurred at the EOB (Wade and Pearson, 2008 and references therein).

At Site 1263, ~~P~~planktonic foraminifers are abundant~~-~~ and their ~~P~~preservation is generally good to moderate. Samples from 109.79 to 99.97 mcd, however, contain strongly fragmented planktonic foraminifers, with non-broken specimens dominated by heavily calcified *Globigerinatheca* spp. (Zachos et al., 2004). ~~At Site 1263,~~ Unfortunately, species of the hantkeninid group are not well preserved, and occur as fragments of variable size, including tubulospines and partial specimens (several chambers). Entire or partially preserved specimens ~~of one of the primary marker species for the EOB, the~~ of hantkeninids as well as loose tubulospines~~, (the genera *Cribrohantkenina* and~~

~~*Hantkenina*)~~ have been observed from the bottom sample (107.29 mcd) up to 97.91 mcd. No specimen of *Hantkenina* spp. nor even tubulospine were seen from 97.14 mcd upward (~~Fig. 2;~~ Table S1, Supplement). ~~are not well preserved, and occur as fragments of variable size, including hantkeninid spines and partial specimens (several chambers). We primarily studied benthic foraminifera, so that we scanned through large samples, containing thousands of specimens of planktonic foraminifera.~~ Therefore, we focused on the Top of *T. cerroazulensis* group (comprising *T. cerroazulensis*, *T. cocoaensis*, and *T. cunialensis*) and the size reduction of the *Pseudohastigerina* lineage, characterized as the Top of >125 µm-sized *Pseudohastigerina micra*. These two bioevents where detected at the same depth as the Top of *Hantkenina* spp., i.e. the three bioevents all fall in between 97.91 and 97.14 mcd (Fig. 2; Table S1, Supplement). Due to the lower resolution of the sampling for planktonic foraminifers than for nannofossils, the three bioevents may not be exactly coeval, but occur with that interval of less than 1 meter~~s~~ (~70 kyr). Nevertheless, we can refine the position of the EOB reported in Zachos et al. (2004), where only core catcher samples were studied, and place the EOB between 97.91 and 97.14 mcd, i.e. at 97.53 mcd (Fig. 2). This position of the EOB is in agreement with the nannofossil bioevent, Bc of *C. subdistichus*, just above that level (96.92 mcd; see Section 3.1).

A further confirmation of this placement of the EOB comes from the benthic foraminifer oxygen isotope data. The EOB occurs between the two main steps in δ^{18}O characterizing the EOT cooling and glaciation ~~in~~ at TDP Sites 12 and 17, where assemblage are pristine (Pearson et al., 2008). At Site 1263, high resolution δ^{18}O data are available only from 96 mcd up. Step 2 is identifiable at 93.4 mcd, at the maximum value of benthic δ^{18}O (Fig., 2; Riesselman et al., 2007; Peck et al., 2010). Step 1 was tentatively placed by Peck et al. (2010) at ~93.8 mcd, but the δ^{18}O curve does not reveal a signal of the first cooling step as clear as at Pacific Site 1218 (Coxall et al., 2005) and nearby Site 522 at Walvis Ridge (Zachos et al., 1996; Coxall and Wilson, 2011). We argue that Step 1 should be placed below 97.53 mcd at Site 1263, not only on the basis of the planktonic foraminiferal and nannofossil bioevents, but also by comparison with the oxygen isotope curve at Site 522, which records a complete and clear δ^{18}O signal for the entire EOT (Fig. 2). The two sites are geographically close, and have comparable sedimentation rate across the EOT (12 m/myr at Site 1263; 9 m/myr at Site 522, Hsü et al., 1984). Because Step 1 and Step 2 occur within ~4 meters at Site 522 (Zachos et al., 1996; Coxall and Wilson, 2011), we can infer that a similar pattern is present at Site 1263, placing the Step 1 between 97.5 and 98.5 mcd (Fig. 2). A δ^{18}O signal similar to the one at Site 522, with Step 1 placed ~2 meters below the EOB~~, but with lower resolution,~~ is recorded at Site 1265 on the Walvis Ridge (lower sampling resolution; sedimentation rate 5.7 m/myr;~~)~~ ~~, where Step 1 can be placed ~2 meters below the EOB (~~Liu et al., 2004). These evidences do not agree with the previous proposed position for Step 1 at only 40 cm below Step 2 (Peck et al., 2010). More oxygen isotope analyses are necessary to definitely place Step 1 in the sediment column at Site 1263.

~~From 96.41 mcd up-section (the first higher sample being at 96.27 mcd) we did not find any fragments of hantkeninid tests and/or loose spines (*Cribohantkenina* and *Hantkenina alabamensis*), whereas these were consistently present in samples below that level (Fig. 2). The sample at 96.41 mcd contained rare spines, but no partial specimens (Fig. 2). We thus recorded the Top of *H. alabamensis*, the traditional marker for the EOB (e.g. Coccioni, 1988; Premoli Silva and Jenkins, 1993; Pearson et al., 2008), at 97.91 mcd~~.~~, and placed the EOB above 96.41 mcd (1263A-10H-5, 32-34cm, 96.27 mcd; Table 1; Fig. 2). The benthic foraminifera at Site 1263 show some evidence of reworking (Zachos et al., 2004), but this was limited to a few samples~~.~~, so we consider that the uppermost sample with partial tests of hantkeninids marks the uppermost Eocene. This observation differs from that in Zachos et al. (2004), where only core catcher samples were studied and the partial specimens in Sample 1263A-10H-CC were not observed (Table 1). Samples from Core 1263A-11H and sample 1263A-10H-CC (99.97-109.79) contain strongly fragmented planktonic foraminifers, with non-broken specimens dominated by heavily calcified *Globigerinatheca* spp. (Zachos et al., 2004).~~

4 Biotic responses

4.1 Calcareous nannofossil preservation and assemblages

At ODP Site 1263 ~~no consistent increase in~~the carbonate content did not increase above the EOB ~~was recorded~~ (Fig. 3; Riesselman et al., 2007), in contrast to other sites, specifically in the Pacific Ocean (e.g. Salamy and Zachos, 1999; Coxall et al., 2005; Coxall and Wilson, 2011).~~,~~ This lack of response is probably due to the location of ~~probably because this s~~Site 1263 ~~was~~ well above the lysocline since the late Eocene (Zachos et al., 2004), so that~~. The carbonate accumulation was not strongly affected by potential CCD deepening, because the~~ CaCO$_3$ (wt%) was and remained generally high, and was not affected by CCD deepening (Fig. 3; Riesselman et al., 2007). The deeper Site 1262, close to Site 1263, was below the lysocline before the rise in CCD, and shows a strong increase in CaCO$_3$ (wt%) across the EOB (from ~5 to > 90%; Liu et al., 2004).

However, t~~T~~he CaCO$_3$ (wt%) at Site 1263 does not reflect the total coccolith absolute abundance (Fig. 3). This~~,~~ ~~suggesting~~ supports that ~~also~~ other calcifying organisms (mainly planktonic foraminifers) contributed consistently to the calcite accumulation in the sediments. To unravel the "true" contribution of each calcifying group to the accumulated $CaCO_3$ (wt%), we need to know the total amount of carbonate produced by calcareous nannoplankton and foraminifers, which is beyond the scope of this study.

Although the site was above the lysocline during the studied time interval, the nannofossil and foraminiferal assemblages show signs of dissolution ~~all~~ throughout ~~along~~ the sequence. ~~Such~~ ~~d~~Dissolution may occur above the lysocline (e.g. Adler et al., 2001; de Villiers, 2005), leading to a reduction in species numbers and an increase of fragmentation with depth, in both nannoplankton (e.g. Berger, 1973; Milliman et al., 1999; Gibbs et al., 2004) and planktonic foraminiferal ~~communities~~ assemblages (e.g. Peterson and Prell, 1985).

At Site 1263 signs of dissolution were detected, in particular, i~~o~~n specimens of *Cyclicargolithus* (Fig. S1, ~~in the~~ Supplement) – one of the least resistant nannoplankton species (Blaj et al., 2009), but also i~~o~~n more robust species ~~like~~ such as *Dictyococcites bisectus* ~~(Fig. S1 in the Supplement)~~. Despite these signs, holococcoliths and abundant small-medium sized *Cyclicargolithus* – which are prone to dissolution (Young et al., 2005; Bown et al., 2008; Blaj et al., 2009) – are present in all samples. We did not see small placoliths (<3 µm) at Site 1263 ~~(<3 µm)~~, possibly due to dissolution, but these were not dominant in the late Eocene (e.g. Persico and Villa, 2004; Villa et al., 2008; Fioroni et al., 2015). The lack of such placoliths does not ~~but does not~~ prevent the identification of the main features ~~in~~ of the medium-large sized ~~assemblage~~taxa. ~~To have more information on dissolution we calculated the~~

Our coccolith dissolution index~~, which~~~~The coccolith dissolution index~~ does not show any ~~large~~ major changes across the EOT (91-98.5 mcd)~~t the EOB~~, but at 90.2 mcd and from 87 mcd upward~~during and after the Oi-1~~ nannofossil dissolution slightly increased~~tensified~~ (Fig. 3). The correlation between the dissolution index and total coccolith abundance is positive (entire interval r = 0.32; *p*-value = 0) and stronger in the upper interval of the studied sequence (r = 0.59; *p*-value = 0.002), but not significant across the EOB~~. In fact~~~~For example, from 90.5 mcd upward the correlation value, r = , is 0.59 (*p*-value = 0.002), instead for the entire interval r = 0.32 (*p*-value = 0)~~. ~~i~~Intervals of increased dissolution do~~id~~ not necessarily correspond to lower ~~values in the total~~ absolute abundances, so that ~~This confirms that the total coccolith abundance and~~ we can infer that ~~the~~ primary signals of the ~~nannofossil~~ nannoplankton ~~assemblage features~~ are preserved in the fossil ~~record~~ assemblages~~,~~ at least across the EOB, with the exception of the primary presence/absence of small specimens.~~, although nannofossil intervals of higher dissolution may be intensehave affected the nannofossil assemblage. (at the episode recorded From at 90.5 mcd mcd up-section, dissolution appears to have more strongly affected the assemblages slightly more .~~

~~The t~~T~~otal absolute coccolith abundance~~s ~~records~~ show ~~a marked decrease across the EOB: within 60 cm (from 96.39 to 95.79 mcd) the abundance rapidly drops by 45%, mainly driven by the loss of large-sized species, in particular of *D. bisectus* (Fig. 3).~~

Nannofossil diversity, as expressed in ~~based on~~ the H index, does not ~~record~~ vary significantly ~~variations at~~across the EOB, but decrease gradually~~.~~ within 1.5 m above the EOB. A more distinct step-wise decrease ~~is recorded~~ at 90 mcd (~~grey bar in~~ Fig. 3)~~)~~ reflects a community structure with fewer dominant species, possibly due ~~which could be explained by the~~to increased dissolution in this interval, and by a community structure with fewer dominant species. Actually, ~~in this interval~~ *Cyclicargolithus* became ~~more~~ dominant in ~~the assemblage~~this interval, while large *Reticulofenestra* decreased in abundance significantly (Fig. 3). The calcareous nannofossil assemblage variations recorded in sample sets A and B are comparable despite the different sampling resolution (Figs. S2 and S3,~~ in the~~ Supplement).~~Total absolute coccolith abundances show a marked decrease ~1.5 m above the EOB: within 60 cm (from 96.39 to 95.79 mcd) the abundance rapidly drops by 45%, mainly driven by the loss of large-sized species, in particular of *D. bisectus* (Fig.~~

~~By comparing~~The trends in ~~the~~ absolute and relative abundances~~, we documented that their trends~~ are very similar (Fig. S2,~~ in the~~ Supplement). Thus, we ~~can infer~~conclude that the dilution/sedimentation rates at Site 1263 were close to constant over time, and that the variations in absolute abundance were linked to biological processes. Total absolute coccolith abundances show a marked decrease ~1.5 m above the EOB (Fig. 3): within 60 cm (from 96.39 to 95.79 mcd) the abundance rapidly drops by 45%, mainly driven by the loss of large-sized species, ~~, in particular of *D. bisectus* (Fig. 3).The absolute abundances of all~~ most ~~the large-sized species decreased markedly across 1.5 m above the EOB (Fig. 3),~~ including ~~the species~~ *D. bisectus*, *Dictyococcites stavensis*, *Reticulofenestra umbilicus*, *Reticulofenestra samodurovii*, *Reticulofenestra hillae*, and *Reticulofenestra* ~~sp.1~~*circus* group (see taxonomical remarks in the Supplement)~~, and *Reticulofenestra daviesii*~~. Among these, *D. bisectus* and *D. stavensis* constitute a significant part (up to 28%) of the assemblage. The medium-~~-~~sized *Reticulofenestra daviesii* also shows a decrease ~1.5 m above the EOB, contrary to what was reported at ODP Site~~s~~ 744 (Persico and Villa, 2004), Site 748 (Villa et al., 2008), Site 711 (Fioroni et al., 2015), and Site 1090 (Pea, 2010~~-~~) for the same time interval.

The small-medium *Cyclicargolithus* spp. and *C. floridanus* are the most abundant species (up to 50%), and the 5-7 µm size group is dominant. This group increases ~~in absolute abundance~~ slightly from the bottom ~~upwards~~~~of the studied section up~~upwards,~~-~~ and just above the EOB it records an increase in abundance~~then it increases more, but at the EOB only a slight decrease in absolute abundance is recorded~~. ~~Also,~~ *Coccolithus*~~.~~ *pelagicus* is ~~one of the most~~another important component~~s~~ of the nannofossil assemblage, at a maximum abundance of 27% (Fig. 3). This species increases ~~its~~ in ~~absolute~~ abundance between 96.~~79~~92-92.6 mcd, i.e. ~~across and~~ above the EOB, and then it decreases from 88 mcd upwards. *Sphenolithus* spp. ~~also~~ does not show any marked variation at the EOB, even if this group is not very abundant. The increase of *Cyclicargolithus* and *C. pelagicus* does not coincide with the marked decrease of large reticulofenenstrids indicating ~~: tThis lack of any species, that increase in abundance above the EOB at Site 1263 suggests~~ that the loss of the latter group ~~in large reticulofenestrids~~ was not compensated for by other taxa. ~~leading to a sustained decrease in~~The total coccolith abundance (and export production) thus decreased ~~since~~ above the EOB.

Another component of the assemblage, *Lanternithus minutus*, is generally not abundant, but peaks between 89.6 and 87.12 mcd. *Zygrablithus bijugatus* and *Discoaster* spp. both decreased in abundance ~~before~~ below the EOB (at 98 and 99 mcd, respectively) and~~, thereafter,~~ higher in the section never reached abundances as high as in the ~~late~~upper Eocene (Fig. 3).

4.1.1 Principal component analysis

~~The~~ Results from the PCAs performed on datasets A and B ~~give fairly~~are comparable~~ results~~, ~~either~~ both using the log- or clr-transformation. For dataset A, the Pearson correlation value (r) between the components from the two transformations is 0.90 (p-value=0), confirming that the primary signals in the assemblage are ~~revealed~~ reflected in~~by~~ the multivariate statistical analysis, as long as ~~the~~ normal distribution of the species is maintained. We also compared the PCA results with or without the presence of the marker species, because stratigraphically-controlled species are not distributed along the entire succession, thus affecting PCA outcomes (e.g. Persico and Villa, 2004; Maiorano et al., 2013). ~~Nonetheless, t~~The results obtained with and without the marker species ~~still~~ provide similar trends for both datasets because in the studied interval the marker species are not very abundant (Fig. 4; Table ~~S1~~S2~~, in the~~ Supplement).

In the following discussion, we will focus on the PCA results and the loading species using the log-transformation for datasets A and B (Fig. 4; Tables ~~S1~~ S2 and ~~S2~~S3~~, in the~~ Supplement). The only two significant principal components explain 50% of the total variance in dataset A, and respectively account for 36% and 14%. For dataset B the two components explain 35% (26% and 11% respectively).

Principal component 1 (PC1) of dataset A shows positive values below 96 mcd. A pronounced decrease occurs ~~at~~ 1.5 m above the EOB, and from 96 mcd upwards the PC1 maintains mainly negative values (Fig. 4a). PC1 is negatively loaded by *C. obrutus*, *C. floridanus* small and medium size, and positively by *D. stavensis*, *D. bisectus*, *R. daviesii*, and *R. umbilicus* (Fig. 4a; Table ~~S1~~S2, ~~in the~~ Supplement). The loadings of the other species are too low to be significant. The PC1 of dataset B does not record the same marked drop ~~at~~ above the boundary, but rather a gradual decrease ~~all~~ along the whole sequence (Fig. 4a). Although the main loading species are the same for both datasets (i.e. *C. obrutus*, *Cyclicargolithus* versus *D. bisectus* and *R. umbilicus*), there are some differences ~~can be identified~~ (Tables S2 and ~~S2~~S3, ~~in the~~ Supplement). ~~In particular~~Specifically, the size groups ~~influence~~ of *Cyclicargolithus* ~~size groups~~ do not influence ~~on~~ PC1 in dataset B ~~cannot be detected in dataset B~~ because the size subdivision was not included in the counts ~~in~~of that dataset. As the distribution of large *versus* small-medium sized species on the PCA seems to be important for both datasets, and *Cyclicargolithus* is one of the most abundant species, ~~it is possible that~~ the lack of a detailed size grouping within this genus in dataset B ~~could lead~~ might be the cause of the~~to the~~ difference in the PC1 curves ~~at~~ above the EOB. The higher abundances of *Discoaster* and *R. umbilicus* from the bottom up to 102 mcd in dataset B could also explain some differences in the loading species between the two datasets (Tables ~~S1~~ S2 and ~~S2~~S3, and Fig. S3, ~~in the~~ Supplement).

Principal component 2 (PC2) of dataset A also records an abrupt variation ~~across~~ above the EOB (at 96 mcd): the negative values at the bottom of the succession turn toward positive values above the boundary, ~~and~~ remaining positive up to 89.95 mcd. From ~~89~~ 90 mcd upwards, PC2 displays mainly negative values ~~again~~, except for a peak between 85.68-86.42 mcd (Fig. 4b). The most meaningful species loading on PC2 is *L. minutus* (negative loading). The PC2 is also loaded negatively by *D. stavensis* and *C. floridanus* (5-7 µm), and positively by *C. pelagicus* (3-7 µm and 7-11 µm), *I. recurvus* and *Sphenolithus* spp. (Fig. 4b; Table ~~S1~~ S2, ~~in the~~ Supplement). The PC2 for dataset B shows a trend similar ~~trend as~~ to that for dataset A from 98 mcd upward (Fig. 4b), but it distinctly differs in the lower part of the succession. Again, the PC2 is resolved by the same main loading species *L. minutus* versus *C. pelagicus*, ~~(~~but ~~note that~~ the relative direction (positive or negative) of the loadings is ~~swapped~~ reversed between datasets A and B~~;~~ (Tables ~~S1~~ S2 and ~~S2~~S3, ~~in the~~ Supplement). In particular, *L. minutus* has very strong loadings in both datasets. In dataset B, *L. minutus* has its maximum abundance in the upper Eocene interval ~~that was~~ not sampled in dataset A (Figs. S2 and S3, ~~in the~~ Supplement), likely driving the differences between the two PC2 curves below the EOB (Fig. 4b). ~~The distribution of *L. minutus* becomes more comparable between the~~

~~datasets above 100 mcd, reaching a peak between 89.6 and 87.12 mcd although not as high as during the upper Eocene (Figs. S2 and S3, in the Supplement).~~

In the following discussion, we used the PCA results for dataset A (without ~~the~~ marker species) only, because of its more even sample distribution and direct comparison to the other available nannofossil proxies, i.e. dissolution index, coccolith size distribution and absolute abundance.

4.2 Mean coccolithophore cell size variations

The PC1 curve is mirrored (r=0.~~81~~79; *p*-value=0) by mean cell size estimates (V:SA ratio) of all medium to large-sized (>3 µm) placolith-bearing coccolithophores within the assemblages and of those all ancient alkenone producers combined (i.e. *Cyclicargolithus*, *Reticulofenestra* and *Dictyococcites*; Plancq et al., 2012)~~(~~ (Fig. 5). Fluctuations in mean size are mainly driven by the relative abundance of the different placolith-bearing taxa and their respective size groups, rather than by intra-specific size variations. The mean V:SA ratios were higher (species with large cells were more abundant) during the latest ~~Eocene~~Eocene and early Oligocene, and the size decreased (due to the loss of large species) by 8% ~~across~~ between 96.39 to 95.79 mcd (within ~47 kyr), which is according to our age model ~120 kyr after the EOB.~~, within 60 cm above (from 96.39 to 95.79 mcd), or ~53~~ 47 ~~kyr.~~

The~~Our~~ coccolith dissolution index confirms that preferential dissolution ~~of small species~~ did not bias the V:SA results, as intervals of increased dissolution did not generally correspond to large V:SA (r = -0.12). The only exception is the top, 90-90.3 mcd, interval where a high dissolution peak corresponds to an increase in mean size. In either case, the above V:SA considerations do not include small placoliths (<3 µm), so that our analysis is free from any bias due to the (original) presence or absence of this most dissolution-prone group.

4.3 Benthic foraminiferal assemblage

Benthic foraminifers show partial dissolution or etching, especially between 94.42 mcd and 109.79 mcd, but are generally well preserved, i.e. sufficient for determination at species level (Fenero et al., 2010). The ~~low~~ low-resolution data on benthic foraminifers~~a~~ show that the diversity of the assemblages (~~see~~ Fisher's alpha index curve; Fig. 6) started to decline in the late Eocene (~34.5 Ma; 102.79 mcd), reached its lowest values just below the EOB, then slowly recovered, but never to its Eocene values (Fenero et al., 2010). The decline in diversity was due in part to a decline in relative abundance of the generally rare but species-rich group of rectilinear species with complex apertures ('extinction group' species). Such a decline is observed globally at the end of the Eocene (Thomas, 2007; Hayward et al., 2012). The declining diversity (decreased evenness) was also due to a transient increase in abundance of species indicative of seasonal delivery of food to the sea floor (phytodetritus species, mainly *Epistominella* spp.; ~~~34.04~~33.9-33.~~51~~ 4 Ma; 97.91-91.91 mcd), with a short peak in overall, year-round food delivery ~~at~~ above the ~~E/O boundary~~EOB (buliminid taxa; ~33.~~9~~ 8 Ma; 96.41-96.27 mcd). From ~3 meter ~~After Oi-1~~ above Step 2 (~~(starting at~~ ~33.~~4~~ 3 Ma; 90.41 mcd) up, the abundance of *N. umbonifera*, an indicator of carbonate corrosive bottom waters, increased. Due to this evidence for dissolution, benthic foraminiferal accumulation rates can-not be used to estimate food supply quantitatively and reliably throughout the studied interval.

Evidence for effects of dissolution on benthic foraminiferal assemblages is seen in the interval where *N. umbonifera* is common, but not in the interval with peak abundance of phytodetritus species (thin-walled, solution-sensitive species), and the interval where buliminids peak. These intervals are also not recognized as influenced by carbonate corrosivity in the pore waters (Riesselman et al., 2007). Thus, we conclude that the increased percentage of infaunal taxa is, in this studied section, not due to dissolution, although such an effect is seen in sections with much more severe dissolution effects, such as the Paleocene-Eocene interval of ocean acidification (Foster et al., 2013). Increased abundance of buliminid taxa (though not of phytodetritus taxa) could possibly result from lowered oxygen conditions in bottom or pore waters (e.g. Jorissen et al., 2007). However, there are no indications in the sediment of low oxygen conditions (e.g. lamination), and the overall benthic assemblage does not indicate low oxygen conditions (e.g. the diversity is too high).

5 Discussion

5.1 Nannoplankton abundance and cell size decrease ~~at~~ after the EOB

The distinct variation in nannoplankton abundance and average ~~coccolith~~ size of medium to large placoliths above the EOB at Site 1263 cannot be explained by dissolution – which would affect smaller coccoliths preferentially and lead -to an increase of the mean size of the whole assemblage, opposite to what is observed. It can also not be explained ~~– nor~~ by a change in species diversity, but is mainly linked to changes in community structure (Fig. 3). The drop in total nannofossil abundance (Fig. 3) and mean cell size (Fig. 5) is mainly driven by the decrease in abundance of large *Reticulofenestra* and *Dictyococcites* ~~across~~ 1.5 m (~120 kyr) above the EOB. The mean V:SA estimates for all ancient alkenone producers combined (i.e. *Cyclicargolithus*, *Reticulofenestra* and *Dictyococcites*; Plancq et al., 2012) tightly overlap (Fig. 5) with biometric data of the same group in the Equatorial Atlantic (Ceara Rise, ODP Sites 925 and 929; Pagani et al., 2011; Zhang et al., 2013), while the mean size estimates for combined *Reticulofenestra* and *Dictyococcites* remained relatively stable and coincide with mean values measured at ODP Site 1090 in the Subantarctic Atlantic, where *Cyclicargolithus* spp. were not present and assemblages are likely severely affected by dissolution (Pea, 2010; Pagani et al., 2011). This highlights that the observed patterns in average placolith size at Site 1263 are driven by the decrease in abundance, rather than (intra-specific) size variations of *Reticulofenestra* and *Dictyococcites*.

The assemblages also ~~records~~ illustrate the mid-latitude location of Site 1263, hosting both "subantarctic" and "equatorial" taxa. A striking correspondence ~~with~~ between the mean V:SA of ancient alkenone producers at Site 1263 and Sites 929 and 925 (Fig. 5) would suggest more affinity with tropical assemblages than with high-latitude ones, south of the Subtropical Convergence (STF). The abundance patterns of the larger reticulofenestrids, however, are ~~strikingly~~ more similar to those at Southern Ocean sites (Persico and Villa, 2004; Villa et al., 2008). The mid-latitudinal Site 1263 thus probably records paleobiogeographic patterns in the nannofossil assemblage intermediate between those in equatorial-tropical and subantarctic regions.

The coccolith size-shift and the decreased abundance of large reticulofenestrids ~~across~~ after the EOB may be related to different bio-limiting factors. Under growth-limiting environmental conditions, phytoplankton (coccolithophores) with small cell volume-to-surface area ratios may outcompete larger cells due to lower resource requirements (lower C, P and N cell quota) and generally higher growth rates (e.g. Daniels et al., 2014). A change in overall nutrient regime, such as in coastal upwelling *v~~s.~~ersus* oligotrophic, stratified gyre systems, may also cause a shift in opportunistic *versus*~~vs.~~ specialist taxa (e.g. Falkowski et al., 2004; Dunkley Jones et al., 2008; Henderiks et al., 2012). The 16-37% absolute abundance declines of the reticulofenestrid species ~~*D. bisectus*,~~ *R. umbilicus*, ~~and~~ *R.* ~~*hillae*~~ *samodurovii R. hillae* and *R. circus* group ~~and *R. daviesii*~~ (Figs. 3 and ~~3~~S2, Supplement), are strong indications that these large-celled coccolithophores were at a competitive disadvantage already during or shortly after the EOB. Earlier biometric studies of reticulofenestrid coccoliths point to a similar scenario (Fig. 5), postulating that the macroevolutionary size decrease reflects the long-term decline in pCO_2 (Henderiks and Pagani, 2008; Pagani et al. 2011; Hannisdal et al., 2012). High CO_2 availability during the late Eocene could have supported high diffusive CO_2-uptake rates and photosynthesis even in the largest cells, assuming that Paleogene ~~ancient~~ coccolithophores had no or inefficient CO_2-concentrating mechanism, similar to modern species today (Rost et al., 2003; Bolton and Stoll, 2013), and due to the fact that RUBISCO~~ubisco's~~ specificity for CO_2 increases at higher CO_2 levels (Giordano et al., 2005).

Available paleo-pCO_2 proxy reconstructions from Equatorial regions (Pearson et al., 2009; Pagani et al., 2011; Zhang et al., 2013) indicate a transient decrease in pCO_2 across the studied interval, rather than a distinct drop in pCO_2 ~~at~~ after the EOB, which ~~would~~ appears to be ~~suggested~~ supported by our high-resolution assemblage (PC1) and mean V:SA time series (Fig. 5). ~~Nevertheless, t~~The paleo-pCO_2 proxy data, however, are at much lower time resolution, based on a range of geochemical proxies and assumptions (Pearson et al., 2009; Pagani et al., 2011; Zhang et al., 2013). Therefore ~~, and may therefore~~they may not record the drop in pCO_2 as accurately as our comparative analysis would require. The range of estimated pCO_2 values is fairly wide: mean values are 940 ppmv ~~below~~ before the EOB (standard deviation range 740-1260 ppmv) and 780 ppmv ~~above~~ after the boundary (s.d. range 530-1230 ppmv) (Pearson et al., 2009; Pagani et al., 2011; Zhang et al., 2013; Fig. 5).

Possibly, ~~during~~ shortly after the EOB a threshold level in pCO_2 was reached, below which large reticulofenestrids became limited in their diffusive CO_2-uptake, or other, fast-changing (a)biotic environmental factors limited the ecological success of th~~e same~~ is group. On million-year time scales, atmospheric CO_2 levels appear to have influenced coccolithophore macroevolution more than related long-term changes in temperature, sea level, ocean circulation or global carbon cycling (Hannisdal et al., 2012). Between biotic and abiotic factors, the latter (i.e. nutrient supply, temperature, salinity, etc.) are deemed to be dominant (Benton, 2009), and may have led to a more successful adaptation of the smaller taxa at the expense~~s~~ of ~~the~~ large ones (see discussion below, Section 5.2).

This would not exclude a transient, long-term pCO_2 forcing on coccolithophore evolution (Hannisdal et al., 2012). Interestingly, the decline of large *R. umbilicus* occurred earlier at Site 1263 (~~across the EOB~~ ~33.~~89~~ 8 Ma) than at higher latitudes in the Southern Ocean (~~just above the EOB:~~ ~33.3 Ma at Site 689, Persico and Villa, 2004; ~33.5 Ma at Site 748, Villa et al., 2008). A similar pattern is documented in the timing of its subsequent extinction, occurring earlier at low- and mid-latitudes (32.02 Ma; ~~Gradstein et al., 2012~~Pälike et al., 2006) and later ~~in~~ at high latitudes (31.35 Ma; Gradstein et al., 2012). Henderiks and Pagani (2008) suggested that the generally higher content of CO_2 in polar waters may have sustained *R. umbilicus* populations after it had long disappeared from the tropics.

5.2 Paleoproductivity at Site 1263: nannoplankton and benthic foraminifer signals

At Site 1263, no other phytoplankton than calcareous nannoplankton was detected, and diatoms were ~~also~~ absent in coeval sediments at near-by ~~Deep Sea Drilling Program (~~DSDP~~)~~ Walvis Ridge Sites ~~525~~522-529 (Hsü et al., 1984; Moore et al., 1984). Therefore, our inferences of paleo-primary productivity and export production are based on the nannoplankton and benthic foraminiferal assemblages.

PC2 of the calcareous nannoplankton analysis could be correlated with paleoproductivity and total water column stratification. The strongest negative loading on PC2 is the holococcolith *L. minutus* (Fig. 4b; Table S1, ~~in the~~ Supplement). In modern phytoplankton, the holococcolith-bearing life stages proliferate under oligotrophic conditions (e.g. Winter et al., 1994). Moreover, holococcoliths such as *L. minutus* and *Z. bijugatus* are quite robust (Dunkley Jones et al., 2008), so that dissolution is unlikely to affect their distribution~~ which may be mainly linked to low nutrient availability~~.

The positive loadings on PC2 are the species *C. pelagicus*, *I. recurvus* and *Sphenolithus* spp. A high abundance of *C. pelagicus* has often been considered as indicative for warm-to-temperate temperatures at high-latitudes (e.g. Wei and Wise, 1990; Persico and Villa, 2004; Villa et al., 2008). In the modern oceans, *C. pelagicus* seems to be restricted to temperate-to-cool ~~-~~water, high-nutrient conditions (e.g. Cachao and Moita, 2000; Boeckel et al., 2006), but during the Paleogene it was cosmopolitan (Haq and Lohmann, 1976). The paleoecological preferences of *Sphenolithus* are still controversial, but it has been related to oligotrophic conditions inferring a major nutrient control ~~over~~rather than temperature control on this species during the Paleocene-Eocene thermal maximum (PETM; Agnini et al., 2006) and the EOT (Villa et al., 2008). Increased abundances of *Sphenolithus* have been also related to high-productivity intervals in the early Oligocene (Wade and Pälike, 2004) and across the EOT (Dunkley Jones et al., 2008).

We compared PC2 with the proxy for regional paleoproductivity $\Delta\delta^{13}C_{P\text{-}B}$ (Fig. 6), with lower values corresponding to lower productivity and/or higher stratification.~~ The~~ $\Delta\delta^{13}C_{P\text{-}B}$ data are not available for the interval below 96 mcd (upper Eocene-lower Oligocene), but lower paleoproductivity in general corresponds to negative loadings on PC2, and vice versa. The correlation coefficient between the two curves is 0.33 (*p*-value =0.05), i.e. a significant but not~~ a~~ very strong correlation, possibly due to the lower number of stable isotope data points than nannofossil data points. We infer that PC2 probably reflects lower productivity during the latest Eocene, ~~and~~ with both PC2 and $\Delta\delta^{13}C_{P\text{-}B}$ curves showing ~~a~~ higher productivity ~~signal~~ within ~~at the~~

~~onset of Oi-1~~the EOB and the onset of Step 2 (Fig. 6). In particular, PC2 records a longer interval of positive loadings (higher productivity) ~~above~~ after the EOB, and an initial decrease ~~before in~~ corresponding to~~ence of~~ the highest peak in $\delta^{18}O$ (at~93 mcd; ~33.5~~6~~ Ma), as recorded also by $\Delta\delta^{13}C_{P-B}$. According to the $\Delta\delta^{13}C_{P-B}$, p~~P~~aleoproductivity remained constant above 90 mcd upward, and lower than below Step 2.~~subsequently remained lower from the end of Oi-1 upward.~~ The ~~PC2~~ different trend ~~by~~in PC2 ~~curve~~ from 90 mcd ~~and $\Delta\delta^{13}C_{P-B}$ curves differ from 90.5 mcd~~ upward may be~~, possibly~~ related to increased nannofossil dissolution, in particular above 87 mcd. The increase of dissolution is confirmed by the increased abundance of the benthic foraminifer species *N. umbonifera* ~~(Fig. 6)~~, indicative of more corrosive bottom waters, and ~~or possibly a lower food supply. This is thus in agreement with~~ the intensified dissolution interval recorded by the coccolith dissolution index (compare Figs. 3 and 6).

The benthic foraminiferal assemblage confirms the above interpretation of the PC2, adding information on the nature of the ~~nutrient~~ supply of organic matter to the seafloor, i.e. export productivity (Fig. 6). The increase ~~across the EOB~~in abundance of the phytodetritus-~~consuming~~using species across the EOB indicates an increase in seasonality of food delivery ~~of food~~ to the seafloor, correlated to the interval with positive scores o~~i~~n PC2 (Fig. 6). The interval was~~, though~~ interrupted by a short ~~interval~~ period of increased productivity across the EOB (as showed by the peak in the buliminid species curve at 96.27 mcd; Fig. 6), indicating~~-~~ high, less seasonally interrupted food supply. Seafloor conditions changed ~~A~~after ~~the Oi-1~~Step 2, when the high abundance of *N. umbonifera* and the decrease of phytodetritus and buliminid species ~~are indicative~~ indicate ~~of~~ more corrosive bottom waters, possibly combined with less food arriving at the sea floor and~~/or~~ a less pronounced seasonality (Fig. 6).

~~The v~~Variations in nutrient supply to the photic zone, as reflected in ~~both~~ nannofossil ~~and benthic foraminifer assemblages~~, is a factor that could possibly have ~~driven~~combined with the declining pCO_2 to cause the decrease in the mean coccolith size ~~decrease across~~ after the EOB. ~~In fact, t~~The transient higher availability of nutrients ~~at the~~ between the EOB and the onset of Step 2~~Oi-1~~ (~330 kyr), may have ~~made it possibly for~~ given small opportunistic nannoplankton species a competitive advantage over ~~above the EOB to outcompete~~ large specialist species after this time. The decrease of mean cell size (less biomass per individual) and overall~~, also,~~ decrease in ~~of total~~ nannofossil abundance could have led to less available organic matter or less efficient ballasting of organic matter during transport to the sea floor and, ~~thus,~~ less food for the benthic foraminifers.~~, and smaller nannoplankton could have caused a decrease in delivery of organic matter to the seafloor (and/or higher remineralization).~~ If the smaller size led to decreased efficiency in ballasting, the time of transport from surface to the sea floor could have increased, making remineralization more efficient despite the declining temperatures. Ecosystem structure is the main determinant of efficiency of transfer of organic matter to the sea floor (e.g. Henson et al., 2012), and such important changes as observed in the nannofossil assemblages could have strongly impacted transfer of food to the seafloor, hence benthic foraminiferal assemblages, and influenced the decline in 'Extinction Group' species (Hayward et al., 201~~3~~2; Mancin et al., 2013).

Possibly, ~~major~~ climate-driven instability of the water column ~~during the~~ within 330 kyr after the EOB ~~onset of Oi-1~~ favoured seasonal or episodic upwelling, thus primary productivity in this area, which may also be reflected by the (slightly) increasing trends in ~~but an increase in productivity at the Oi-1 onsetis not reflected in the~~ absolute ~~coccolith~~ abundance of (medium-sized) *Cyclicargolithus* spp., *C.~~occolithus~~ pelagicus* and *Sphenolithus* spp. (Fig. 3). After the major peak in δ^{18}O (~~Oi-1~~Step 2) a more stable system~~, related also to the onset of North Atlantic Deep Water (NADW) production in the early Oligocene (Via and Thomas, 2006),~~ may have allowed the proliferation of more oligotrophic taxa, including holococcoliths, and the establishment of more oligotrophic, stable environmental conditions (Fig. 6).

Previous studies ~~have~~ documented an increase in primary productivity during the late Eocene-early Oligocene, in particular in the Southern Ocean (e.g.~~,~~ Diester-Haass, 1995; Diester-Haass and Zahn, 1996; Salamy and Zachos, 1999; Persico and Villa, 2004; Schumacher and Lazarus, 2004; Anderson and Delaney, 2005). At tropical latitudes, both transient increases (equatorial Atlantic; Diester-Haass and Zachos, 2003) and decreases (e.g. Griffith et al., 2010; Moore et al., 2014) in paleoproductivity have been recorded during the early Oligocene, with a sharp drop in the export productivity ~~i~~during~~n~~ the early Oligocene at ~33.7 Ma (Moore et al., 2014), similar to what we observed in the ~~southeastern~~ SE Atlantic. Schumacher and Lazarus (2004) did not record a significant shift of paleoproductivity ~~at~~ after the EOB in equatorial oceans, but noted a decrease in the early Oligocene (after 31 Ma). An increase in seasonality ~~at~~ after the EOB, similar to the one we recorded at mid-latitudinal Site 1263, was documented at Site 689 in Southern Ocean (Schumacher and Lazarus, 2004), ~~and~~ while seasonality increased just before ~~Oi-1~~Step 2 ~~in the~~at northern high latitudes ~~as well~~ (Eldrett et al., 2009).

5.3 Timing and possible causes of the biotic response at the EOB

Marine faunal and floral ~~species~~ extinctions and community changes were coeval with the climatic deterioration during the late Eocene-early Oligocene (e.g. Adams et al., 1986; Coccioni, 1988; Berggren and Pearson, 2005; Dunkley Jones et al., 2008; Pearson et al., 2008; Tori, 2008; Villa et al., 2008, 2014). At ODP Site 1263, we ~~also~~ see close correspondence between marked changes in the nannoplankton assemblages (i.e. nannofossil abundance and coccolith size decrease) and the ~~extinction of the hantkeninid planktic~~benthic foraminiferal assemblages. ~~Both events occurred at the EOB, pre-dating the onset of Oi-1, i.e. the first major ice sheet expansion on Antarctica. Extinction events are usually rapid (10-100 kyr; Gibbs et al., 2005; Raffi et al., 2006).~~ The nannoplankton did not suffer significant extinctions at the ~~same boundary~~ EOB as the planktonic foraminiferal assemblage did, but the change in the community was as ~~relatively~~ fast as extinction events (which occur within 10-100 kyr; Gibbs et al., 2005; Raffi et al., 2006), taking place within ~~~53~~ 47 kyr.

The ~~timing of the~~ main shifts in the nannoplanktonic community ~~was~~ occurred during the EOT climatic transition, ~~after the beginning of the climate change of the EOT (i.e. probably~~ ~250 kyr after the Step 1~~relatively early~~, and ~120 kyr after the EOB~~) during the transient climate change across the EOB~~, ~~and~~ but pre-dated ~~significant~~ the major cooling and increase in Antarctic ice sheet volume (i.e. Step 2) by about ~~440~~ 200 kyr~~rs (i.e. Oi-1)~~. Therefore, ~~fossil planktonic~~nannofossil assemblages prove to be~~are fundamentally important~~ sensitive and accurate tools to investigate climate thresholds and the early impacts of climate change on biotic systems~~or crossing of threshold levels during climate change on biotic systems~~.

Benthic foraminiferal changes at Site 1263 ~~likewise~~ started before the EOB, as observed at other sites (Thomas, 1990, 2007), and the faunal turnover persisted into the early Oligocene. The benthic faunas in general show a decline in abundance of rectilinear species with complex apertures, possibly linked to the decline in nannoplankton species which they may have consumed~~which may have been used by the rectilinear benthics~~ (as e.g. hypothesized by Hayward et al., 2012, Mancin et al., 2013). The increase in phytodetritus-using species was possibly linked to more episodic upwelling and thus productivity and transport to the sea floor, and potentially blooming of more opportunistic nannoplankton species. Unfortunately, the lower resolution of the benthic foraminifer data compared to the nannofossil data does not allow to unravel the exact timing of the benthic fauna response ~~across the EOBalong~~during the EOT, and also does not allow exact correlation to changes in nannofossil assemblages.

At Site 1263 and in Southern Ocean records (Persico and Villa, 2004; Villa et al., 2008) the large reticulofenestrids declined in abundance rapidly ~~at above~~after the EOB. Persico and Villa (2004) and Villa et al. (2008, 2014) inferred a strong influence of SST cooling on coccolithophores, and the drop in SST ~~across~~ shortly after the EOB at high-latitudes is ~~also~~ confirmed by a decrease of 5°C in $U^{K'}_{37}$-based SST (Liu et al., 2009). In contrast, at Site 1263 planktonic foraminifer Mg/Ca data record no significant change in SST at that time (Peck et al., 2010; Fig. 5), as at ODP Sites 925 and 929 (tropical western Atlantic) where $U^{K'}_{37}$-based SSTs ~~also~~ show no ~~relevant~~ significant cooling (Liu et al., 2009; Fig. 5). Fairly stable SSTs were also documented in the tropics, using Mg/Ca-based SST reconstructions (Lear et al., 2008). The temperatures at mid-latitudinal Site 1263 thus may have been stable, like those in the tropics, rather than cooling, as inferred for high latitudes in the Southern Ocean (e.g. Persico and Villa, 2004; Villa et al., 2008; Liu et al., 2009; Villa et al., 2014).

If this is true, SST may not have been the main environmental factor affecting the nannoplankton assemblages at Site 1263 ~~across~~ after the EOB. Andruleit et al. (2003) documented that ~~for modern coccolithophores in tropical-subtropical regions~~ temperature changes may be of less importance for modern coccolithophores in tropical-subtropical regions, but the lower temperature at high latitudes can approach the vital limits for coccolithophores (Baumann et al., 1997), and become important as a bio-limiting factor.

Changes in the phytoplankton community could be related to a global influence of declining pCO_2. Unfortunately the estimates ~~available~~ from alkenone- and boron isotopes lack the resolution to unravel the variation ~~at~~ across and after the EOB (Fig. 5) in detail, ~~but~~ and leave open the possibility that ~~falling~~ pCO_2 falling below a certain threshold-level could have played a role in driving the reorganization in the nannoplankton community. Alternatively, our combined biotic and geochemical proxy data (i.e. nannofossil and benthic foraminiferal assemblages, and $\Delta\delta^{13}C_{P\text{-}B}$) suggest an increase in nutrient and food supply just ~~after~~ after the EOB (Fig. 6), which would have favored opportunistic taxa over low-nutrient selected, specialist species. ~~We conclude that the large reticulofenestrids were clearly at an ecological disadvantage, either due to changes in nutrient supply and/or pCO_2, whereas *Cyclicargolithus* and *Coccolithus* remained unaffected, or slightly increased in absolute abundance.~~ Most large reticulofenestrids (except *R. hillae* and *R. circus* group~~*eticulofenestra* sp.1~~) never recovered to ~~pre-EOB~~previous abundances, despite a return to more stratified conditions after ~~the Oi-1 event~~Step 2. It is unlikely that increased dissolution ~~after the Oi-1~~above 87 mcd (33 Ma) ~~event unlikely~~ explains the loss of large, heavily calcified taxa, but the decrease in size of coccoliths may have ~~may~~ also have led to enhanced remineralization of organic matter and less food supply to the benthic communities.

~~A regional increase in nutrients shortly after the EOB was also postulated to have occurred at low latitudes, based on a decrease in nannofossil species diversity at Tanzanian sites (Dunkley Jones et al., 2008). At Site 1263, no marked change in diversity was recorded at the EOB (Fig. 3). The diversity and species richness of fossil assemblages, however, are strongly affected by dissolution, or by reworking and taxonomic errors (Lazarus, 2011; Lloyd et al., 2012). The Tanzanian sites~~

~~indeed reveal remarkable and pristine marine microfossil preservation (Dunkley Jones et al., 2008; Pearson et al., 2008), rarely matched by other Eocene-Oligocene deep-sea records.~~

There appears to be a latitudinal gradient in the timing of nannofossil abundance decreases. The total abundance decreases were first detected in the Southern Ocean (late Eocene; Persico and Villa, 2004), then at mid-latitude (~~at~~ after the EOB; this study), and finally at the equator (after Step 2, ~~the Oi-1~~as inferred from a decrease in nannofossil species diversity at Tanzanian sites; Dunkley Jones et al., 2008). ~~So that~~ This observation may suggest a direct temperature effect on nannoplankton abundance since nannofossil floras reflect the pattern of cooling, which started ~~the cooling started~~ and was most pronounced at high latitudes. On the other hand,~~,~~ ~~or indirect~~ high-latitude cooling may have impact~~s~~ed ~~on~~ the global nutrient regimes and ocean circulation. Since regional dissolution bias may also have affected the comparison of absolute coccolith abundance, additional studies on well-preserved material will be necessary to confirm the timing and character of the response at different latitudes and in different ocean basins. Nevertheless, a meridional gradient in biotic response is expected, given the different environmental sensitivities and biogeographic ranges of different phytoplankton species (e.g. Wei and Wise, 1990; Monechi et al., 2000; Persico and Villa, 2004; Villa et al., 2008), and the diachroneity of the onset of cooling (Pearson et al., 2008).

6 Conclusions

High-resolution analyses of the calcareous nannofossil and foraminiferal assemblages refine the biostratigraphy at ODP Site 1263 (Walvis Ridge), and demonstrate distinct assemblage and abundance changes in marine biota ~~at~~ across the Eocene-Oligocene ~~boundary~~transition. The biotic response of calcareous nannoplankton was very rapid (~~~53~~ 47 kyr), ~~similar to the hantkenid extinction event, and~~ follow~~ed~~ing the EOB ~~of~~by ~120 kyr and pre-dat~~ed~~ing the climatic ~~Oi-1~~Step 2 event by ~~44~~200 kyr.

The ecological success of ~~the~~ smaller-sized coccolithophore species *versus* the drastic decrease of large~~ ones~~ reticulofenestrids, and the overall decrease of nannoplankton productivity ~~across~~ after the EOB likely ~~may have~~ affected the benthic foraminiferal community (e.g. decrease in rectilinear species due to changes in nannoplankton floras), with increased seasonality driving the transient increased abundance of phytodetritus-using species. After Step 2 and in particular after 33.3 Ma~~Oi-1~~, both nannoplankton and benthic records at Site 1263 were~~are~~ affected by intensified dissolution and corrosivity of bottom waters.

We conclude that the planktonic community reacted to ~~some~~ fast-changing environmental conditions, possibly seasonally increased nutrient supply to the photic zone, global cooling or lowered CO_2-availability, and/or the crossing of a threshold-level ~~along~~ in the longer-term ~~(transient)~~ climate and environmental changes suggested by available proxy data, such as the transient pCO_2 decline during the late Eocene-early Oligocene.

Supplement data file contains: Table S1 (planktonic foraminiferal marker species); Tables ~~S1~~ S2 and ~~S2~~ S3 (loading species for datasets A and B); taxonomic remarks; Fig. S1 (plate of main species); Figs. S2 and S3 (plotted curves of all the distinguished species in datasets A and B).

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References

Adams, C. G., Butterlin, J., and Samanta, B. K.: Larger foraminifera and events at the Eocene-Oligocene boundary in the Indo–West Pacific region, in: Terminal Eocene Events, edited by: Pomerol, C. and Premoli Silva, I., Elsevier, Amsterdam, 237–252, 1986.

Adler, M., Hensen, C., Wenzhöfer, F., Pfeifer, K., and Schulz, H. D.: Modelling of calcite dissolution by oxic respiration in supralysoclinal deep-sea sediments, Mar. Geol., 177, 167–189, 2001.

Agnini, C., Fornaciari, E., Rio, D., Tateo, F., Backman, J., and Giusberti, L.: Responses of calcareous nannofossil assemblages, mineralogy and geochemistry to the environmental perturbations across the Paleocene/ Eocene boundary in the Venetian Pre-Alps, Mar. Micropaleontol., 63, 19–38, 2006.

Agnini, C., Fornaciari, E., Raffi, I., Catanzariti, R., Pälike, H., Backman, J., and Rio, D.: Biozonation and biochronology of Paleogene calcareous nannofossils from low and middle latitudes, Newsletters on Stratigraphy, 47, 131–181, 2014.

Aitchison, J.: The statistical analysis of compositional data. Chapman and Hall, London, 416 pp., 1986.

Anderson, L. D. and Delaney, L. M.: Middle Eocene to early Oligocene paleoceanography from the Agulhas Ridge, Southern Ocean (Ocean Drilling Program Leg 177, Site 1090), Paleoceanography, 20, PA1013, doi:10.1029/2004PA001043, 2005.

~~Andruleit, H.: A filtration technique for quantitative studies of coccoliths, Micropaleontology, 42, 403–406, 1996.~~

Andruleit, H., Stäger, S., Rogalla, U., and Čepek, P.: Living coccolithophores in the northern Arabian Sea: ecological tolerances and environmental control. Mar. Micropaleontol., 49, 157–181, 2003.

Aubry, M.-P.: Late Paleogene calcareous nannoplankton evolution; a tale of climatic deterioration, in: Eocene-Oligocene Climatic and Biotic Evolution, edited by: Prothero, D. R. and Berggren, W. A., Princeton University Press, 272–309, 1992.

Auer, G., Piller, W. E., and Harzhauser, M.: High-resolution calcareous nannoplankton palaeoecology as a proxy for small-scale environmental changes in the Early Miocene, Mar. Micropaleontol., 111, 53–65, 2014.

Backman, J.: Quantitative calcareous nannofossil biochronology of middle Eocene through early Oligocene sediment from DSDP Sites 522 and 523, Abhandlungen der Geologischen Bundesanstalt, Vienna, 39, 21–31, 1987.

Barker, P. F. and Thomas, E.: Origin, signature and palaeoclimatic influence of the Antarctic Circumpolar Current, Earth Science Reviews, 66, 143–162, 2004.

Baumann, K.-H., Andruleit, H., Schröder-Ritzrau, A., and Samtleben, C.: Spatial and temporal dynamics of coccolithophore communities during non-production phases in the Norwegian-Greenland Sea, in: Contributions to the Micropaleontology and Paleoceanography of the Northern North Atlantic, edited by: Hass, H. C. and Kaminski, M. A., Grzybowski Foundation Special Publication, 5, 227–243, 1997.

Beaufort, L., Probert, I., and Buchet, N.: Effects of acidification and primary production on coccolith weight: Implications for carbonate transfer from the surface to the deep ocean, Geochem. Geophy. Geosy., 8, 1–18, 2007.

Benson, R. H.: The origin of the psychrosphere as recorded in changes of deep-sea ostracode assemblages, Lethaia, 8, 69–83, 1975.

Benton, M. J.: The Red Queen and the Court Jester: species diversity and the role of biotic and abiotic factors through time, Science, 323, 728–732, 2009.

Berger, W. H.: Deep-sea carbonates: evidence for a coccolith lysocline, Deep-Sea Research and Oceanographic Abstracts, 20, 917–921, 1973.

Berggren, W. A. and Pearson, P. N.: A revised tropical to subtropical Paleogene planktonic foraminifera zonation, J. Foramin. Res., 35, 279–298, 2005.

Berggren, W. A., Kent, D. V., Swisher, C. C., and Aubry, M.-P. A revised Cenozoic geochronology and chronostratigraphy, in: Geochronology, time scales and global stratigraphic correlation, SEPM Spec. Publ., 54, 129–212, 1995.

Blaj, T., Backman, J., and Raffi, I.: Late Eocene to Oligocene preservation history and biochronology of calcareous nannofossils from paleo-equatorial Pacific Ocean sediments, Riv. Ital. Paleontol. S., 115, 67–85, 2009.

Boeckel, B., Baumann, K.-H., Henrich, R., and Kinkel, H.: Coccolith distribution patterns in South Atlantic and Southern Ocean surface sediments in relation to environmental gradients, Deep-Sea Res. Pt. I, 53, 1073–1099, 2006.

Bohaty, S. M., Zachos, J. C., and Delaney, M. L.: Foraminiferal Mg/Ca evidence for Southern Ocean cooling across the Eocene/Oligocene transition, Earth Planet. Sc. Lett., 317, 251–261, 2012.

Bollmann, J., Brabec, B., Cortes, M., and Geisen, M.: Determination of absolute coccolith abundances in deep-sea sediments by spiking with microbeads and spraying (SMS method), Mar. Micropaleontol., 38, 29–38, 1999.

Bolton, C. T. and Stoll, H.: Late Miocene threshold response of marine algae to carbon dioxide limitation, Nature, 500, 558–562, 2013.

Bordiga, M., Beaufort, L., Cobianchi, M., Lupi, C., Mancin, N., Luciani, V., Pelosi, N., and Sprovieri, M.: Calcareous plankton and geochemistry from the ODP site 1209B in the NW Pacific Ocean (Shatsky Rise): new data to interpret calcite dissolution and paleoproductivity changes of the last 450 ka, Palaeogeogr. Palaeocl., 371, 93–108, 2013.

Bordiga, M., Bartol, M., and Henderiks, J.: Absolute nannofossil abundance estimates: Quantifying the pros and cons of different techniques, Revue de micropaléontologie, http://dx.doi.org/10.1016/j.revmic.2015.05.002, 2015.

Boscolo-Galazzo, F., Thomas, E., and Giusberti, L.: Benthic foraminiferal response to the Middle Eocene Climatic Optimum (MECO) in the South-Eastern Atlantic (ODP Site 1263), Palaeogeogr. Palaeocl., 417, 432–444, 2015.

Bown, P. R. and Dunkley Jones, T.: New Paleogene calcareous nannofossil taxa from coastal Tanzania: Tanzania Drilling Project Sites 11 to 14, J.~~ournal of ~~Nannoplankton Res.~~earch~~, 28, 17–34, 2006.

Bown, P. R. and Young, J. R.: Techniques, in: Calcareous Nannofossil Biostratigraphy, edited by: Bown, P. R., Chapman and Hall, Cambridge, 16–28, 1998.

Bown, P. R., Lees, J. A., and Young, J. R.: Calcareous nannoplankton evolution and diversity through time, in: Coccolithiphores, edited by: Thierstein, H., R. and Young J. R., Springer Berlin Heidelberg, 481–508, 2004.

Bown, P. R., Dunkley Jones, T., Lees, J. A., Randell, R. D., Mizzi, J. A., Pearson, P. N., Coxall, H. K., Young, J.R., Nicholas, C. J., Karega, A., Singano, J., and Wade, B. S.:A Paleogene calcareous microfossil Konservat-Lagerstätte from the Kilwa Group of coastal Tanzania, Geol. Soc. Am. Bull., 120, 3–12, 2008

Bremer, M. L. and Lohmann, G. P.: Evidence for primary control of the distribution of certain Atlantic Ocean benthonic foraminifera by degree of carbonate saturation, Deep-Sea Res., 29, 987–998, 1982.

~~Brown, R. E., Koeberl, C., Montanari, A., and Bice, D. M.: Evidence for a change in Milankovitch forcing caused by extraterrestrial events at Massignano, Italy, Eocene-Oligocene boundary GSSP, in: The Late Eocene Earth – Hothouse, Icehouse, and Impacts, edited by: Koeberl, C. and Montanari, A., Geol. S. Am. S., 452, 119–137, 2009.~~

Buccianti, A. and Esposito, P.: Insights into Late Quaternary calcareous nannoplankton assemblages under the theory of statistical analysis for compositional data, Palaeogeogr. Palaeocli., 202, 209–277, 2004.

Cachao, M. and Moita, M. T.: *Coccolithus pelagicus*, a productivity proxy related to moderate fronts off Western Iberia, Mar. Micropaleontol., 39, 131–155, 2000.

Coccioni, R.: The genera *Hantkenina* and *Cribrohantkenina* (foraminifera) in the Massignano section (Ancona, Italy), in: The Eocene–Oligocene boundary in the Marche-Umbria basin (Italy), edited by: Premoli Silva, I., Coccioni, R., and Montanari, A., International Subcommission on the Paleogene Stratigraphy, Eocene Oligocene Meeting, Ancona, Spec. Publ., 2, 81–96, 1988.

Coxall, H. K. and Pearson, P. N.: Taxonomy, biostratigraphy, and phylogeny of the Hantkeninidae (*Clavigerinella*, *Hantkenina*, and *Cribrohantkenina*), in: Atlas of Eocene Planktonic Foraminifera, edited by: Pearson, P. N., Olsson, R. K., Huber, B. T., Hemleben, C., and Berggren, W. A., Cushman Foundation Special Publication, 41, 216–256, 2006.

Coxall, H. K. and Pearson, P. N.: The Eocene-Oligocene transition, in: Deep-time perspectives on climate change: marrying the signal from computer models and biological proxies, edited by: Williams, M., et al., Geological Society (London), Micropalaeontological Society, 351–387, 2007.

Coxall, H. K. and Wilson, P. A.: Early Oligocene glaciation and productivity in the eastern equatorial Pacific: insights into global carbon cycling, Paleoceanography, 26, doi:10.1029/2010PA002021, 2011.

Coxall, H. K., Wilson, P. A., Pälike, H., Lear, C. H., and Backman, J.: Rapid stepwise onset of Antarctic glaciation and deeper calcite compensation in the Pacific Ocean, Nature, 433, 53–57, 2005.

Daniels, C. J., Sheward, R. M., and Poulton, A. J.: Biogeochemical implications of comparative growth rates of *Emiliania huxleyi* and *Coccolithus* species, Biogeosciences, 11, 6915–6925, doi:10.5194/bg-11-6915-2014, 2014.

De Kaenel, E. and Villa, G.: Oligocene-Miocene calcareous nannofossil biostratigraphy and paleoecology from the Iberia abyssal plain, in: Proceedings ODP, Scientific Results, College Station, TX (Ocean Drilling Program), 149, 79–145, 1996.

De Villiers, S.: Foraminiferal shell-weight evidence for sedimentary calcite dissolution above the lysocline. Deep-Sea Res. Pt. I, 52, 671-680, 2005.

DeConto, R. M. and Pollard, D.: Rapid Cenozoic glaciation of Antarctica induced by declining atmospheric CO_2, Nature, 421, 245–249, 2003.

Diester-Haass, L.: Middle Eocene to early Oligocene paleoceanography of the Antarctic Ocean (Maud Rise, ODP Leg 113, Site 689): change from low productivity to a high productivity ocean, Palaeogeogr. Palaeocl., 113, 311–334, 1995.

Diester-Haass, L. and Zachos, J. C.: The Eocene-Oligocene transition in the Equatorial Atlantic (ODP Site 325), paleoproductivity increase and positive $\delta^{13}C$ excursion, in: from greenhouse to icehouse: the marine Eocene-Oligocene transition, Prothero, D. R., Ivany, L. C., and Nesbitt, E. A., Columbia University Press, New York, 397–416, 2003.

Diester-Haass, L. and Zahn, R.: Eocene-Oligocene transition in the Southern Ocean: history of water mass circulation and biological productivity, Geology, 24, 163–166, 1996.

Dockery III, D. T.: Punctuated succession of marine mollusks in the northern Gulf Coastal Plain, Palaios, 1, 582–589, 1986.

Dunkley Jones, T., Bown, P. R., Pearson, P. N., Wade, B. S., Coxall, H. K., and Lear, C. H.: Major shift in calcareous phytoplankton assemblages through the Eocene-Oligocene transition of Tanzania and their implications for low-latitude primary production, Paleoceanography, 23, PA4204, doi:10.1029/2008PA001640, 2008.

Eldrett, J. S., Greenwood, D. R., Harding, I. C., and Hubber, M.: Increased seasonality through the Eocene to Oligocene transition in northern high latitudes, Nature, 459, 969–973, 2009.

Falkowski, P. G., Katz, M. E., Knoll, A. H., Quigg, A., Raven, J. A., Schofield, O., and Tayler, F. J. R.: The evolution of modern eukaryotic plankton, Science, 305, 354–360, 2004.

Fenero, R., Thomas, E., Alegret, L., and Molina, E.: Evolución paleoambiental del tránsito Eoceno-Oligoceno en el Atlántico sur (Sondeo 1263) basada en foraminíferos bentónicos, Geogaceta, 49, 3–6, 2010 (in Spanish).

Fioroni, C., Villa, G., Persico, D., and Jovane, L.: Middle Eocene-Lower Oligocene calcareous nannofossil biostratigraphy and paleoceanographic implications from Site 711 (equatorial Indian Ocean), Mar. Micropaleontol., 118, 50–62, 2015.

Foster, L. C., Schmidt, D. N., Thomas, E., Arndt, S., and Ridgwell, A.: Surviving rapid climate change in the deep sea during the Paleogene hyperthermals, Proceedings of the National Academy of Sciences, 110, 9273–9276, 2013.

Geisen, M., Bollmann, J., Herrle, J. O., Mutterlose, J., and Young, J. R.: Calibration of the random settling technique for calculation of absolute abundances of calcareous nannoplankton, Micropaleontology, 45, 437–442, 1999.

Gibbs, S. J., Shackleton, N. J., and Young, J. R.: Identification of dissolution patterns in nannofossil assemblages: a high-resolution comparison of synchronous records from Ceara Rise, ODP Leg 154, Paleoceanography, 19, PA1029, doi:10.1029/2003PA000958, 2004.

Gibbs, S. J., Young, J. R., Bralower, T. J., and Shackleton, N. J.: Nannofossil evolutionary events in the mid-Pliocene: an assessment of the degree of synchrony in the extinctions of *Reticulofenestra pseudoumbilicus* and *Sphenolithus abies*, Palaeogeogr. Palaeocl., 217, 155–172, 2005.

Gibbs, S. J., Bown, P. R., Murphy, B. H., Sluijs, A., Edgar, K. M., Pälike, H., Bolton, C. T., and Zachos, J. C.: Interactive comment on "Scaled biotic disruption during early Eocene global warming events"-, Biogeosciences Discuss., 9, C618–C620, www.biogeosciences-discuss.net/9/C618/2012/, 2012.

Giordano, M., Beardall, J., and Raven, A.: CO_2 concentrating mechanisms in algae: mechanisms, environmental modulation, and evolution, Annu. Rev. Plant. Biol., 56, 99–131, 2005.

Goldner, A., Herold, N., and Huber, M.: Antarctic glaciation caused ocean circulation changes at the Eocene–Oligocene transition, Nature, 511, 574–578, 2014.

Gooday, A. J.: Benthic foraminifera (Protista) as tools in deep-water palaeoceanography: environmental influences on faunal characteristics, Adv. Mar. Biol., 46, 1–90, 2003.

Gooday, A. J. and Jorisssen, F. J.: Benthic foraminiferal biogeography: controls on global distribution patterns in deep-water settings, Annual Reviews of Marine Science, 4, 237–262, 2012.

Gradstein, F. M., Ogg, J. G., Schmitz, M., and Ogg, G.: The Geologic Time Scale 2012, Vol. 2, Elsevier, 1144 pp., 2012.

Griffith, E., Calhoun, M., Thomas, E., Averyt, K., Erhardt, A., Bralower, T., Lyle, M., Olivarez-Lyle, A., and Paytan, A.: Export productivity and carbonate accumulation in the Pacific Basin at the transition from greenhouse to icehouse climate (Late Eocene to Early Oligocene), Paleoceanography, 25: PA3212, doi:10.1029/2010PA001932, 2010.

Hammer, Ø. and Harper, D. A. T.: Paleontological data analysis, Blackwell, Malden, USA, 2006.

Hammer, Ø., Harper, D. A. T., and Ryan, P. D.: PAST: Paleontological Statistics Software Package for education and data analysis, Palaeontologia Electronica, 4, 1–9, http://palaeo-electronica.org/2001_2001/past/issue2001_2001.htm, 2001.

Hannisdal, B., Henderiks, J., and Liow, L. H.: Long-term evolutionary and ecological responses of calcifying phytoplankton to changes in atmospheric CO_2, Glob. Change Biol., 18, 3504–3516, 2012.

Haq, B. U. and Lohmann, G. P.: Early Cenozoic calcareous nannoplankton biogeography of the Atlantic Ocean, Mar. Micropaleontol., 1, 119–194, 1976.

Hayek, L.-A. C. and Buzas, M. A.: Surveying natural populations: quantitative tools for assessing biodiversity, Columbia University Press, 590 pp., 2010.

Hayward, B. W., Kawagata, S., Sabaa, A. T., Grenfell, H. R., van Kerckhoven, L., Johnson, K., and Thomas, E.: The last global extinction (Mid-Pleistocene) of deep-sea benthic foraminifera (Chrysalogoniidae, Ellipsoidinidae, Glandulonodosariidae, Plectofrondiculariidae, Pleurostomellidae, Stilostomellidae), their Late Cretaceous-Cenozoic history and taxonomy. Cushman Foundation For Foraminiferal Research, Spec. Publ., 43, 408 pp., 2012.

Henderiks, J.: Coccolithophore size rules - reconstructing ancient cell geometry and cellular calcite quota from fossil coccoliths, Mar. Micropaleontol., 67, 143–154, 2008.

Henderiks, J. and Pagani, M.: Refining ancient carbon dioxide estimates: significance of coccolithophore cell size for alkenone-based pCO_2 records, Paleoceanography, 22, PA3202, doi:10.1029/2006PA001399, 2007.

Henderiks, J. and Pagani, M.: Coccolithophore cell size and Paleogene decline in atmospheric CO_2, Earth Planet. Sc. Lett., 269, 576–584, 2008.

Henderiks, J., Winter, A., Elbrächter, M., Feistel, R., van der Plas, A. K., Nausch, G., and Barlow, R.: Environmental controls on *Emiliania huxleyi* morphotypes in the Benguela coastal upwelling system (SE Atlantic), Mar. Ecol. Prog. Ser., 448, 51–66, 2012.

Henson, S. A., Sanders, R., and Madsen, E.: Global patterns in efficiency of particulate organic carbon export and transfer to the deep ocean, Global Biogeochem.~~ical~~ Cy.~~cles~~, 26, GB-1028, doi:10.1029/2011GB004099, 2012.

Hsü, K. J., LaBrecque, J. L., Carman Jr, M. F., and Shipboard Scientific Party: Site 522, in: DSDP, Initial Reports, College Station, TX, 73, 187–270, 1984.

Hyland, E., Murphy, B., Varela, P., Marks, K., Colwell, L., Tori, F., Monechi, S., Cleaveland, L., Brinkhuis, H., Van Mourik, C. A., Coccioni, R., Bice, D., and Montanari, A.: Integrated stratigraphic and astrochronologic calibration of the Eocene-Oligocene transition in the Monte Cagnero section (northeastern Apennines, Italy): a potential parastratotype for the Massignano global stratotype section and point (GSSP), in: The Late Eocene Earth: Hothouse, Icehouse, and Impacts, edited by: Koeberl, C. and Montanari, A., Geol. S. Am. S., 452, 303–322, 2009.

Jennions, S. M., Thomas, E., Schimdt, D. N., Lunt, D., and Ridgwell, A.: Changes in benthic ecosystems and ocean circulation in the Southeast Atlantic across Eocene Thermal Maximum 2, Paleoceanography, 30, doi:10.1002/2015PA002821, 2015.

Jorissen, F. J., de Stigter, H. C., and Widmark, J. G. V.: A conceptual model explaining benthic foraminiferal microhabitats, Mar. Micropaleontol., 26, 3–15, 1995.

~~Jennions et al., 2015 → ELLEN.~~

Jorissen, F. J., Fontanier, C., and Thomas, E.: Paleoceanographical proxies based on deep-sea benthic foraminiferal assemblage characteristics, in: Proxies in Late Cenozoic Paleoceanography: Pt. 2: Biological tracers and biomarkers, edited by: Hillaire-Marcel, C. and de Vernal, A., Elsevier, 263–326, 2007.

Katz, M. E., Miller, K. G., Wright, J. D., Wade, B. S., Browning, J. V., Cramer, B. S., and Rosenthal, Y.: Stepwise transition from the Eocene greenhouse to the Oligocene icehouse, Nat. Geosci., 1, 329–334, 2008.

Keller, G: Stepwise mass extinctions and impact events: Late Eocene to early Oligocene, Mar. Micropaleontol., 10, 267–293, 1986.

Kennett, J. P.: Cenozoic evolution of Antarctic glaciation, the circum-Antarctic Ocean, and their impact on global paleoceanography, J. Geophys. Res., 82, 3843–3860, 1977.

Koch, C. and Young, J. R.: A simple weighing and dilution technique for determining absolute abundances of coccoliths from sediment samples, Journal of Nannoplankton Research, 29, 67–69, 2007.

Kucera, M. and Malmgren, B. A.: Logratio transformation of compositional data – a resolution of the constant sum constraint, Mar. Micropaleontol., 34, 117–120, 1998.

Ladant, J.-B., Donnadieu, Y., and Dumas, C.: Links between CO$_2$, glaciation nd water flow: reconciling the Cenozoic history of the Antarctic Circumpolar Current, Clim. Past., 10, 1957-1966, 2014.

~~Lazarus, D. B.: The deep-sea microfossil record of macroevolutionary change in plankton and its study, in: Comparing geological and fossil records: implications for biodiversity studies, edited by: McGowan, A. J. and Smith, A. B., Geol. Soc., London, Spec. Publ., 358, 141–166, 2011.~~

Lear, C. H., Bailey, T. R., Pearson, P. N., Coxall, H. K., and Rosenthal, Y.: Cooling and ice growth across the Eocene-Oligocene transition, Geology, 36, 251–254, 2008.

Liu, Z., Tuo, S., Zhao, Q., Cheng, X., and Huang, W.: Deep-water earliest Oligocene Glacial Maximum (EOGM) in South Atlantic, Chinese Sci.~~ence~~ Bull.~~etin~~, 49, 2190–2197, 2004.

Liu, Z., Pagani, M., Zinniker, D., DeConto, R. M., Huber, M., Brinkhuis, H., Shah, S. R., Leckie, R. M., and Pearson, A.: Global cooling during the Eocene-Oligocene climate transition, Science, 323, 1187–1190, 2009.

~~Lloyd, G. T., Young, J. R., and Smith, A. B.: Comparative quality and fidelity of deep-sea and land-based nannofossil records, Geology, 40, 155–158, 2012.~~

Lyle, M., Wilson, P. A., Janecek, T. R., et al.: Leg 199 Summary, in: Proceedings ODP, Initial Reports, College Station, TX (Ocean Drilling Program), 199, 1–87, 2002.

MacArthur, R. H.: On the relative abundance of species, Am. Nat., 94, 25–36, 1960.

Maiorano, P., Tarantino, F., Marino, M., and De Lange, G. J.: Paleoenvironmental conditions at Core KC01B (Ionina Sea) through MIS 13-9: evidence from calcareous nannofossil assemblages, Quatern. Int., 288, 97–111, 2013.

Mancin, N., Hayward, B. H., Trattenero, I., Cobianchi, M., and Lupi, C.: Can the morphology of deep-sea benthic foraminifera reveal what caused their extinction during the mid-Pleistocene Climate Transition?, Mar. Micopaleontol., 104, 53–70, 2013.

Marino, M. and Flores, J. A.: Middle Eocene to early Oligocene calcareous nannofossil stratigraphy at Leg 177 Site 1090, Mar. Micropaleontol., 45, 291–307, 2002.

Maronna, R., Martin, R. D., and Yohai, V. J.: Robust statistics: Theory and methods, Wiley J., New York, 2006.

Martini, E.: Standard Tertiary and Quaternary calcareous nannoplankton zonation, Proc. 2nd Conf. Planktonic Microfossils, Rome, 2, 739–786, 1971.

Meng, J. and McKenna, M. C.: Faunal turnovers of Palaeogene mammals from the Mongolian Plateau, Nature, 394, 364–367, 1998.

Merico, A., Tyrrell, T., and Wilson, P. A.: Eocene/Oligocene ocean de-acidification linked to Antarctic glaciation by sea-level fall, Nature 452, 979–982, 2008.

~~Miller, K. G., Wright, J., and Fairbanks, R.: Unlocking the icehouse: Oligocene-Miocene oxygen isotopes, eustasy and margin erosion, J. Geophys. Res., 96, 6829–6848, 1991.~~

Miller, K. G., Wright, J. D., Katz, M. E., Wade, B. S., Browning, J. V., Cramer, B. S., and Rosenthal, Y.: Climate threshold at the Eocene-Oligocene transition: Antarctic ice sheet influence on ocean circulation, Geol. Soc. Am. Spec. Pap., 452, 169–178, 2009.

Milliman, J. D., Troy, P. J., Balch, W. M., Adams, A. K., Li, Y.-H., and Mackenzie, F. T.: Biologically mediated dissolution of calcium carbonate above the chemical lysocline? Deep-Sea Res. Pt. I, 46, 1653–1669, 1999.

Mix, A. C., Morey, A. E., Pisias, N. G., and Hostetler, S. W.: Foraminiferal faunal estimates of paleotemperature: circumventing the no-analog problem yields cool ice age tropics, Paleoceanography, 14, 350–359, doi:10.1029/1999PA900012, 1999.

Monechi, S., Buccianti, A., and Gardin, S.: Biotic signals from nannoflora across the iridium anomaly in the upper Eocene of the Massignano section: evidence from statistical analysis, Mar. Micropaleontol., 39, 219–237, 2000.

Moolna, A. and Rickaby, R. E. M.: Interaction of the coccolithophore *Gephyrocapsa oceanica* with its carbon environment: response to a recreated high-CO_2 geological past, Geobiology, 10, 72–81, 2012.

Moore, T. C., Rabinowitz, P. D., et al.: Site 525-529, in: Deep Sea Drilling Project, Initial Reports, US Government Printing Office, Washington, DC, USA, 74, 41–465, 1984.

Moore, T. C., Wade, B. S., Westerhold, T., Erhardt, A., M., Coxall, H. K., Baldauf, J., and Wagner, M.: Equatorial Pacific productivity changes near the Eocene-Oligocene boundary, Paleoceanography, 29, 825–844, doi:10.1002/2014PA002656, 2014.

Norris, R. D., Wilson, P. A., Blum, P., and the Expedition 342 Scientists:~~,~~ Proceedings IODP, 342, College Station, TX (Integrated Ocean Drilling Program), doi:10.2204/iodp. proc.342.2014, 2014.

Ocean Drilling Stratigraphic Network, Plate Tectonic Reconstruction Service: http://www.odsn.de/odsn/services/paleomap/paleomap.html, last access: 10 April 2015, 2011.

~~Okada, H. and Bukry, D.: Supplementary modification and introduction of code numbers to the low-latitude coccolith biostratigraphic zonation (Bukry, 1973; 1975), Mar. Micropaleontol., 5, 321–325, 1980.~~

Ortiz, S. and Thomas, E.: Deep-sea benthic foraminiferal turnover during the early middle Eocene transition at Walvis Ridge (SE Atlantic), Palaeogeogr. Palaeocl., 417, 126–136, 2015.

Formatted: No underline

Pagani, M., Huber, M., Liu, Z., Bohaty, S. M., Henderiks, J., Sijp, W., Krishnan, S., and DeConto, R. M.: The role of carbon dioxide during the onset of Antarctic glaciation, Science, 334, 1261–1264, 2011.

Pälike, H., Norris, R. D., Herrle, J. O., Wilson, P. A., Coxall, H. K., Lear, C. H., Shackleton, N. J., Tripati, A. K., and Wade, B. S.: The heartbeat of the Oligocene climate system, Science, 314, 1894–1898, 2006.

Pea, L.: Eocene-Oligocene paleoceanography of the subantarctic South Atlantic: calcareous nannofossil reconstructions of temperature, nutrient, and dissolution history, Ph.D. thesis, Department of Earth Sciences, University of Parma, Italy, 210 pp., 2010.

Pearson, K.: Mathematical contributions to the theory of evolution. On a form of spurious correlation which may arise when indices are used in the measurement of organisms, P. R. Soc. London, 60, 489–498, 1896.

Pearson, P. N., van Dogen, B. E., Nicholas, C. J., Pancost, R. D., Schouten, S., Singano, J. M., and Wade, B. S.: Stable warm tropical climate through the Eocene Epoch, Geology, 35, 211–214, 2007.

Pearson, P. N., McMillan, I. K., Wade, B. S., Dunkley Jones, T., Coxall, H. K., Bown, P. R., and Lear, C. H.: Extinction and environmental change across the Eocene-Oligocene boundary in Tanzania, Geology, 36, 179–182, 2008.

Pearson, P. N., Gavin, L. F., and Wade, B. S.: Atmospheric carbon dioxide through the Eocene–Oligocene climate transition, Nature, 461, 1110–1114, 2009.

Peck, V. L., Yu, J., Kender, S., and Riesselman, C. R.: Shifting ocean carbonate chemistry during the Eocene-Oligocene climate transition: implications for deep-ocean Mg/Ca paleothermometry, Paleoceanography, 25, doi:10.1029/2009PA001906, 2010.

Persico, D. and Villa, G.: Eocene-Oligocene calcareous nannofossils from Maud Rise and Kerguelen Plateau (Antarctica): paleoecological and paleoceanographic implications, Mar. Micropaleontol., 52, 153–179, 2004.

Peterson, L. C. and Prell, W. L.: Carbonate dissolution in recent sediments of the eastern equatorial Indian Ocean: preservation patterns and carbonate loss above the lysocline, Mar. Geol., 64, 259–290, 1985.

Plancq, J., Grossi, V., Henderiks, J., Simon, L., and Mattioli, E.: Alkenone producers during late Oligocene–early Miocene revisited, Paleoceanography, 27, PA1202, doi:10.1029/2011PA002164, 2012.

Premoli Silva, I. and Jenkins, D. G.: Decision on the Eocene-Oligocene boundary stratotype, Episodes, 16, 379–382, 1993.

Raffi, I., Backman, J., Fornaciari, E., Pälike, H., Rio, D., Lourens, L., and Hilgen, F.: A review of calcareous nannofossil astrobiochronology encompassing the past 25 million years, Quaternary Sci. Rev., 25, 3113–3137, 2006.

Riesselman, C. R., Dunbar, R. B., Mucciarone, D. A., and Kitasei, S. S.: High resolution stable isotope and carbonate variability during the early Oligocene climate transition: Walvis Ridge (ODP Site 1263), in: Antarctica: A Keystone in a Changing World-Online Proceedings of the 10th ISAES, edited by: Cooper, A. K., Raymond, C. R., and the 10th ISAES Editorial Team, US Geol. Surv., doi:10.3133/of2007-1047.srp095, 2007.

Rost, B., Riebesell, U., Burkhardt, S., and Sültemeyer, D.: Carbon acquisition of bloom-forming marine phytoplankton, Limnol. Oceanogr., 48, 55–67, 2003.

Rugenstein, M., Stocchi, P., von der Heijdt, A., Dijkstra, H., and Brinkhuis, H.: Emplacement of Antarctic ice sheet mass circumpolar ocean flow, Global Planet. Change, 118, 16–24, 2014.

Saavedra-Pellitero, M., Flores, J. A., Baumann, K.-H., and Sierro, F. J.: Coccolith distribution patterns in surface sediments of Equatorial and Southeastern Pacific Ocean, Geobios, 43, 131–149, 2010.

Salamy, K. A. and Zachos, J. C.: Latest Eocene-early Oligocene climate change and Southern Ocean fertility: inferences from sediment accumulation and stable isotope data, Palaeogeogr. Palaeocl., 145, 61–77, 1999.

Sarnthein, M. and Winn, K.: Reconstruction of low and middle latitude export productivity, 30,000 years BP to present: implication for global carbon reservoir, in: Climate-Ocean Interaction, edited by: Schlesinger, M. E., Kluwer Academic Publishers, 319–342, 1990.

Schumacher, S. and Lazarus, D.: Regional differences in pelagic productivity in the late Eocene to early Oligocene - a comparison of southern high latitudes and lower latitudes, Palaeogeogr. Palaeocl., 214, 243–263, 2004.

Sijp, W. P., von der Heydt, A. S., Dijkstra, H. A., Flögel, S., Douglas, P. J., and Bijl, P. K.: The role of ocean gateways on cooling climate on long time scales, Global Planet. Change, 119, 1–22, 2014.

Spencer-Cervato, C.: The Cenozoic deep sea microfossil record: explorations of the DSDP/ODP sample set using the Neptune Database, Palaeontol. Electron., 2, 2, 270 pp., 1999.

Thomas, E.: Late Cretaceous through Neogene deep-sea benthic foraminifers (Maud Rise, Weddell Sea, Antarctica), in: Proceedings ODP, Scientific Results, College Station, TX (Ocean Drilling Program), 113, 571–594, 1990.

Thomas, E.: Middle Eocene - late Oligocene bathyal benthic foraminifera (Weddell Sea): faunal changes and implications for ocean circulation, in: Late Eocene-Oligocene climatic and biotic evolution, edited by: Prothero, D. R., and Berggren, W. A., Princeton University Press, 245–271, 1992.

Thomas, E.: Cenozoic mass extinctions in the deep sea: what disturbs the largest habitat on Earth?, in: Large ecosystem perturbations: causes and consequences, edited by: Monechi, S., Coccioni, R., and Rampino, M., Geol. S. Am. S., 424, 1–23, 2007.

Thomas, E. and Gooday, A. J.: Cenozoic deep-sea benthic foraminifers: tracers for changes in oceanic productivity?, Geology, 24, 355–358, 1996.

Tori, F.: Variabilità climatica e ciclicità nell'intervallo Eocene Oligocene: dati dai nannofossili calcarei, Ph.D. thesis, Department of Earth Sciences, University of Florence, Italy, 222 pp., 2008 (in Italian).

~~Via, R. K. and Thomas, D. J.: Evolution of Atlantic thermohaline circulation: Early Oligocene onset of deep-water production in the North Atlantic, Geology, 34, 441–444, 2006.~~

Villa, G., Fioroni, C., Pea, L., Bohaty, S., and Persico, D.: Middle Eocene-late Oligocene climate variability: calcareous nannofossil response at Kerguelen Plateau, Site 748, Mar. Micropaleontol., 69, 173–192, 2008.

Villa, G., Fioroni, C., Persico, D., Roberts, A. P., and Florindo, F.: Middle Eocene to Late Oligoce ne Antarctic glaciation/deglaciation and Southern Ocean productivity, Paleoceanography, 29, 223–237, doi:10.1002/2013PA002518, 2014.

Wade, B. S. and Pälike, H.: Oligocene climate dynamics, Paleoceanography, 19, PA4019, doi:10.1029/2004PA001042, 2004.

Wade, B. S. and Pearson, P. N.: Planktonic foraminiferal turnover, diversity fluctuations and geochemical signals across the Eocene/Oligocene boundary in Tanzania, Mar. Micropaleontol., 68, 244–255, 2008.

Wei, W. and Wise, S. W.: Biogeographic gradients of middle Eocene–Oligocene calcareous nannoplankton in the South Atlantic Ocean, Palaeogeogr. Palaeocl., 79, 29–61, 1990.

Winter, A., Jordan, R. W., and Roth, P. H.: Biogeography of living coccolithophores in ocean waters, in: Coccolithophores, edited by: Winter, A. and Siesser, W. G., 161–177, 1994.

Young, J. R., Geisen, M., and Probert, I.: A review of selected aspects of coccolithophore biology with implications for paleodiversity estimation, Micropaleontology, 51,267–288, doi:10.2113/gsmicropal.51.4.267, 2005

Young, J. R., Bown P.R., and Lees, J. A.: Nannotax3 website, International Nannoplankton Association, 21 Apr. 2014, URL: http://http://ina.tmsoc.org/Nannotax3, last access: 21 March 2015, 2014.

Zachos, J. C. and Kump, L. R.: Carbon cycle feedbacks and the initiation of Antarctic glaciation in the earliest Oligocene, Global Planet. Change, 47, 51–66, 2005.

Zachos, J. C., Quinn, T. M., and Salamy, K. A.: High-resolution (104 years) deep-sea foraminiferal stable isotope records of the Eocene-Oligocene climate transition, Pal~~a~~eoceanography, 11, 251–266, doi:10.1029/96PA00571, 1996.

Zachos, J., Pagani, M., Sloan, L., Thomas, E., and Billups, K.: Trends, rhythms, and aberrations in global climate 65 Ma to present, Science, 292, 686–693, 2001.

Zachos, J. C., Kroon, D., Blum, P., and Shipboard Scientific Party: Site 1263, in: Proceedings ODP, Initial Reports, College Station, TX (Ocean Drilling Program), 208, 1–87, 2004.

Zhang, J., Wang, P., Li, Q., Cheng, X., Jin, H., and Zhang, S.: Western equatorial Pacific productivity and carbonate dissolution over the last 550 kyr: foraminiferal and nannofossil evidence from ODP Hole 807A, Mar. Micropaleontol., 64, 121–140, 2007.

Zhang, Y. G., Pagani, M., Liu, Z., Bohaty, S. M., and DeConto, R. M.: A 40-milion-year history of atmospheric CO_2, Philos. T. Roy. Soc. A., 371, 20130096, 2013.

Table caption

Table 1. Calcareous nannofossil and planktonic foraminiferal (underlined) bioevents as identified in this study (at meter composite depth, mcd), and the mcd reported by the Shipboard Scientific Party (Zachos et al., 2004). Note that for the planktonic foraminiferal bioevents the average depth is reported. For each bioevent, the ages available in the most recent literature are given~~, as well as the location of the reference sites~~. N.A.: not available datum; *: ages not included in the sedimentation rate estimate.

Figure captions

Figure 1. Paleogeographic reconstruction at 33 Ma (modified from Ocean Drilling Stratigraphic Network, Plate Tectonic Reconstruction Service, www.odsn.de/odsn/services/paleomap/paleomap.html) showing location of ODP Site 1263 (black dot) on Walvis Ridge. The positions of the other sites (white squares) used for comparison and cited in the text are also given.

Figure 2. Eocene-Oligocene stratigraphy of Site 1263 and DSDP Site 522 (Walv~~b~~is Ridge). Stable oxygen isotope stratigraphy ($\delta^{18}O$, ‰) DSDP Site 522 (Zachos et al., 1996) compared to that at Site

1263 (Riesselman et al., 2007).~~,~~ ~~nannofossil~~ Absolute abundances of ~~N~~nannofossil marker species ~~absolute abundances~~ (N g^{-1}; note 10^7-10^8 change in scale among curves) for dataset A (grey line) and their relative percentages (%) for datasets A (black line) and B (black dashed). A 5 pt. smoothed curve is shown for the species *C. subdistichus*~~-~~.~~,~~ ~~number of specimens > 3 chambers per gram of sediment and presence of spines of the planktonic foraminifer *Hantkenina alabamensis*.~~ Note the changes in horizontal scale~~s~~ among curves. Calcareous nannofossil and planktonic foraminiferal datums are highlighted. B: Base occurrence; T: Top occurrence; Bc: Base common occurrence.

Figure 3. Calcareous nannofossil abundance and distribution against depth (mcd) at Site 1263 (dataset A). $CaCO_3$ (wt%; Riesselman et al., 2007), coccolith dissolution index (%), H index, and the total absolute coccolith abundance (N g^{-1}). Error bars indicates the~~ and the~~ ~~mean~~ standard deviation (± 1 s.d., in %) of replicate counts~~ percentage on 5 samples are plotted against depth~~. The absolute (N g^{-1}, black ~~solid~~ line) and relative (%, ~~grey dotted~~green line) abundances of the main species ~~which~~ constituting~~e~~ the assemblage are ~~displayed~~shown. For *Cyclicargolithus* spp. and *C. pelagicus* ~~also~~ the absolute abundances of ~~the~~ different size groups are shown. The grey vertical bar ~~close to the dissolution index identifies~~marks an interval of major dissolution (87 to 83 mcd). The positions of EOB and Step 2 are reported.

Figure 4. Distribution patterns of PC1 **(a)** and PC2 **(b)** obtained from the PCA for the datasets A and B ~~(light green curves)~~. Loadings of calcareous nannofossil taxa on the two principal components of the whole studied succession for dataset A are reported. The shaded boxes represent the most relevant loaded species. Shaded area: PCs (dataset A) obtained omitting the marker species in the dataset. Red line: PCs (dataset A) obtained inserting also the marker species. The positions of EOB and Step 2 are reported.

Figure 5. ~~Coccolith total abundance (N g^{-1}),~~ PC1 and cell-size trends during the Eocene-Oligocene at Site 1263. The average cell V:SA (µm) of all placolith-bearing species (green area), *Reticulofenestra-Dictyococcites-Cyclicargolithus* (red solid line) and *Reticulofenestra-Dictyococcites* (green dotted line) are reported. The average cell V:SA of ODP 925 (black circles; Pagani et al., 2011), DSDP 516 (white triangles; Henderiks and Pagani, 2008), DSDP 511-277

(white squares) and ODP 1090 (black squares) from the southern ocean (Pagani et al., 2011), and pCO$_2$ (ppm) alkenone-based from ODP 925 (~~black~~ white circles; Pagani et al., 2011; Zhang et al., 2013), ODP 929 (~~white~~ black circles; Pagani et al., 2011), and pCO$_2$ boron isotope-based from TDP17~~2~~/12~~7~~ (grey triangles; Pearson et al., 2009) are also shown. For comparison with sea surface temperature (SST) proxies, the Mg/Ca (mmol/mol; Peck et al., 2010) at Site 1263 and the SST from $U^{k'}_{37}$ at low latitude in the Atlantic Ocean (Liu et al., 2009) are also displayed. The positions of EOB and Step 2 at Site 1263 are reported.

Figure 6. Paleoproductivity indices from nannofossil (PC2) and benthic foraminifer ($\Delta\delta^{13}C_{P-B}$ calculated from data in Riesselman et al.~~,~~ (2007) and Peck et al.~~,~~ (2010); Fisher's alpha index - diversity proxy, extinction group species, phytodetritus~~-~~ using species, buliminid species and the species *Nuttalides umbonifera*) datums are plotted against depth. The positions of EOB and Step 2 are reported.

Table 1

	This study		**Shipboard Scientific Party** (Zachos et al., 2004)	**Ages**	
Datum	**Interval (hole-core-section, cm)**	**Depth (mcd)**	**Average Depth (mcd)**	**Age (Ma)**	**References**
T *Isthmolithus recurvus*	B-3H-5, 115-116	83.19	86	32.7	Lyle et al. (2002)
T *Coccolithus formosus*	A-9H-4, 9-10	85.16	86	32.92	Pälike et al. (2006)
Bc *Sphenolithus akropodus*	A-9H-4, 100-102	86.34	N.A.	33.31 *	Pälike et al. (2006)
B *Chiasmolithus altus*	B-4H-2, 131-132	89.4	N.A.		
B *Sphenolithus akropodus*	B-4H-3, 50-52	90.09	N.A.	33.88 *	Pälike et al. (2006)
Bc *Clausicoccus subdistichus*	A-10H-4, 141-142	96.92	94.77		
T *Hantkenina* spp.	A-10H-5, 32-34/B-4H, CC	97.53	104.5	33.89	Gradstein et al. (2012)
Pseudohastigerina size reduction	A-10H-5, 32-34/B-4H, CC	97.53	N.A.	33.89	Gradstein et al. (2012)
T *Turborotalia cerroazulensis* group	A-10H-5, 32-34/B-4H, CC	97.53	N.A.		
T *Discoaster saipanensis*	B-5H-3, 50-52	102.27	104.1	34.44	Pälike et al. (2006)
T *Discoaster barbadiensis*	B-5H-4, 0-2	103.27	N.A.	34.77	Pälike et al. (2006)

B *Sphenolithus tribulosus*	B-5H-4, 50-52	103.77	N.A.	-	-

~~Table 1~~

	~~**This study**~~		~~**Shipboard Scientific Party** (Zachos et al., 2004)~~	~~**Ages**~~		
~~**Datum**~~	~~**Interval (hole-core-section, cm)**~~	~~**Depth (mcd)**~~	~~**Average Depth (mcd)**~~	~~**Age (Ma)**~~	~~**Site/Area**~~	~~**References**~~
~~T *Isthmolithus recurvus*~~	~~B-3H-5, 115-116~~	~~83.19~~	~~86~~	~~32.7~~	~~Leg 199~~	~~Lyle et al. (2002)~~
~~T *Coccolithus formosus*~~	~~A-9H-4, 9-10~~	~~85.16~~	~~86~~	~~32.92~~	~~Site 1218~~	~~Gradstein et al. (2012)~~
~~Bc *Sphenolithus akropodus*~~	~~A-9H-4, 100-102~~	~~86.34~~	~~N.A.~~			
~~B *Chiasmolithus altus*~~	~~B-4H-2, 131-132~~	~~89.4~~	~~N.A.~~	~~33.31*~~	~~Site 1218~~	~~Pälike et al. (2006)~~
~~B *Sphenolithus akropodus*~~	~~B-4H-3, 50-52~~	~~90.09~~	~~N.A.~~			
~~AB *Clausicoccus obrutus*~~	~~A-10H-4, 141-142~~	~~96~~	~~94.77~~	~~33.85*~~	~~Massignan GSSP~~	~~Brown et al. (2009)~~
~~T *Hantkenina* spp.~~	~~A-10H-5, 32-34~~	~~96.27~~	~~104.5~~	~~33.89~~	~~Mediterranean~~	~~Gradstein et al. (2012)~~
~~T *Discoaster saipanensis*~~	~~B-5H-3, 50-52~~	~~102.27~~	~~104.1~~	~~34.44~~	~~Site 1218~~	~~Gradstein et al. (2012)~~
~~T *Discoaster barbadiensis*~~	~~B-5H-4, 0-2~~	~~103.27~~	~~N.A.~~	~~34.76~~	~~Site 1218~~	~~Gradstein et al. (2012)~~
~~B *Sphenolithus tribulosus*~~	~~B-5H-4, 50-52~~	~~103.77~~	~~N.A.~~	-	-	-