

Dear Editor,

In the following, you can find a point-by-point replay to your additional comments on statistical analysis.

Dear authors,

sorry for the delay, but it took me some times to carefully check your statistical analysis. I was particularly worried by your fig 10. You mention that the PCA analysis allows to discriminate between three different faunal assemblages. You justify this result by the possibility of grouping the data points of fig 10 into three shaded areas. The problem I see is that these three shaded area have been drawn by hand, and do not seem to be delimited by statistical criteria. Using your dataset, I calculated the ellipse containing 95% nd . If you proceed like this, group 3 (PETM) is not clearly distinguishable from group 1. This similarity is sustained by further analyses (e.g., cluster) even if some of the traditional statistical tests attest their significant difference (variance test). The conclusion should then be that the faunal assemblages of group 1 and 3 are similar, although group 3 displays a greater variability. This point must be clarified. I suggest you to use discriminant analysis methods, like the LDA, or variance tests (such as MANOVA). This would really strengthen your conclusions.

In Figure 10, I described my samples as subdividable in three different floral assemblages (calcareous nannoplankton are algae) and I have never claimed these groups represent ellipses containing 95% of the data points. Anyway, this is a good point and I have added 95% ellipses for Paleocene and Eocene samples in Figure 10. As you wrote, the PETM samples are obviously widely dispersed and are not surrounded by any ellipse anymore.

What is still pretty clear from PCA is that Paleocene and post-PETM Eocene are statistical different. This is not obvious from what you can get from the literature available. In fact, statistical analyses performed (see for an instance Gibbs et al., 2006) are conceived to point out extinction and speciation rates during the event rather than changes in the relative abundance of the assemblages. Our approach is based on the fact that the changes in abundance of the major components of the assemblages are as much important as the extinction and speciation rates and give a different perspective to the variations observed in the PETM calcareous nannoplankton datasets.

Regarding the difference between group 1 and group 3, I do not totally agree when you wrote that *"the floral assemblages of group 1 and 3 are similar, although group 3 displays a greater variability"* because most of the samples representing the CORE CIE are in fact distinguishable using Component 2 in the PCA analysis. To support my view, I have followed your advice and I have applied a variance analysis (MANOVA) to my dataset. The result is that the three groups are easily distinguishable one from each other.

To address your comment, I have added an additional figure, Figure 10B, which is a scatter graph + biplot plot obtained applying the MANOVA technique. The text has been changed accordingly.

Also, it is not clear whether the "reworking" data should be included in the analysis, since you lost the stratigraphical signal.

As well explained in the available literature, during the late Paleocene and early Eocene hyperthermals, the weathering is enhanced (e.g., Zachos et al. 2006 and reference herein) and, as a consequence, reworked calcareous nannofossils are transported to the ocean and then display a remarkable increase in their relative abundance especially in marginal setting (e.g., Agnini et al. 2007; 2009). This group is thus considered a good proxy for increase weathering and used as a paleoceanographic indicator.

In addition, I fully understand the reasons of log-transforming the raw dataset. However I question the validity of the replacing values at -0.698970004 as 0 is an informative value in your study. This must be clarified. I would finally suggest you to use abundance data, despite their numerous biases, in the calculation of diversity indices of similarity between the different groups. This should help to discuss about the faunal turnover through the PETM.

With regard to your comment, I think that we have fully answered to reviewer 1 and followed her/his suggestions about how to implement the raw data in order to 1) avoid the closed-sum effect and 2) decrease the sensitivity of PCA to zero values (see reviewer 1's comment on this issue). This is essentially the reasons why we have modified our original statistical analysis and this has been done to follow your previous suggestion. Since you understand the reason why we accepted reviewer 1's suggestion, you may

also understand the importance of possibly reducing the zero value effect. These changes do not substantially modify the results obtained with raw data but I would maintain these two mat transformations because even if they decrease the values of PC1 and PC2, they guarantee for a better fit of the dataset to the statistical method applied in this study.

Regarding fig 6, what are the dashed lines representing ?

Good point. Grey and black dash lines are linear trendlines for Paleocene and Eocene samples, respectively. This has been added to the caption of Figure 6.

Finally, the abbreviations used in the data tables are not described in the text and all cells must be filled.

I'm not sure to have understood to which figures you refer to. You may refer to supplementary figures since I do not see this problem in Table 1. In this table, all the abbreviations are explained in the text or as footnotes directly on the table caption.

Table S1. There are no abbreviations.

Table S2. There are no abbreviations. Explanation of the content of each excel sheet is provided in the read me file (READ ME_Supplementary tables.docx).

Table S3. There are no abbreviations. I have added the description of the first column (Sample CIC/07) that was previously missed.

Table S4. There are no abbreviations. I have added the description of the first column (Sample CIC/07) that was previously missed.

I hope to have answered to your extra comments satisfactorily. Looking to hearing from you soon for your final decision.

Best Regards,
Claudia Agnini

1

2 **Stable isotope and calcareous nannofossil assemblage record of the late Paleocene**
3 **and early Eocene (Cicogna section)**

4

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20

21 **Abstract.** We present records of stable carbon and oxygen isotopes, CaCO₃ content, and changes in
22 calcareous nannofossil assemblages across an 81 m thick section of upper Paleocene-lower Eocene
23 marine sedimentary rocks now exposed along the Cicogna Stream in northeast Italy. The studied
24 stratigraphic section represents sediment accumulation in a bathyal hemipelagic setting from
25 approximately 57.5 to 52.2 Ma, a multi-million-year time interval characterized by perturbations in
26 the global carbon cycle and changes in calcareous nannofossil assemblages. The bulk carbonate $\delta^{13}\text{C}$
27 profile for the Cicogna section, once placed on a common time scale, resembles that at several other
28 locations across the world, and includes both a long-term drop in $\delta^{13}\text{C}$, and multiple short-term
29 carbon isotope excursions (CIEs). This precise correlation of widely separated $\delta^{13}\text{C}$ records in marine
30 sequences results from temporal changes in the carbon composition of the exogenic carbon cycle.
31 However, diagenesis has likely modified the $\delta^{13}\text{C}$ record at Cicogna, an interpretation supported by
32 variations in bulk carbonate $\delta^{18}\text{O}$, which do not conform to expectations for a primary signal. The
33 record of CaCO₃ content reflects a combination of carbonate dilution and dissolution, as also
34 inferred at other sites. Our detailed documentation and statistical analysis of calcareous nannofossil
35 assemblages show major differences before, during and after the Paleocene Eocene Thermal
36 Maximum. Other CIEs in our lower Paleogene section do not exhibit such a distinctive change;
37 instead, these events are sometimes characterized by variations restricted to a limited number of
38 taxa and transient shifts in the relative abundance of primary assemblage components. Both long-
39 lasting and short-lived modifications to calcareous nannofossil assemblages preferentially affected
40 nannoliths or holococcoliths such as *Discoaster*, *Fasciculithus*, *Rhomboaster/Tribrachiatus*,
41 *Spenolithus* and *Zygrhablithus*, which underwent distinct variations in abundance as well as
42 permanent evolutionary changes in terms of appearances and disappearances. By contrast,
43 placoliths such as *Coccolithus* and *Toweius*, which represent the main component of the
44 assemblages, were characterized by a gradual decline in abundance over time. Comparisons of

45 detailed nannofossil assemblage records at the Cicogna section and at ODP Site 1262 support the
46 idea that variations in the relative and absolute abundances, even some minor changes, were
47 globally synchronous. An obvious link is through climate forcing and carbon cycling, although the
48 linkages between variations in calcareous nanoplankton, changes in $\delta^{13}\text{C}$ records and
49 oceanography will need additional work.

50

51 **INDEX TERMS**

52 Paleocene, Eocene, calcareous nannofossils, stable isotopes, paleoclimate, Tethys

53

54 **1 INTRODUCTION**

55

56 A remarkable interval of global warming occurred from the middle Paleocene to the early Eocene,
57 between approximately 59 and 51 million years ago (Ma). This inference comes from a variety of
58 proxies (Huber and Caballero, 2011; Hollis et al., 2012), including the stable oxygen isotope ($\delta^{18}\text{O}$)
59 composition of benthic foraminifera (**Figure 1**). The precise timing of the long-term temperature
60 rise remains somewhat unconstrained, because absolute ages across the early Eocene remain
61 unsolidified. Throughout this work, we assume that the Option-1 (WO-1) time scale presented by
62 Westerhold et al. (2008) is correct (**Table 1**), but acknowledge that an offset of ca 400 kyr may occur
63 within the time interval of interest (Vandenberghe et al., 2012). Debate also surrounds the
64 magnitude and distribution of the temperature warming. Earth's surface temperatures, at least at
65 high latitudes and in the deep sea, seem to have risen by at least 6°C from ca. 59 to 51 Ma (Zachos
66 et al., 2008; Bijl et al., 2009; Huber and Caballero, 2011; Hollis et al., 2012). Indeed, the latter date
67 marks the acme of the Early Eocene Climatic Optimum (EECO), the warmest sustained time interval
68 of the Cenozoic (Zachos et al., 2008; Cramer et al., 2009; Hollis et al., 2012). Such a rise in

69 temperature is not obvious at low latitudes with current data (Pearson et al., 2007; Huber et al.,
70 2011).

71 Somehow related to long-term global warming were a series of major perturbations in the
72 global carbon cycle, as clearly indicated by stable carbon isotope ($\delta^{13}\text{C}$) records in benthic
73 foraminifera (**Figure 1**) and bulk carbonate in numerous marine sediment sequences (Shackleton,
74 1986; Corfield, 1994; Cramer et al., 2003; Zachos et al., 2008; 2010; Westerhold et al., 2011; Slotnick
75 et al., 2012). An overall increase in $\delta^{13}\text{C}$ occurred through most of the Paleocene, which climaxed in
76 a Cenozoic high at ca. 57.5 Ma (Westerhold et al., 2011), commonly referred to as the Paleocene
77 carbon isotope maximum (PCIM). From this time, $\delta^{13}\text{C}$ generally decreased to ca. 52.5 Ma. However,
78 when examined at higher temporal resolution, multiple $\delta^{13}\text{C}$ records show several short-term (<200
79 kyr) negative carbon isotope excursions (CIEs) (Cramer et al., 2003; Lourens et al., 2005; Nicolo et
80 al., 2007; Agnini et al., 2009; Zachos et al., 2010; Slotnick et al., 2012). Some of these CIEs clearly
81 coincided with rapid warming (above references). The most prominent and most widely
82 documented example of these “hyperthermals” was the Paleocene-Eocene Thermal Maximum
83 (PETM) at ca. 55.5 Ma, but other apparently similar events occurred at ca. 53.7 Ma (H1 or Eocene
84 Thermal Maximum 2, ETM-2), and at ca. 52.5 Ma (K/X, sometimes called ETM-3).

85 The early Paleogene in general, and the hyperthermals in particular, have attracted
86 considerable geoscience research. On one level, this is because these time intervals represent a
87 range of possible past analogues for understanding the effects of global warming and massive
88 carbon emissions (cf. Keeling and Whorf, 2004; Zachos et al., 2008). On another level, this is because
89 the long-term and short-term temperature and carbon cycle perturbations provide new
90 perspectives for how systems on Earth’s surface operate. The PCIM probably represents a
91 tremendous storage of ^{13}C -depleted carbon somewhere on Earth’s shallow surface (Shackleton,
92 1986; Kurtz et al., 2003; Komar et al., 2013). In turn, the CIEs probably signify rapid and large inputs

93 of ^{13}C -depleted carbon into the ocean and atmosphere (Dickens et al., 1997; Lourens et al., 2005;
94 Zeebe et al., 2009). The middle Paleocene through early Eocene shows us that Earth's climate and
95 carbon reservoirs were extremely dynamic during past times of global warmth. However, the
96 composition and whereabouts of large quantities of transferable ^{13}C -depleted carbon (e.g., seafloor
97 methane, peat, permafrost) remain uncertain (above references). Indeed, it is not clear if and how
98 the long-term and short-term carbon cycle perturbations were related to one another, or to Earth
99 surface temperatures.

100 The above context presents a series of basic questions to the geoscience community. Two of
101 these are the focus of our study: (1) What is the correct template for understanding carbon cycling
102 during the early Paleogene? Major changes in fluxes of ^{13}C -depleted carbon to the ocean or
103 atmosphere should give predictable and coherent signals in the $\delta^{13}\text{C}$ of carbon-bearing phases
104 across Earth, as well as the distribution of carbonate dissolution on the seafloor. This is not yet
105 established. For example, several recently published $\delta^{13}\text{C}$ records (Kirtland-Turner et al., 2014;
106 Slotnick et al., 2015a; Payros et al., 2015) do not precisely correlate with those at other locations
107 (Cramer et al., 2003; Zachos et al., 2010; Slotnick et al., 2012, 2015b), at least with available
108 stratigraphy. (2) How did marine calcifying organisms respond to major early Paleogene
109 perturbations in temperature and carbon cycling, both in terms of evolution and preservation? The
110 prominent changes in temperature and carbon fluxes almost assuredly caused large variations in
111 seawater pH and carbonate ion concentration (CO_3^{2-}) (Dickens et al., 1997; Zachos et al., 2005; Kump
112 et al., 2009; Zeebe et al., 2009; Leon-Rodriguez and Dickens, 2010; ; Hönisch et al., 2012; Pälike et
113 al., 2012), although the response should depend on location and carbon fluxes involved (Dickens,
114 2000; Zeebe and Westbroek, 2003; Komar et al., 2013). Such changes might also affect the ability of
115 living organisms to calcify (Riebesell et al., 2000, 2008; Kleypas et al., 2006; Iglesias-Rodriguez et al.,

116 2008; Stillman and Paganini, 2015), which might impact the fossil record (Agnini et al., 2006; Raffi
117 and De Bernardi, 2008; Erba et al., 2010; Hönisch et al., 2012).

118 In regards to both questions, calcareous nanoplankton are an obvious group of organisms to
119 focus on. This is because they are a main component of open ocean primary production (Milliman,
120 1993; Winter et al., 1994; Rost and Riebesell, 2004), because they are the dominate the output of
121 carbonate in the ocean (Ziveri et al., 1999; Hay, 2004), and because they exhibit marked changes in
122 species composition from the middle Paleocene through the early Eocene (Romein, 1979; Aubry,
123 1998, Bown et al., 2004; Gibbs et al., 2012). While numerous studies have examined calcareous
124 nanofossils across the PETM from different perspectives (e.g., Bralower, 2002; Stoll and Bains,
125 2003; Gibbs et al., 2006a; 2006b; Agnini et al., 2007a; Mutterlose et al. 2007; Bown and Pearson,
126 2009; Jiang and Wise, 2009, Self-Trail et al., 2012), the relationship between these organisms and
127 carbon cycle perturbations before and after this short-lived warming episode remain poorly
128 documented (Gibbs et al., 2012). It seems possible that the high rate of calcareous nanofossil
129 taxonomic evolution (appearances and extinctions), as well as distinct changes in calcareous
130 nanofossil abundance patterns may provide excellent stratigraphic control across the early
131 Paleogene (Bukry, 1973; Perch-Nielsen, 1985; Backman, 1986, Agnini et al., 2014). Moreover, if the
132 exact relationship between changes in nanofossil assemblages and global carbon cycle
133 perturbations were known, key time intervals could be rapidly identified for more detailed work.
134 Finally, changes in calcareous nanofossils across the early Paleogene provide insights about the
135 response of an important part of the sediment forming marine biota to changes in climate and
136 carbon cycling.

137 Very few stratigraphic sections presently have detailed and coupled records of stable isotopes,
138 carbonate content, and calcareous nanofossil abundances across the broad late Paleocene-early
139 Eocene interval. The two notable exeptions are Ocean Drilling Program (ODP) Site 1262 (southeast

140 Atlantic) (Agnini et al., 2007b; Zachos et al., 2010) and Deep Sea Drilling Project (DSDP) Site 577
141 (northwest Pacific) (Shackleton, 1986; Dickens and Backman, 2013) (**Figure 2**). Here we present
142 geochemical records ($\delta^{13}\text{C}$, $\delta^{18}\text{O}$ and CaCO_3 content) and calcareous nannofossil census data from
143 the Cicogna section in northeast Italy (**Figures 2, 3**). These data are compared with similar
144 information from Sites 1262 and 577. We show that the Cicogna section provides an important
145 template for understanding potential relationships between climate, carbon cycling and the biotic
146 evolution of calcareous nanoplankton.

147

148 **2 THE CICOGNA SECTION**

149

150 The Cicogna section crops out along the Cicogna Stream near the village of Tassei in the Belluno
151 Province, northeast Italy (**Figure 3**). From a regional geological perspective, the sedimentary rocks
152 of this section belong to the Belluno Basin. This basin represents part of a paleogeographic domain
153 that formed when Jurassic rifting created a series of N–S oriented structural highs (platforms) and
154 lows (basins), which persisted through much of the Paleogene (Bernoulli and Jenkyns, 1974;
155 Bernoulli et al., 1979; Winterer and Bosellini, 1981). Importantly, from the Cretaceous to the middle-
156 late Eocene, expanded deep sea sediment successions accumulated within the basins at nominally
157 30°N latitude (Stefani and Grandesso, 1991; Agnini et al., 2006; 2011; Zattin et al., 2006).

158 The Cicogna section consists of two lithostratigraphic units (**Figure 3**). The lower portion is a
159 well-exposed upper Paleocene and lower Eocene unit referred to as Scaglia Rossa *sensu lato* (**Figures**
160 **3, 4**) (Giusberti et al., 2007; Dallanave et al., 2009). Based on benthic foraminiferal assemblages, the
161 early Paleogene marls of this unit represent lithified pelagic and hemipelagic sediment that
162 accumulated at middle to lower bathyal water depth, likely between 600 m and 1000 m and not
163 deeper than 1500 m (Giusberti et al., 2007; 2015). The upper portion is a thick early to middle

164 Eocene unit called the Belluno Flysch (**Figures 3, 4**). This unit represents a synorogenic deposit on
165 the flanks of the former Trento and Friuli platforms (Grandesso, 1976; Doglioni and Bosellini, 1987).

166 Once corrected for bed strike and dip (ca. 315°N; ca. 45°) and bends in the stream, the Scaglia
167 Rossa at Cicogna measures 80 m in terms of stratigraphic height (Dallanave et al., 2009). The Belluno
168 Flysch measures 1 m in the Cicogna section. Furthermore, the section of interest can be subdivided
169 into several subunits (**Figure 4**). The lower 20 m is comprised of distinctive alternating beds of gray-
170 greenish to purple marls and calcareous marls, the latter defined by carbonate contents higher than
171 60% (**Figure 3c**). This is overlain by approximately 9 m of pink-red marls with much less lithologic
172 alternation. At 28.7 m, the sedimentary package is broken sharply by an approximately 3 m thick
173 red to brownish-red interval of clayey marls with sporadic grey-green cm-scale spots and lenses
174 (**Figure 3f, g**). This has been called the Clay Marl Unit (CMU), and records the core of the prominent
175 negative $\delta^{13}\text{C}$ excursion associated with the PETM at multiple outcrop sites within the Belluno Basin
176 (Agnini et al., 2006; 2007a; Giusberti et al., 2007). Above the CMU, from 31.7 to 39.2 m, the section
177 continues with deposition of rhythmic alternations of marls and calcareous marls (**Figure 3g**). Above
178 this 8.5 m thick interval, at ca. 40.5 m, spatic calcite crystals occur. Generally, couplets of marl and
179 calcareous marl couplets become less evident from 40.5 m until 54 m, where such couplets reoccur
180 (**Figure 3d**). At 75.5 m, a thin calcarenitic bed is encountered, presaging the onset of the Belluno
181 Flysch. This turbidite is followed by a temporary return to hemipelagic sedimentation that ends at
182 80.6 m. Above, sedimentation of the Belluno Flysch begins in earnest (**Figures 3b, 4**).

183 The basic stratigraphy of the Scaglia Rossa in the Cicogna section, including both polarity chron
184 boundaries and key calcareous nannofossil biohorizons has been published (Giusberti et al., 2007;
185 Dallanave et al., 2009). The combined biomagnetostratigraphy indicates that the 81 m of interest
186 spans polarity Chron C25r to Chron 23r, and calcareous nannofossil biozones CP6 to CP10 (Okada
187 and Bukry, 1980) or CNP10 to CNE4 (Agnini et al., 2014). Thus, the section represents a 5.3 million

188 year (Myr) long time interval, from about 57.5 to 52.2 Ma on the W0-1 time scale. This also implies
189 an average sedimentation rate (SR) of ca. 15 m/myr. Although the deposition of hemipelagic
190 sediment might suggest relatively constant SRs over time, the PETM and possibly other
191 hyperthermal events in the Belluno Basin were characterized by higher SRs (Giusberti et al. 2007;
192 Agnini et al., 2009; Tipple et al., 2011; Krishnan et al., 2015).

193 The Scaglia Rossa at Cicogna appears to record fairly continuous sediment accumulation at
194 moderately high deposition rates. This is important because it affords longer time duration than
195 most shallow ocean sites, greater time resolution than most deep ocean sites (**Figure 2**), and an
196 overall different environmental setting. Many early Paleogene records, especially those from paleo-
197 shelf environments, such as in Egypt (e.g. Aubry and Salem, 2012) and New Jersey (Mixon and
198 Powars, 1994; Harris et al., 2010), or from early deep sea drilling expeditions, such as in the Indian
199 Ocean (Slotnick et al., 2015b), are discontinuous, either because of hiatuses or core gaps. Much of
200 the detailed work and current understanding of stable carbon isotope stratigraphy and calcareous
201 nannofossil variations across the broad early Paleogene, therefore, has come from deep-sea drilling
202 sites with multiple holes but slow sedimentation rates, although we note the work in Clarence
203 Valley, New Zealand (**Figure 2**), another area that contains several paleo-slope sections with
204 moderately high sedimentation rates (Nicolo et al., 2007; Slotnick et al., 2012, 2015b; Dallanave et
205 al., 2015). For the Cicogna section, detailed stable isotope and CaCO₃ records are currently lacking,
206 as well as detailed calcareous nannofossil assemblage information, which we present here.

207

208 **3 MATERIAL AND METHODS**

209

210 **3.1 Samples**

211

212 A total of 492 samples were chiseled from outcrops along the section. Samples were selected so as
213 to be as fresh and unaltered as possible. This included chipping off weathered surfaces while in the
214 field. Each sample was calibrated to height (**Figure 4**). Samples then were split, with one portion
215 powdered in an agate ball mill, and subsequently freeze-dried.

216

217 **3.2 Geochemistry**

218

219 Each powdered samples was analyzed for bulk sediment stable isotope composition at the Stable
220 Isotope Laboratory, University of Southampton, UK. A known mass (~80 µg) was placed into a
221 headspace vial, dried overnight, and flushed with helium. 10 mL of 100 % phosphoric acid was added
222 to each sample and allowed to react. The liberated CO₂ gas was measured using an EUROPA
223 Scientific GEO 20-20 mass spectrometer fitted with a microCAPS for carbonate analysis. Results are
224 reported in standard delta notation relative to Vienna Pee Dee Belemnite (VPDB). An in-house
225 standard of Carrara Marble, calibrated to NBS-19 Limestone, was measured multiple times to
226 evaluate accuracy and precision. The external analytical precision (1σ), based on these replicate
227 analyses, was 0.028 ‰ for δ¹³C and 0.057 ‰ for δ¹⁸O.

228 The amount of CaCO₃ in each sample was calculated from the beam height response during
229 isotope mass spectrometer measurements (Spofforth et al., 2010). The liberated CO₂ gas, when
230 squeezed up in the bellows, is measured and generates a current, the beam height. The pressure of
231 CO₂ gas is directly proportional to the beam height and therefore the mass of carbonate in the
232 sample. Over 100 samples of pure CaCO₃, with masses between 200 and 480 µg, were analyzed to
233 establish a linear relationship between beam height and carbonate content ($\text{CaCO}_3 = mx + b$; $R^2 =$
234 $0.94 - 0.99$). Results were validated by analyzing 30 samples on a C-H-N-O elemental analyzer.

235

236 3.3 Calcareous nannofossils

237

238 The un-powdered sample split was examined for calcareous nannofossils. Raw sediments were
239 processed to prepare standard smear slides (Bown and Young, 1998). To assess the reproducibility
240 of our counting methods in every single sample, a pivotal sample was prepared 10 times by two
241 different operators. Repeated counts of the identical sample performed by different analysts gave
242 similar results (sd <2-5 %). Particle density estimates (Baccelle and Bosellini, 1965) were not carried
243 out because samples have a high range in the terrigenous content (22 to 90 %). An increase or
244 decrease of the silicoclastic component is mainly related to the major or minor efficiency of the
245 chemical and mechanical weathering on land (Agnini et al., 2009). In the studied sediments, the
246 variation in the amount of the terrigenous content through time has modified the density of the
247 allochemic particle component. Consequently, calcareous nannofossil absolute abundances could
248 not be estimated correctly using a homogeneous/constant particle density or by weighing the same
249 amount of sediment for each smear slide. However, the scope of semi-quantitative counts
250 performed in this study is to recognize the precise position of biostratigraphic biohorizons rather
251 than use these data as a proxy of the paleoproductivity of taxa. Essentially, the identification of the
252 appearance or disappearance of any given taxon is not affected by its stratigraphic abundance
253 pattern, which obviously reduces the negative effect of the variable abundance of the silicoclastic
254 component throughout the section. Samples were examined under a Zeiss light microscope at
255 1250× magnification. Calcareous nannofossils were determined using taxonomy proposed by Aubry
256 (1984, 1988, 1989, 1990, 1999), Perch-Nielsen (1985) and Bown (2005).

257 A total of 200 samples were examined, providing an average time resolution of ca. 25 kyr. A
258 preliminary qualitative estimate of the abundance and preservation state of calcareous nannofossil
259 assemblages was performed for all samples. An initial large batch (185) was analyzed primarily to

260 provide biostratigraphic control for the Cicogna section, and the basic results have been presented
261 by Dallanave et al. (2009). We re-checked and refined the positions of some biohorizons by
262 examining 15 additional samples, primarily across some of the CIEs, such as B1/B2, PETM, H1 and
263 H2, and K/X (Cramer et al., 2003). The calcareous nannofossil biostratigraphic schemes used by
264 Dallanave et al. (2009) were those of Martini (1971) and Okada and Bukry (1980). The new zonal
265 scheme of Agnini et al. (2014) is also used here. Biohorizon nomenclature follows that given by
266 Agnini et al. (2014): Base (B), Base common (Bc), Top (T) and Top common (Tc).

267 Calcareous nannofossil biostratigraphic results are based on semi-quantitative analyses, which
268 is based on counts of the number of specimens of selected taxa present in a prefixed area, 1 mm²
269 or 3 long traverses (modified after Backman and Shackleton, 1983). Calcareous nannofossil
270 paleoecological results are instead based on relative abundances of calcareous nannofossil taxa
271 (percent of the total assemblage), calculated from counts of at least 300 specimens.

272 To capture changes in calcareous nannofossil assemblages we also use a statistical approach.
273 Principal Component Analysis (PCA) was preferred to other methods, as for instance non-metric
274 dimensional scaling (MDS) procedure for which a small number of axes are chosen prior to the
275 analysis and the data are fitted to these dimensions (Hammer et al., 2001). However, non-metric
276 MDS results were performed and are available as supplementary data (Figure S2). Multivariate
277 ANalysis Of Variance (MANOVA) was carried out on our dataset to determine if significant
278 differences are present among the three groups of samples recognized with PCA analysis.

279 Principal component analysis (PCA) and MANOVA was/were performed on the percentages of 15
280 subgroups using the statistical software package, PAST ver. 2.17c (Hammer et al., 2001). Such-The
281 former analysis is often used for examining paleontological data (e.g., Buccianti et al., 2006; Kucera
282 and Malmgren, 1998; Watkins and Self-Trail, 1992; Thibault and Gardin, 2010; Marino et al., 2012;
283 Bordiga et al., 2015), as it can point out hypothetical variables (components) that explain much of

284 the variance in a multidimensional data set. The first principal component accounts for the most
285 variability in any dataset examined. Each succeeding component has the highest variance possible
286 relative to the preceding components (Hammer et al., 2001). This method also increases the
287 symmetry, homoscedasticity and linearity of the data set (Aitchison, 1986). The chosen subgroups
288 were: *Chiasmolithus*, *Coccolithus*, *Ellipsolithus*, *Discoaster*, *Ericsonia*, *Fasciculithus*, *Girgisia*,
289 *Octolithus*, *Prinsius*, *Sphenolithus*, *Toweius*, *Rhombaster/Tribrachiatus*, *Zyghrablithus*, reworked
290 forms, and “others”.

291 **4 RESULTS**

292

293 **4.1 Carbon isotopes**

294

295 The bulk rock $\delta^{13}\text{C}$ record for the Cicogna section can be described, in a general sense, as a long-
296 term decrease of approximately 3 ‰, punctuated by a series of negative CIEs (**Figure 4**). The most
297 prominent low in $\delta^{13}\text{C}$ coincides with the CMU.

298 Previously established polarity chron boundaries and key calcareous nannofossil biohorizons at
299 the Cicogna section (Dallanave et al., 2009) provide very good stratigraphic framework. Once placed
300 onto a common time scale, in this case WO-1 (Westerhold et al., 2008), the $\delta^{13}\text{C}$ record at Cicogna
301 is fairly similar to those generated using upper Paleocene and lower Eocene marine carbonate at
302 other locations (Cramer et al., 2003; Zachos et al., 2010; Slotnick et al., 2012). This includes, for
303 example, bulk carbonate $\delta^{13}\text{C}$ records at ODP Site 1262, and DSDP Site 577 (**Figure 5**) The relatively
304 high $\delta^{13}\text{C}$ values near the base of the Cicogna section document the late stages of the PCIM, which
305 was centered within C25r (**Figure 1**). The overall drop in $\delta^{13}\text{C}$ across the section marks the long-term
306 global decrease in $\delta^{13}\text{C}$ that lasted through Chron C24n (**Figure 1**). The record contains multiple
307 negative shifts in $\delta^{13}\text{C}$. There is, however, an intriguing difference: across the Cicogna section, the

308 long-term 3 ‰ shift in bulk carbonate $\delta^{13}\text{C}$ values is generally offset from that in bulk carbonate
309 $\delta^{13}\text{C}$ records at Sites 1262 and 577 by approximately -1 ‰.

310 The superimposed CIEs are considered to correspond to CIEs found in $\delta^{13}\text{C}$ records from
311 elsewhere, some of which represent known or inferred hyperthermal events (Cramer et al., 2003;
312 Lourens et al., 2005; Nicolo et al., 2007; Zachos et al., 2010; Slotnick et al., 2012). There are three
313 pairs of CIEs below the CMU (**Figure 4**), and during the initial upper Paleocene long-term decline in
314 $\delta^{13}\text{C}$. These correspond with the B1/B2, C1/C2 and D1/D2 CIEs documented by others (Cramer et
315 al., 2003; Zachos et al., 2010). Notably, at Site 1262, the B1/B2 CIEs occur during middle C25n, and
316 the C1/C2 CIEs occur at the start of C24r (**Figure 5**). The same is true at Cicogna. Interestingly, at
317 Cicogna, the B2 and C2 CIEs show greater magnitudes than the B1 and C1 CIEs, and these paired
318 excursions are more pronounced than at all other locations examined to date. An additional paired
319 CIE occurs in the uppermost Paleocene (**Figure 4**). This may correlate to a fourth set of late
320 Paleocene CIEs documented at Site 1262 (Zachos et al., 2010).

321 The lower Eocene portion of the $\delta^{13}\text{C}$ record at Cicogna (**Figure 4**) begins at the CMU, which
322 marks the PETM (Giusberti et al., 2007; Dallanave et al., 2009). As at many locations, the PETM is
323 characterized by a prominent negative CIE. The shift in $\delta^{13}\text{C}$ at Cicogna is approximately -2.5 ‰, a
324 decrease that begins abruptly at 28.7 m and returns more gradually to near pre-excursion values by
325 about 33 m. From approximately 33 to 54 m, the $\delta^{13}\text{C}$ curve shows a relatively smooth trend. At 54
326 m, a pair of CIEs begin, with the first pair having a magnitude of about 1.0 ‰. These are the H1/H2
327 events (Cramer et al., 2003), which occurred in the upper part of Chron C24r (Lourens et al, 2005;
328 Zachos et al., 2010; Dickens and Backman, 2013; Dallanave et al., 2015). Above the H1/H2 CIEs, and
329 within Chron C24n, are a series of smaller (0.4 to 0.6 ‰) CIEs. Those at approximately 60, 65 and 72
330 m, are correlated with the I1/I2, J and K/X events, respectively. In summary, the $\delta^{13}\text{C}$ record at
331 Cicogna correlates with that at ODP Site 1262 (Zachos et al., 2010) and DSDP Site 577 (Dickens and

332 Backman, 2013) (**Figure 5**), as well as at several other locations (Cramer et al., 2003; Slotnick et al.,
333 2012; 2015b). This is important because it enables comparison and discussion between widely
334 separated sedimentary records within a firm temporal framework.

335

336 **4.2 Oxygen isotopes**

337

338 The $\delta^{18}\text{O}$ values range from -1.08 to -3.64 ‰ with a mean value of -1.96 ‰ and a standard deviation
339 (1σ) of 0.50 ‰ (**Figure 4**). However, at the broad scale, $\delta^{18}\text{O}$ increases upsection, with Paleocene
340 samples averaging -2.10 ‰ and Eocene samples averaging -1.89 ‰. This trend is noteworthy
341 because $\delta^{18}\text{O}$ values should decrease upsection, if the composition of the CaCO_3 was principally
342 reflecting rising global temperatures through the early Eocene. The 1σ of $\delta^{18}\text{O}$ values also increases
343 upsection, being 0.33 ‰ across Paleocene samples and 0.56 ‰ across Eocene samples.

344 There is virtually no correlation ($r^2 = 0.014$; $r=0.12$) between $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ values across all
345 samples (**Figure 6**). However, most “short-term” CIEs do display decreases in $\delta^{18}\text{O}$ (**Figure 4**). An
346 interval of anomalously low $\delta^{18}\text{O}$ values occurs from 39.9 m to 40.9 m, where the spatic calcite was
347 observed.

348

349 **4.3 Carbonate content**

350

351 The CaCO_3 content varies between 9.4 and 77.7 % across the sample suite, with a mean value of
352 54.3 % and a 1σ of 8.2 % (**Figure 4**). Two important findings emerge from the CaCO_3 content record.
353 First, from 39 m to 54 m, where we find limited variance in the $\delta^{13}\text{C}$ curve, CaCO_3 content averages
354 52.1 % with a 1σ of 4.9 %. Thus, while the average is similar to that calculated for the entire section,
355 the standard deviation is much less. At Site 1262, the corresponding time interval is also

356 characterized by limited variance in $\delta^{13}\text{C}$ values and carbonate contents, the latter inferred from the
357 abundance of Fe counts in XRF scans (Zachos et al., 2010). Second, across all samples, the CaCO_3
358 content co-varies somewhat ($r=0.29$) with $\delta^{13}\text{C}$ (**Figure 6**). This is because several lows in CaCO_3
359 content coincide with minima in $\delta^{13}\text{C}$, as is obvious for the B1/B2, PETM and H1/H2 events (**Figure**
360 **4**).

361

362 **4.4 Calcareous nannofossils**

363

364 Calcareous nannofossils are generally abundant, diverse, and moderately well preserved. The sole
365 exception is across a 10 cm interval from 28.75 to 28.85 m, which corresponds to the onset of the
366 CIE that marks the PETM. The three samples from this interval are virtually barren of calcareous
367 nannofossils.

368 Secondary overgrowth of calcite can partially or wholly blur species-specific morphological
369 features. Such diagenetic alteration, however, only marginally influences the relative, semi-
370 quantitative and absolute abundance of calcareous nannofossil taxa (Toffanin et al., 2013). Calcite
371 dissolution, on the other hand, can significantly affect the relative abundances of various calcareous
372 nannofossils within a given volume of sediment. This is because the removal of more dissolution
373 susceptible taxa, such as *Toweius* and holococcoliths, necessarily increases the abundance of less
374 dissolution susceptible taxa, such as discoasters (Roth and Thierstein, 1972; Adelseck et al., 1973;
375 Roth, 1983; Bornemann and Mutterlose, 2008; Toffanin et al., 2013). In general, moderate to strong
376 calcite dissolution also decreases the total abundance of calcareous nannofossils within a given
377 volume of sediment (Adelseck et al., 1973; Toffanin et al., 2011). In the Cicogna section, calcite
378 overgrowth on discoasters is the prevalent process affecting calcareous nannofossil assemblages
379 (**Plate I**). Most assemblages display high abundances (>1000 specimens/ mm^2) and a high diversity,

380 which include more fragile taxa. It follows that dissolution has not severely altered most
381 assemblages in samples from the Cicogna section. Rather, the calcareous nannofossil record is
382 considered to represent a genuine paleoecological signal.

383 Nannofossil assemblages from the Cicogna section display several general trends (**Figures 7-9**).
384 At the most basic level, there is a decrease in the total number of nannofossils (N/mm^2) with
385 decreasing age. Paleocene samples average approximately 2600 specimens/ mm^2 , whereas Eocene
386 samples above the H1/H2 events average approximately 1200 specimens/ mm^2 . This decrease in
387 abundance broadly corresponds to a change in calcareous nannofossil composition, as supported
388 through a series of additional observations at the Cicogna section (**Figures 7-9**):

- 389 • *Coccolithus* and *Toweius* constitute nearly half of the assemblages considering the entire
390 section. However, these genera show a clear decrease in abundance upsection, with a mean
391 value of 60 % in Paleocene samples and 35 % in Eocene samples;
- 392 • *Zyghrablithus bijugatus* shows a low mean value of approximately 4 % in the Paleocene,
393 followed by a sharp increase in the basal part of the Eocene, and a mean value of
394 approximately 25 % upsection in the Eocene. Hence, the abundance of this taxon expands
395 on behalf of *Coccolithus* and *Toweius*;
- 396 • *Sphenolithus* decreases progressively during the Paleocene, suddenly disappears at the
397 onset of the PETM, before returning to and exceeding pre-PETM values in the lower Eocene.
398 Thus, the abundance of sphenoliths also expands on behalf of *Coccolithus* and *Toweius*;
- 399 • *Fasciculithus* shows a severe decline in abundance and species diversity at the onset of the
400 PETM (28.70 m), leading up to their extinction at 34.73 m;
- 401 • *Octolithus* is rare throughout most of the studied section, but displays high abundances from
402 approximately 14.7 m to 27.5 m in the upper Paleocene;

- 403 • *Discoaster* does not show any distinct change in abundance except for a single peak at the
404 onset of the PETM;
- 405 • Several Cretaceous and early Paleocene species constitute minor reworked components
406 throughout the section. Notably, the intervals marked by the PETM, H1/H2 and, to a lesser
407 extent, B1/B2 CIEs are characterized by higher abundances of these reworked components;
- 408 • Representatives of placolith genera, such as *Prinsius*, *Ericsonia*, *Chiasmolithus* and *Girgisia*,
409 are minor components of most samples. *Prinsius* displays a marked permanent decrease in
410 abundance from a mean value of approximately 6 % to 2.5 % across the Paleocene/Eocene
411 boundary. By contrast, *Ericsonia* does not show a prominent difference in abundance
412 between Paleocene and Eocene assemblages, but increases in abundance during known and
413 suspected hyperthermal events;
- 414 • The Calcareous Nannofossil Excursion Taxa (CNET), which include *Discoaster araneus* and
415 the genus *Rhomboaster* are present during the CIE of the PETM. The evolution of the
416 *Rhomboaster/Tribrachiatus* plexus started at the onset of the PETM, when *Rhomboaster* and
417 *T. bramlettei* first appeared, and continued into the lower Eocene with the successive
418 appearances of *T. contortus* and *T. orthostylus* (Raffi et al., 2005; Agnini et al., 2006; 2007b).

419 Beyond the above variations, evolutionary appearances and extinctions occur during the
420 studied time interval (**Figures 7-9**). Most of these species belong to *Discoaster*, *Sphenolithus* and the
421 *Rhomboaster/Tribrachiatus* lineage, and include *D. multiradiatus*, *D. diastypus*, *D. lodoensis*, *S.*
422 *radians*, *S. anarrhopus*, *T. bramlettei*, *T. contortus* and *T. orthostylus*. The biohorizons defined using
423 these species are exceptionally useful for biostratigraphy and, interestingly, often occur close to
424 changes in $\delta^{13}\text{C}$.

425 All assemblage data were used for PCA analysis. This indicates that PC1 (41.3 %) and PC2 (14.7
426 %) together account for 56 % of the variance in the dataset. The PCA graph (**Figure 10A8**; **Figure S1**

427 **supporting material**) shows that samples can be ~~easily~~ subdivided into three subgroups. The first
428 two populations of samples are distinguished because of their different positions along the x-axis
429 (PC1). The third population is much more dispersed but a possible discrimination can be separated
430 from the other two seems to be because hypothesized because of its different position along the y-
431 axis (PC2). The use of a different statistical procedure, as for instance MDS, does not substantially
432 change these results (**Figure S2**). To further support the subdivision of the study samples in three
433 subgroup, we applied the MANOVA analysis to our data set (Figure 10B). The result clearly
434 confirmed that that Paleocene, PETM and Eocene samples are in fact isolated one from each other.
435

436 5 DISCUSSION

437

438 5.1 Integrated stratigraphy and a carbon isotope template

439

440 Polarity chron boundaries and calcareous nannofossil biohorizons (**Table 1; Figure 4**) provide a solid
441 stratigraphic framework for the Cicogna section. Calcareous nannofossil biohorizons, including
442 additional ones defined here, align in same stratigraphic order when compared to other locations,
443 such as ODP Site 1262 and DSDP Site 577 (**Table 1; Figure 11**). The Cicogna section represents
444 sediment accumulation between 57.5 and 52.2 Ma on the WO-1 time scale (Dallanave et al., 2009).
445 The average SR was ca.15.2 m/Myr, although this must have varied (**Figures 3, 11**). The CMU, which
446 marks the “core” of the PETM and ca. 80-100 kyr, showing a higher sedimentation rate than much
447 of the remaining record (Dallanave et al., 2009; Krishnan et al., 2015).

448 Once placed into the above stratigraphic framework, the bulk carbonate $\delta^{13}\text{C}$ profile
449 documented at Cicogna correlates well to that generated at ODP Site 1262 (**Figure 5**). In fact, it is
450 similar to $\delta^{13}\text{C}$ profiles generated at multiple locations (**Figure 2, Figure S4**), as long as records have

451 been properly calibrated in both the depth and time domains. This includes accounting for core
452 stretching and core gaps at scientific drilling sites, such as at DSDP Site 577 (Dickens and Backman,
453 2013), and accounting for changing strike and dip along land sections, such as done at Cicogna
454 (**Figure 3**). During late Paleocene and early Eocene times, the Cicogna section records the long-term
455 decrease in $\delta^{13}\text{C}$. Superimposed on this drop were multiple, often paired, negative CIEs. The PETM
456 definitively represents the most prominent CIE, but several other CIEs occurred before and after.
457 Importantly, the relative positions of polarity chron boundaries, key calcareous nannofossil
458 biohorizons and CIEs at Cicogna align well with those found at other locations (**Table 1; Figures 5,**
459 **11**).

460 A clearly recognizable $\delta^{13}\text{C}$ pattern spans the late Paleocene through the early Eocene at several
461 locations (Cramer et al., 2003; Nicolo et al., 2007; Galeotti et al., 2010; Zachos et al., 2010; Slotnick
462 et al., 2012, 2015b), although the total number of CIEs remains uncertain. At Cicogna, the problem
463 lies in the interval surrounding the K/X event, which broadly corresponds to the start of the EECO
464 (see discussion in Slotnick et al., 2012). We cannot confirm with our sample resolution whether a
465 series of short-term, small amplitude CIEs mark this time, an idea suggested from $\delta^{13}\text{C}$ records of
466 the Clarence Valley sections (Slotnick et al., 2012; 2015b). However, as at other locations, such as
467 Site 1262, no significant CIEs occurred within the 1.6 Myrs between the PETM and the H-1/ETM-2
468 event (**Figure 5**).

469 The time-correlative $\delta^{13}\text{C}$ template implies changes in the mean ocean $\delta^{13}\text{C}$ of dissolved
470 inorganic carbon (DIC). In turn, these compositional changes very likely represent variations in fluxes
471 of highly ^{13}C -depleted carbon to and from the ocean or atmosphere, such as changes in the release
472 and storage of organic carbon (Shackleton, 1986; Dickens et al., 1997; Kurtz et al., 2003; Deconto et
473 al., 2010; Komar et al., 2013). The $\delta^{13}\text{C}$ record at Cicogna offers no direct insight on the location of
474 this carbon (e.g., seafloor methane, permafrost, peat). However, it does support an important

475 concept: the magnitudes of given CIEs appear somewhat related to one another and to the long-
476 term $\delta^{13}\text{C}$ record. In particular, the PETM occurred about halfway between the long-term high and
477 low in $\delta^{13}\text{C}$, and heralded a relatively long time interval lacking CIEs. A generic explanation is that a
478 very large mass of ^{13}C -depleted carbon was injected from some organic reservoir into the ocean or
479 atmosphere during the PETM, and that the reservoir needed to recharge for considerable time
480 before the next injection (H-1/ETM-2) could occur (Dickens et al., 2003; Kurtz et al., 2003; Lunt et
481 al., 2011; Komar et al., 2013).

482 The overall -1 ‰ offset of the $\delta^{13}\text{C}$ curve between the records at Cicogna and at Sites 577 and
483 1262 (**Figure 5**) warrants brief discussion. It probably does not reflect wholesale diagenesis and
484 resetting of the primary signal at any of these sections. Otherwise, a recognizable correlative $\delta^{13}\text{C}$
485 record and well-preserved nannofossils (**Plate I**) would not be found at all three locations. In fact, it
486 is difficult to modify the original $\delta^{13}\text{C}$ composition of carbonate over appreciable distance (> than
487 several meters) in marine sedimentary sequences dominated by fine grained calcite, even those
488 now exposed on land as lithified rock, such as at Cicogna or in the Clarence Valley. This is because
489 the carbon water/rock ratio remains low, because almost all carbon exists in carbonate, and because
490 temperature minimally influences carbon isotope fractionation (Matter et al., 1977; Scholle and
491 Arthur, 1980; Frank et al., 1999). Instead, the offset in the $\delta^{13}\text{C}$ curves probably relates to differences
492 in the composition of the original carbonate, a concept that we return to later.

493 However, local dissolution and re-precipitation of carbonate definitely has occurred in the
494 Cicogna section. This can be observed in the overgrowths of secondary calcite on discoasters and
495 *Rhombaster/Tribrachiatus* (**Plate I**). This process should dampen the original CIEs, because on the
496 meter-scale, dissolution and re-precipitation of carbonate would involve $\delta^{13}\text{C}$ gradients in the DIC
497 of surrounding pore water (Matter et al., 1977; Scholle and Arthur, 1980). This may explain, in part,

498 why the magnitude of early Paleogene CIEs in bulk carbonate records are often muted relative to
499 those found in other carbon-bearing phases (Slotnick et al., 2015b).

500

501 **5.2 Oxygen isotopes and a problem recording past temperatures**

502

503 The $\delta^{18}\text{O}$ record at Cicogna is intriguing because many of the CIEs are characterized by negative
504 excursions but absolute values of $\delta^{18}\text{O}$ generally and unexpectedly increase upsection (**Figure 4**).
505 Similar results have been documented in bulk carbonate stable isotope records at other locations,
506 such as ODP Site 1215 (Leon-Rodriguez and Dickens, 2010) and Mead Stream (Slotnick et al., 2012).
507 Even the $\delta^{18}\text{O}$ record of bulk carbonate at Site 1262 shows minimal long-term change from the late
508 Paleocene to the early Eocene (Zachos et al., 2010), the time when high-latitude surface
509 temperatures and deep ocean temperatures presumably increased by 5-6 °C, and one might expect
510 a >1 ‰ decrease in the $\delta^{18}\text{O}$ of marine carbonate.

511 Like previous workers, we cannot discount the notion that temperatures at low and high
512 latitudes responded differently across the early Paleogene (Pearson et al., 2007; Huber and
513 Caballero, 2011). Unlike for carbon isotopes, however, local dissolution and re-precipitation of
514 carbonate should significantly impact the $\delta^{18}\text{O}$ of marine carbonate. This is because the oxygen
515 water/rock ratio would be high before lithification, and because temperature strongly influences
516 oxygen isotope fractionation (Matter et al., 1975; Scholle and Arthur, 1980; Frank et al., 1999). In
517 general, as calcite-rich sediments and surrounding pore water are buried to higher temperatures
518 along a geothermal gradient, local dissolution and re-precipitation of carbonate shifts carbonate
519 $\delta^{18}\text{O}$ to lower values (above references; Schrag et al., 1995). It is likely that, during sediment burial,
520 the bulk carbonate $\delta^{18}\text{O}$ records in many lower Paleogene sections, including at Cicogna, have been
521 modified. We suggest that a signal of surface ocean temperature changes remains in the Cicogna

522 section, which gives rise to short-term $\delta^{18}\text{O}$ excursions that coincide with CIEs and several known or
523 suspected hyperthermal events. However, the entire $\delta^{18}\text{O}$ record at this location likely has shifted
524 to more negative values preferentially with increasing burial depth and age. This partly explains the
525 observed relationship between bulk carbonate $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$, which lies along a trajectory expected
526 for diagenesis (**Figure 6**). A potential test of this idea would be to show that the overgrowths on
527 nanofossils (**Plate I**) have a significantly lower $\delta^{18}\text{O}$ than the primary core carbonate of nanofossil
528 tests.

529

530 **5.3 Calcareous nanofossil assemblages within the context of correlative stable isotope records**

531

532 A detailed stable carbon isotope curve provides a powerful tool to place past changes in calcareous
533 nanofossil assemblages into a highly resolved framework. This is because, as implied above, truly
534 global changes in the $\delta^{13}\text{C}$ composition of the ocean should occur within the cycling time of carbon
535 through ocean, which is <2000 years at present-day and presumably for the entire Cenozoic
536 (Broecker and Peng, 1982; Shackleton, 1986; Dickens et al., 1997).

537 Across the study interval at Cicogna, several calcareous nanofossil taxa appear or disappear
538 (**Table 1**). Moreover, their abundances also change between these horizons (**Figures 7-9**). One might
539 hypothesize that these changes in nanofossil assemblages were related to the established (e.g.,
540 the PETM, H1/ETM-2 and K/X) and potential (e.g., the B1/B2, I1/I2) hyperthermal events that
541 occurred during the late Paleocene and early Eocene (**Figures 1, 5**). However, the timing between
542 recorded evolutionary appearances and extinctions of calcareous nanofossils and perturbations in
543 $\delta^{13}\text{C}$ are variable (**Figures 7-9**). For instance, several significant calcareous nanofossil changes
544 observed close to H1/H2 hyperthermals (e.g., *B. T. othostylus*, *B. S. radians*, *B. S. villae*, *Tc. D.*
545 *multiradiatus*, *T. T. contortus*) predate these events. By contrast, several biotic changes observed

546 close to the B1/B2 CIEs (e.g., *B D. delicatus*, *Tc S. anarrhopus*, *B D. multiradiatus*, *T Ericsonia robusta*)
547 happened at the end of these events. The PETM seems to provide the only case when a negative
548 CIE precisely corresponds to major changes in calcareous nannofossil assemblages.

549 Profound changes in calcareous nannofossil assemblages occurred across the PETM in several
550 locations (**Figure 2**), both in terms of relative abundances and increases in origination and extinction
551 rates (Aubry, 1998; Bown et al., 2004; Raffi et al., 2005; Gibbs et al., 2006a; Agnini et al., 2007a; Self-
552 Trail et al., 2012). At Cicogna, the assemblages show remarkable, though mostly transient, relative
553 abundance variations across the PETM, including an increase in *Coccolithus*, a decrease in
554 *Zygrhablithus*, *Sphenolithus*, *Toweius* and *Prinsius*, and an extinction of most fasciculith species
555 (**Figure 8**). Not surprisingly, these changes are very similar to those in the Forada section, which is
556 also located in the Belluno Basin (Agnini et al., 2007a).

557 Although these changes in relative abundance of taxa alone represent a notable difference with
558 respect to background conditions, most of the changes are transient and/or local when compared
559 with other datasets (Bralower, 2002; Gibbs et al., 2006b; Agnini et al., 2007b; Angori et al., 2007;
560 Mutterlose et al., 2007). For instance, an increase in abundance of *Discoaster* and *Fasciculithus* was
561 reported for some of the PETM section studied (e.g., Bralower, 2002; Tremolada and Bralower,
562 2004; Raffi et al. 2009), but these assemblage variations were not observed in other sections (e.g.,
563 Gibbs et al. 2006; Agnini et al., 2007a; Self-Trail et al., 2012). The only global calcareous nannofossil
564 assemblage features of the PETM are represented by the evolutionary appearance of
565 *Rhomboaster/Tribrachiatus* lineage, the presence during the CIE of short-lived species such as
566 *Discoaster areneus*, and the disappearance of several species of fasciculiths (Raffi et al., 2005; Agnini
567 et al., 2007a).

568 While changes in calcareous nannoplankton assemblages during the PETM have been
569 investigated at high resolution at different locations (e.g., Bralower, 2002, Gibbs et al., 2006b; Agnini

570 et al. 2007a), the longer-term perspective in which such changes occurred during the early
571 Paleogene has remained uncertain (Gibbs et al., 2012). The record at Cicogna provides this
572 opportunity.

573 The PCA analysis of calcareous nannofossil census data (%) indicates that two principal
574 components (PC1 and PC2) account for most (56.0%) of the variability in our 15 selected subgroups.

575 Such analysis also permits the studied samples to be subdivided into ~~three-two~~ populations and a
576 possible widely dispersed group (Figure 10A). The first two populations are distinguished because
577 of a major difference along the -x-axis representing PC1, whereas the third population seems to
578 stands out because of a ~~significant~~ difference along the y-axis representing PC2. Importantly, each
579 of these three populations constitutes a homogeneous group in the time domain: Group 1 includes
580 all upper Paleocene samples (Paleocene samples and B1/B2 events); Group 2 consists of almost all
581 lower Eocene samples (Eocene samples, H1/H2 events and K event); Group 3 comprises samples
582 that span the PETM (both core and recovery), and two samples that come from sediment deposited
583 during the core of the H1 and B2 events (Figure 10). These results indicate that late Paleocene
584 calcareous nannofossil assemblages are statistically different in their composition from those of
585 early Eocene samples. ~~Moreover, the~~To check if calcareous nannofossil assemblages across the
586 PETM, ~~and the climax of the B2 and H1 events,~~ are statistically different from those of either the
587 late Paleocene or the early Eocene, we performed a MANOVA analysis, which pointed out that
588 ellipses containing 95% of the data points for each group (late Paleocene, early Eocene and PETM)
589 are virtually not overlapping one to each other suggesting that three statistically different
590 populations are recognized across the PETM, the late Paleocene and the early Eocene background
591 assemblages, and the PETM fossil associations.

592

593 The general shift in the relative abundance of placoliths (i.e., *Coccolithus*, *Toweius* and *Prinsius*),
594 the major component of the late Paleocene assemblages, to nannoliths/holococcoliths (i.e.,
595 *Sphenolithus* and *Zygrhablithus*), the major component of the early Eocene assemblages, largely
596 explains the PC1 component [or Axis 1 \(Figure 10\)](#). By contrast, the dramatic shift toward negative
597 values in the PC2 component [or Axis 2](#) during the PETM happens because of the increase of *Ericsonia*
598 and reworking and the presence of *Rhomboaster-Tibrachiatus* plexus. Presumably, this relates to
599 peculiar paleoenvironmental conditions that developed during the event. One can hypothesize that
600 this may have been a major difference in the physicochemical parameters of sea surface waters,
601 such as higher temperatures, higher nutrient concentration or reduced carbonate saturation state.

602 Statistical analysis of our data from Cicogna does not highlight any prominent short-term
603 changes in calcareous nannofossil assemblages, other than across the PETM and perhaps the B2 and
604 H1 events. However, several biohorizons occur around the B1/B2 events. Specifically, these are the
605 Bc *Z. bijugatus*, the brief high abundance of *Octolithus* spp., the evolutionary onset of the *D.*
606 *delicatus*/*D. multiradiatus* lineage, the presence of the short-ranged *E. robusta*, the final radiation
607 of late Paleocene fasciculiths (i.e., *F. richardii* group, *F. hayi*, *F. liliana*, *F. alanii*), and the Tc of *S.*
608 *anarrhopus*. All these happened at Cicogna and at Site 1262 within Chron C25n (Agnini et al., 2007b;
609 Dallanave et al., 2009; **Figure 11**), which spanned only 0.54 Myr (Westerhold et al., 2008). These
610 near-synchronous events are intriguing because while the various nannofossils represent only minor
611 components of late Paleogene assemblages, they were destined to become either an abundant
612 constituent of Eocene populations (e.g., *Z. bijugatus* and the *D. delicatus*/*D. multiradiatus* lineage),
613 or extinct after having been a distinctive element of Paleocene assemblages (e.g. *Fasciculithus* spp.
614 and *S. anarrhopus*). Following the PCIM, the long-term increase in temperature and decrease in $\delta^{13}\text{C}$
615 (**Figure 1**) coincided with a series of minor changes in nannofossil assemblages, which subsequently
616 became important, presumably for evolutionary reasons.

617 Similar to the late Paleocene, calcareous nannofossil assemblages after the PETM do not show
618 major rearrangements of common taxa during the early Eocene. Instead, minor components of
619 these assemblages exhibit a sequence of closely spaced biohorizons. The sequence of these
620 biohorizons is: T *Fasciculithus*, B *D. diastypus*, B *T. contortus*, T *T. bramlettei*, Tc *D. multiradiatus*, T
621 *T. contortus*, B *T. orthostylus*, B *S. radians*, T *D. multiradiatus*, B *D. lodoensis*, B *G. gammation* and
622 Bc *D. lodoensis* (**Table 1**). Within the resolution of available paleomagnetic and $\delta^{13}\text{C}$ data, all these
623 biohorizons are virtually synchronous between the Cicogna section and ODP Site 1262 (**Figure 11**).
624 They also almost all occurred in near synchrony at Site 577 (Dickens and Backman, 2013), although
625 the precise correlation remains uncertain, given problems with coring disturbance and subtleties in
626 age models at this location.

627 Importantly, for stratigraphic purposes, the B and Bc of *D. lodoensis* are approximately coeval
628 at all three locations and spaced apart by about 750 kyr. Unless one examines samples in detail,
629 these two biohorizons can be confused and result in an erroneous assignment of early Eocene ages.

630 The evolutionary appearances and extinctions amongst early Eocene nannofossil assemblages
631 may suggest the presence of uneven communities living in an extreme climate in which alterations
632 of environmental conditions, even minor, might trigger evolutionary changes or prominent
633 variations in abundances of a limited number of taxa that typically do not represent the dominant
634 component of assemblages. explained possible explanation is a generally higher tolerance of
635 cosmopolitan taxa to variations in environmental conditions (Boucot, 1975; Winter et al., 1994). In
636 contrast, highly specialized taxa that are adapted to a particular ecological niche, may display
637 greater sensitivity to modifications in the photic zone environment (MacArthur and Wilson, 1967;
638 Pianka, 1970; Baumann et al., 2005).

639 In summary, several genera of calcareous nannofossils, such as *Rhomboaster*, *Tribrachiatulus*,
640 *Sphenolithus*, *Discoaster* and *Zygrhablithus* were, at least to some extent, affected during the late

641 Paleocene-early Eocene transition, because they show an increased rate of taxonomic evolution
642 (**Figure 11**). However these genera are all minor groups in terms of overall abundance, at least in
643 most lower Paleogene sediment sequences, and they all belong to nannoliths and holococcoliths. It
644 appears that these organisms were more sensitive to environmental changes than heterococcoliths,
645 for example the cosmopolitan genera *Coccolithus* and *Toweius*.

646

647 **5.4 Early Paleogene calcareous nannofossil evolution**

648

649 Any comprehensive paleoenvironmental interpretation involving early Paleogene calcareous
650 nannofossils remains tentative because many taxa, such as *Rhomboaster/Tribrachiatus*, *Discoaster*,
651 *Sphenolithus* and *Zygrhablithus*, are extinct. Still, some single species or species groups are
652 considered to be useful for reconstructions of paleoenvironmental conditions (Geisen et al., 2004).
653 With that viewpoint, and with an understanding of modern holococcolith/nannolith ecology and
654 classical biogeographical model, we provide a scenario regarding late Paleocene-early Eocene
655 calcareous nannofossil evolution.

656 Modern holococcolithophores have numerous tiny rhombohedral calcite crystallites, and are
657 considered as haploid stages of certain heterococcolithophores, which can live in just about any
658 marine photic zone environment, although higher abundances and diversity are typical in
659 oligotrophic settings (Billard and Inouye, 2004). The most common Paleogene holococcolith was
660 *Zygrhablithus bijugatus*. This taxon has been interpreted as a *K*-specialist more adapted to stable
661 environments and oligotrophic conditions (Aubry, 1998; Bralower, 2002; Tremolada and Bralower,
662 2004; Agnini et al., 2007a; Self-Trail et al., 2012). Nannolith is a term used to describe peculiar
663 morphotypes usually observed in association with coccoliths, but lacking the typical features of
664 heterococcoliths or holococcoliths. *Ceratolithus cristatus*, a modern nannolith, has been observed

665 on the same cell together with *Neosphaera coccolithomorpha* (Alcolber and Jordan, 1997),
666 suggesting that the nannolith stage (*C. cristatus*) corresponds to the holococcolith stage in other
667 taxa (Young et al., 2005). Paleogene nannoliths include taxa with peculiar morphologies such as
668 *Discoaster*, *Fasciculithus* and *Sphenolithus*. These genera often have been associated with warm
669 waters and oligotrophic environments and are almost unanimously interpreted as *K*-specialists (Haq
670 and Lohmann, 1976; Backman, 1986; Wei and Wise, 1990; Bralower, 2002; Gibbs et al., 2004; 2006a;
671 2006b; Agnini et al., 2007a). *K* specialists fluctuate at or near the carrying capacity (*K*) of the
672 environment in which they thrive (MacArthur and Wilson, 1967), and are usually characterized by
673 long individual life-cycles and low reproductive potential. The *K*-specialist strategy is advantageous
674 in highly stable, typically oligotrophic environments, which allows the evolution of stenotopy and
675 where organisms compete by specialization and habitat partitioning (Hallock, 1987; Premoli Silva
676 and Sliter, 1999). The narrow range of adaptability to changes in habitat or ecological conditions
677 stimulates a rapid speciation.

678 At present, it is commonly accepted that modern holococcoliths and nannoliths are not
679 produced by autonomous organisms; rather, they are stages in the life cycle of coccolithophores.,
680 Moreover, the passage between the two stages may be triggered by environmental factors (Billard
681 and Inouye, 2004).

682 Hence, though Paleogene holococcoliths/nannoliths have no direct descendants in present-day
683 oceans, they may very well have shared similar physiological features and life cycles with modern
684 taxa. Assuming this is the case, the increase in the relative abundance of holococcoliths and
685 nannoliths at the expense of heterococcoliths as well as the higher rates of evolution shown by
686 holococcoliths and nannoliths may suggest conditions in which highly specialized taxa could flourish
687 and rapidly evolve. This scenario is consistent with the idea, based on laboratory and modern ocean
688 data, that the calcareous nannoplankton response to environmental change is species or group

689 specific rather than homogeneous across the entire assemblage (Riebesell et al., 2000; Langer et al.,
690 2006; Iglesias-Rodriguez et al., 2008; Lohbeck et al., 2012). Variations in the thermal and chemical
691 structure of photic zone waters may thus account for the observed changes in the early Paleogene
692 calcareous nannofossil assemblages.

693

694 **5.5 Carbon isotope of surface waters during the early Paleogene**

695

696 Like at Cicogna, well-preserved calcareous nannofossils dominate bulk sediment carbonate contents
697 of early Paleogene strata at Sites 577 and 1262 (Backman, 1986; Zachos et al., 2004; Dickens and
698 Backman, 2013). Given that the nannofossil assemblages are fairly similar (**Figure 11**), a really basic
699 question returns: why is the overall early Paleogene bulk carbonate $\delta^{13}\text{C}$ record at Cicogna less by
700 approximately 1 ‰?

701 A variety of explanations for the $\delta^{13}\text{C}$ offset can be offered. For example, sediments at Cicogna had
702 greater amounts of organic matter, and during burial diagenesis, a fraction of this carbon was
703 consistently added so as to decrease the $\delta^{13}\text{C}$ of pore water DIC. We note, though, that C_{org} contents
704 (wt %) at the proximal Forada section generally have values less than 0.1 wt % (Giusberti et al.,
705 2007). Similar C_{org} contents are found at ODP Site 1262, where values range from 0.0 to 0.3 wt %
706 (Zachos et al., 2004).

707 A cursory examination of early Paleogene bulk carbonate $\delta^{13}\text{C}$ records from other sites of the
708 North Atlantic/western Tethys region (e.g., Sites 550 and 1051; **Figure 2**) shows a commonality:
709 these locations also display negative 0.5 to 1 ‰ offsets relative to correlative records at Sites 577
710 and 1262 (Cramer et al., 2003). The $\delta^{13}\text{C}$ of DIC in modern surface waters (<100 m) ranges by about
711 2 ‰, because of the differences in temperature, primary productivity and water mass mixing
712 (Kroopnick, 1985; Tagliabue and Bopp, 2008). Notably, however, gradients in $\delta^{13}\text{C}$ of surface water

713 DIC are gradual, such that large regions have fairly similar values. It is possible that bulk carbonate
714 $\delta^{13}\text{C}$ values in early Paleogene North Atlantic sections record lower values than locations near the
715 Equator or in southern latitudes because of past ocean circulation.

716

717 **6 SUMMARY AND CONCLUSIONS**

718

719 We generate records of bulk carbonate $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$, CaCO_3 content and calcareous nannofossil
720 assemblages from the Cicogna section, a marine sedimentary succession that now crops out along
721 a stream in the Southern Alps of northeast Italy. The combined geochemical and calcareous
722 nannofossil results allow us to generate a detailed stratigraphy for the section, as well as to explore
723 relationships between stable isotope variations and nannofossil assemblages. Most lower
724 Paleogene sections examined to date lack such coupled data sets.

725 The $\delta^{13}\text{C}$ record and calcareous nannofossil assemblages show that the section spans ~5.3 Myr
726 of the late Paleocene and early Eocene interval, from 57.5 to 52.2 Ma on the WO-1 timescale. This
727 is consistent with previous paleomagnetic information and preliminary calcareous nannofossil
728 biostratigraphy (Dallanave et al., 2009), but provides a more detailed stratigraphic framework, one
729 appropriate for correlations to other locations around the world. In particular, the fairly well
730 resolved $\delta^{13}\text{C}$ record shows long-term and short variations that correspond to those found in several
731 other sections, including an established series of negative CIEs. The most prominent CIE marks the
732 PETM, while other less pronounced CIEs represent the H-1, K/X and other “events” documented
733 elsewhere. The $\delta^{13}\text{C}$ variations observed at Cicogna clearly reflect global changes in the fluxes of
734 carbon to and from the ocean and atmosphere.

735 PCA analysis of calcareous nannofossil assemblages shows three distinct sample clusters. Late
736 Paleocene and early Eocene assemblages were distinctly different from each other and from that of

737 the PETM. Indeed, the PETM, the most intense hyperthermal during the late Paleocene - early
738 Eocene, was characterized by a unique calcareous nannofossil assemblage composition. This
739 suggests that the brief episode of extreme warming permanently modified the composition of
740 calcareous nanoplankton through an increase in the rate of taxonomic evolution (Gibbs et al.,
741 2006a). Less prominent hyperthermal events do not show significant variations in the main
742 components of assemblages, but rather were characterized by a series of changes affecting a limited
743 number of rare taxa. These taxa may have been less tolerant to environmental changes in their
744 habitats.

745 More common taxa, essentially consisting of placoliths, such as the cosmopolitan *Coccolithus*
746 and *Toweius*, display a progressive long-term decrease interrupted by transient changes in their
747 relative abundance but virtually no extinction or origination events occur in these groups during the
748 pertinent time interval. Species belonging to nannoliths and holococcoliths (*Discoaster*,
749 *Fasciculithus*, *Rhomboaster/Tribrachiatus*, *Sphenolithus* and *Zygrhablithus*), generally show a higher
750 rate of evolution and a higher concentration of biohorizons close to $\delta^{13}\text{C}$ perturbations. In
751 conclusion, calcareous nanoplankton show a different response of the various components of the
752 assemblages, this is consistent with a species or taxonomic unit sensitivity of calcareous
753 phytoplankton to paleoenvironmental perturbations. This evolutionary climate-forced model is
754 supported by data from ODP Site 1262, which demonstrate that these changes are global and
755 synchronous between middle latitudes in the western Tethys region and the South Atlantic.

756

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764 **8** **FIGURE AND TABLE CAPTIONS**

765 **Figure 1.** Middle Paleocene to middle Eocene (64 to 48 Ma) stable isotope ($\delta^{13}\text{C}$ and $\delta^{18}\text{O}$) records
766 of benthic foraminifera from multiple locations (Zachos et al., 2008) placed on the Option 1 (W01)
767 time scale of Westerhold et al. (2008). Also shown are positions of polarity chrons and calcareous
768 nannofossil biozones for this time interval, both from the CP Biozone scheme (Okada and Bukry,
769 1980) and the CN Biozone scheme (Agnini et al., 2014). Various “events” are noted within this
770 chronostratigraphic framework, including the Paleocene carbon isotope maximum (PCIM), the
771 Paleocene-Eocene thermal maximum (PETM), the H-1/ETM-2 event, the K/X event, and the Early
772 Eocene Climatic Optimum (EECO). To the right is the general lithologic column and
773 magnetostratigraphy of the Cicogna section (Dallanave et al., 2009).

774
775 **Figure 2.** Paleogeographic map indicating approximate locations at 55 Ma for several key sites with
776 detailed stable isotope records across the late Paleocene and early Eocene. These include (marked
777 with black dots and star) the Cicogna section (NE Italy, this study), DSDP Site 577 (Shatsky Rise,
778 Dickens and Backman, 2013), ODP Sites 1051 (Blake Nose, Ogg and Bardot, 2001), 1215 (central
779 Pacific, Raffi et al., 2005), and 1262 (Walvis Ridge, Westerhold et al., 2008), and the Clarence Valley
780 (CV) sections (New Zealand, Slotnick et al., 2015b). The grey areas represent plate fragments, while
781 the black lines show present-day shorelines. Boxes next to site locations show average
782 sedimentation rates from the base of Chron C25n to the base of Chron C23r (57.20 - 52.36 Ma). The
783 base map is from <http://www.odsn.de/odsn/services/paleomap/paleomap.html>. Red triangles are
784 locations where a decrease in diversity of *Fasciculithus* spp. has been documented near the PETM.
785 Locations include the Clarence Valley sections, central Pacific (ODP Sites 1215, 1220, 1221), western
786 Pacific (DSDP Site 577 and ODP Site 865), South Atlantic (Walvis Ridge, DSDP Site 527, ODP Sites
787 1262, 1263-1267; Maud Rise, ODP Site 690), equatorial Atlantic (Ceara Rise, ODP Site 929, Demerara

788 Rise, ODP Sites 1259, 1260), northwestern Atlantic (New Jersey Margin land sections, ODP Site 1051,
789 IODP Site U1403, U1409), northeastern Atlantic (Bay of Biscay DSDP Sites 401, 549 and 550, Zumaya
790 land section), Indian Ocean (DSDP Site 213; ODP 672; Kerguelen Plateau, ODP Site 1135)(Backman,
791 1986; Aubry, 1999; Bralower, 2002; Dupuis et al., 2003; Tremolada and Bralower, 2004; Bralower
792 and Mutterlose, 1995; Monechi et al., 2000; Gibbs et al., 2004; Raffi et al., 2005; Agnini et al., 2007a;
793 Angori et al., 2007, Mutterlose et al., 2007; Jiang and Wise, 2009; Shamrock, 2010; Norris et al, 2014;
794 Dallanave et al., 2015).

795

796 **Figure 3.** The location and representative photographs of the Cicogna section in northeast Italy. (a)
797 Geographic map showing the main Late Cretaceous–early Paleogene paleogeographic domains of
798 the Italian Southern Alps (modified after Cati et al., 1989); (b) Geological map of the local area
799 (modified after Costa et al, 1996 indicating also the location of the Cicogna section (red asterisk)); (c)
800 Alternating beds of Paleocene gray-green marls and calcareous marls (0-20 m); (d) The Scaglia Rossa
801 *sensu lato* overlain by the Belluno Flysch; (e) Marl/calcareous marl couplets in the lower Eocene
802 portion of the section (approximately 40.0-70.0 m); (f) The base of the Clay Marl Unit, which denotes
803 the onset of the PETM (approximately 28.7-29.3 m); (g) The brownish-red interval of clayey marls
804 with sporadic grey-green cm-scale spots and lenses, the CMU, overlain by prominent rhythmic
805 alternations of marls and calcareous marls (approximately 28.7-33.0 m).

806

807 **Figure 4.** The Cicogna section with records of bulk carbonate $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ data, and CaCO_3 content.
808 Calcareous nannofossil biostratigraphy (CP and NP biozones) and magnetostratigraphy are after
809 Dallanave et al. (2009), CN biozones are also reported. Orange and yellow bands mark major $\delta^{13}\text{C}$
810 excursions. Dashed lines indicate minor CIEs that have been labeled elsewhere (e.g., E1/E2, F and

811 G; Cramer et al., 2003), whereas dotted lines indicate minor changes in $\delta^{13}\text{C}$ that appear to occur
812 also at ODP Site 1262 (see also **Figure 5**).

813

814 **Figure 5.** Stratigraphic correlation between upper Paleocene and lower Eocene sections at Cicogna,
815 ODP Site 1262 (Zachos et al., 2010), and DSDP Site 577 (Cramer et al., 2003; Dickens and Backman,
816 2013). All three sites have independently derived nannofossil biohorizons, polarity chrons and $\delta^{13}\text{C}$
817 records, which account for subtle temporal offsets. . Color bands and symbols are the same as in
818 Figure 4. Note the missing record at Site 577 that corresponds with known core gaps.

819

820 **Figure 6.** Plots of (a) bulk carbonate $\delta^{13}\text{C}$ versus bulk oxygen $\delta^{18}\text{O}$, and (b) bulk carbonate $\delta^{13}\text{C}$ versus
821 CaCO_3 content for samples from the Cicogna section. The black arrow shows the expected effect of
822 burial diagenesis. Grey and black dash lines are linear trendlines for Paleocene and Eocene samples,
823 respectively. → Note the clear distinction in $\delta^{13}\text{C}$ for Paleocene and Eocene samples, which relates
824 to a long-term decrease in $\delta^{13}\text{C}$ (**Figure 5**).

825

826 **Figure 7.** Relative (%) and semi-quantitative (N/mm^2) abundances of selected calcareous
827 nannofossil genera across the Cicogna section. Also shown are the lithostratigraphy,
828 magnetostratigraphy, biostratigraphy and carbon isotope ($\delta^{13}\text{C}$) stratigraphy at the Cicogna section
829 (**Figure 4**). Color bands and symbols are the same as in previous figures.

830

831 **Figure 8.** Relative (%) and semi-quantitative (N/mm^2) abundances of selected, mainly late
832 Paleocene, calcareous nannofossil taxa across the Cicogna section. Also shown are the
833 lithostratigraphy, magnetostratigraphy, biostratigraphy and carbon isotope ($\delta^{13}\text{C}$) stratigraphy at
834 the Cicogna section (**Figure 4**). Color bands and symbols are the same as in previous figures.

835 **Figure 9.** Relative (%) and semi-quantitative (N/mm²) abundances of selected, mainly early Eocene,
836 calcareous nannofossil taxa across the Cicogna section. Also shown are the lithostratigraphy,
837 magnetostratigraphy, biostratigraphy and carbon isotope ($\delta^{13}\text{C}$) stratigraphy at the Cicogna section
838 (**Figure 4**). Color bands and symbols are the same as in previous figures.

839

840 **Figure 10.** Statistical Analyses of calcareous nannofossil percentage data of the Cicogna section.

841 Calcareous nannofossils are subdivided into 15 subgroups (*Chiasmolithus*, *Coccolithus*, *Ellipsolithus*,
842 *Discoaster*, *Ericsonia*, *Fasciculithus*, *Girgisia*, *Octolithus*, *Prinsius*, *Sphenolithus*, *Toweius*,
843 *Rhombaster/Tribrachiatus*, *Zyghrablithus*, reworking, others). **A)** Principal Component Analysis

844 (PCA) of calcareous nannofossil percentage data of the Cicogna section. Calcareous nannofossils are
845 subdivided into 15 subgroups (*Chiasmolithus*, *Coccolithus*, *Ellipsolithus*, *Discoaster*, *Ericsonia*,
846 *Fasciculithus*, *Girgisia*, *Octolithus*, *Prinsius*, *Sphenolithus*, *Toweius*, *Rhombaster/Tribrachiatus*,
847 *Zyghrablithus*, reworking, others). Scatter plot of percentage data of calcareous nannofossil taxa of

848 samples from the Cicogna section in terms of the first and second component. Each sample is
849 represented by a circle and labelled. Different colors serve to separate sub-sets of samples having

850 the same age. Green and blue shaded areas are the ellipses containing 95% of the data points of
851 Paleocene Group and Eocene group, respectively; **B)** Multivariate Analysis Of VAriance (MANOVA).

852 Scatter graph and biplot. Each sample is represented by a black symbol (quadrangle and diamond)
853 and labelled. Green, red and blue shaded areas are the ellipses containing 95% of the data points of

854 Paleocene Group, PETM group and Eocene group, respectively.

855

856 **Figure 11.** Comparison of $\delta^{13}\text{C}$ profiles and semi-quantitative abundance patterns of selected
857 calcareous nannofossil taxa from the Cicogna section and ODP Site 1262. Calcareous nannofossil

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858 biohorizons from DSDP Site 577 are reported in the right part of the figure. Orange and yellow bands
859 mark CIEs shown in previous figures.). Color bands and symbols are the same as in previous figures.

860

861 **Plate I.** Images of selected calcareous nannofossil taxa from samples of the Cicogna section. Scale
862 bar 10µm.

863 **1.** *Discoaster lodoensis* Bramlette and Riedel, 1954. Parallel light. Parallel nicols. Sample CIC/07-492.

864 **2-3.** *Girgisia gammation* (Bramlette Sullivan, 1961) Varol 1989. Crossed nicols. Sample CIC/07-437.

865 **4-5.** *Chiphragmalithus calathus* Bramlette and Sullivan, 1961; 4.Parallel light; 5. Crossed nicols.

866 Sample CIC/07-447. **6-7.** *Sphenolithus radians* Deflandre in Grassé, 1952. 6. Crossed nicols 0°; 7.

867 Crossed nicols 45°. Sample CIC/07-437. **8.** *Tribrachiatulus orthostylus* Shamrai, 1963. Parallel light.

868 Sample 208-1262A-11H- 1, 149. Sample CIC/07-447. **9-10.** *Tribrachiatulus contortus* (Stradner 1958)

869 Bukry 1972. Parallel light. Sample CIC/07-335. **11-13.** *Zyghrablithus bijugatus* (Deflandre in

870 Deflandre and Fert, 1954) Deflandre, 1959. Crossed nicols. Sample CIC/07-437. **14.** *Discoaster*

871 *salisburgensis* Stradner, 1961. Parallel light. Sample CIC/07-335. **15.** *Discoaster diastypus* Bramlette

872 and Sullivan, 1961. Parallel light. Sample CIC/07-335. **16.** *Fasciculithus tympaniformis* Hay and

873 Mohler in Hay et al. 1967. Crossed nicols. Sample CIC/07-335. **17.** *Octolithus multiplus* (Perch-

874 Nielsen, 1973) Romein, 1979. Crossed nicols. Sample CIC/07-122. **18.** *Discoaster multiradiatus*

875 Bramlette and Riedel 1954. Parallel light. Sample CIC/07-122. **19.** *Toweius pertusus* (Sullivan, 1965)

876 Romein, 1979. Crossed nicols. Sample CIC/07-122. **20.** *Toweius occultatus* (Locker, 1967) Perch-

877 Nielsen, 1971. Crossed nicols. Sample CIC/07-122. **21.** *Toweius eminens* (Bramlette and Sullivan,

878 1961) Perch-Nielsen, 1971. Crossed nicols. Sample CIC/07-029. **22.** *Toweius eminens* (Bramlette and

879 Sullivan, 1961) Perch-Nielsen, 1971. Crossed nicols. Sample CIC/07-029. **23.** *Toweius eminens*

880 (Bramlette and Sullivan, 1961) Perch-Nielsen, 1971. Crossed nicols. Sample CIC/07-122. **24.** *Prinsius*

881 *bisulcus* (Stradner, 1963) Hay and Mohler, 1967. Crossed nicols. Sample CIC/07-029. **25.** *Ericsonia*

882 *robusta* Bramlette and Sullivan 1961. Crossed nicols. Sample CIC/07-029. **26-27.** *Sphenolithus*
883 *anarrhopus* Bukry and Bramlette, 1969. 24. Crossed nicols 0°; 25. Crossed nicols 45°. Sample CIC/07-
884 029. **28-29.** *Zyghrablithus bijugatus* (Deflandre in Deflandre and Fert, 1954) Deflandre, 1959.
885 Crossed nicols. Sample CIC/07-122. **30.** *Thoracosphaera saxea* (Stradner, 1961). Crossed nicols.
886 Sample CIC/07-122.

887

888 **Table 1.** Stratigraphic heights and ages of polarity chron boundaries, key calcareous nannofossil
889 datums, and CIEs at the Cicogna Section and ODP Site 1262.

890

891 9. SUPPLEMENTARY INFORMATION

892 **S12.** Additional information on statistical analysis

893 Nannofossil data from the Cicogna section (NE Italy) were subjected to statistical analysis using the
894 program PAST.

895 For PCA analysis, we additionally provide the biplot and the loading graphs of Component 1 and
896 Component 2 (**Figure S1**).

897 For non-metric multidimensional scaling (MDS) analysis, the species counts were combined to
898 produce a matrix of 15 genera. A square root transformation, used to standardize the matrix, was
899 chosen to minimize the influence of dominant taxa on the ordination (Schneider et al., 2011). Non-
900 metric multidimensional scaling (MDS), using the Bray–Curtis distance metric (**Figure S2**) was
901 applied in order to avoid assumptions as much as possible and guarantee the preservation of the
902 relative differences between the samples (McCune and Grace, 2002).

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907 Oregon, 2002.

908

909 **S2. Further explanation regarding biostratigraphic calcareous nannofossil counts**

910 The high abundance, widespread distribution and rapid evolution of calcareous nannofossils make
911 them one of the most powerful tool to date Cenozoic marine sediments. The use of semi-
912 quantitative counting and the gathering of high resolved datasets greatly enhance their correlation
913 potential (Backman et al., 2012; Agnini et al., 2014).

914 The methodology used in this study for samples of ODP Site 1262 is that proposed by Backman
915 and Shackleton (1983), which consists in counting the number of calcareous nannofossils belonging
916 to a specific taxon present in a prefixed area (1 mm²). Because of significant dilution by terrigenous
917 material in samples from the Cicogna section, we extended the study area to 9 mm². To further
918 appreciate the importance of semi-quantitative estimates and high-resolution sampling, we
919 compare the Top *D. multiradiatus* and Base *D. lodoensis* as recorded from the Cicogna section, ODP
920 Site 1262 and DSDP Site 550 (**Figure S3**). At Cicogna and ODP Site 1262, we provide detailed
921 abundance patterns of these two taxa. *Discoaster multiradiatus* shows a first decrease in abundance
922 preceding the H1 event and a definitive disappearance just before the onset of the I1 event.
923 *Discoaster lodoensis* displays a first presence in the I1 event, which is followed by an interval of
924 absence that eventually leads to its continuous and common presence close to the onset of the X
925 event (**Figure S3**). Datasets from the Cicogna section and ODP Site 1262 allow a very detailed
926 characterization of these two biohorizons and the recognition of peculiar features that are not
927 present in the low-resolution qualitative biostratigraphic data available for DSDP Site 550. As a
928 consequence, the stratigraphic position of Top *D. multiradiatus* and Base *D. lodoensis* at DSDP Site
929 550 are inaccurate. We hope that this simple exercise could serve to emphasize the crucial
930 importance of producing high-resolution semi-quantitative data to obtain the most reliable
931 biostratigraphic results.

932

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961
962 **S3. Looking through “frosty glass”: Comparison to records at ODP Site 690**

963 We have presented fairly detailed records of bulk carbonate $\delta^{13}\text{C}$ and quantified calcareous
964 nannofossil assemblages for the lower Paleogene section at Cicogna, and compared these records
965 with those at the only two locations with similar information. From this comparison, we suggest
966 that a very detailed template exists for the alignment of $\delta^{13}\text{C}$ records and calcareous nannofossil
967 assemblage counts across the early Paleogene (**Figure 11**), one with much higher resolution than
968 given in most previous work, and one most likely related to changes in past global carbon cycling,
969 oceanography, and calcareous nannoplankton evolution.

970 Significant variations in calcareous nannofossil abundances definitely happened at multiple
971 locations during the PETM (Bralower, 2002, and references noted in main text). However, it is by no

972 means clear whether such changes extended across the broader early Paleogene, nor how such
973 changes might compare to those across the PETM. One can certainly speculate that variations in
974 calcareous nannofossil abundance records and bulk carbonate $\delta^{13}\text{C}$ records might correlate in fine
975 temporal detail across widely distributed sites throughout the early Paleogene, given well-
976 established calcareous nannofossil biozone schemes (Martini, 1971; Okada and Bukry, 1980; Agnini
977 et al., 2014), and a growing appreciation of a very dynamic carbon cycle over this time interval.
978 Nonetheless, the generation of detailed and coupled multi-million year records for quantified
979 calcareous nannofossil abundances and bulk carbonate $\delta^{13}\text{C}$ perplexed one of the referees for this
980 paper, who insisted that we needed to make comparisons with existing work at ODP Site 690 and
981 to explain discrepancies.

982 The lower Paleogene record at Site 690 provides a very good example in which to highlight the
983 basic background and importance of our work. Three holes were drilled and cored at ODP Site 690
984 on Maud Rise (South Atlantic; Figure 1) in 1987 using the advanced piston corer (APC) (Barker et al.,
985 1988). Sediment recovery within each core was nearly 100 %, although some cores were shorter
986 than the full 9.7 m. However, most of the lower Paleogene sequence was retrieved in only one of
987 the holes, 690B (Barker et al., 1988). This is important, because m-scale gaps generally occur
988 between successive cores during APC operations (Ruddiman et al., 1987; Lisiecki and Herbert, 2007).
989 The early Paleogene section at Site 690 is, almost assuredly, incomplete, with “missing” portions at
990 each core break.

991 Sediment from Core 690B-19H has been the focus of numerous papers, as it contains the PETM
992 (Kennett and Stott, 1991; Bains et al., 1999; Bralower, 2002). However, correlating this core to the
993 surrounding sedimentary record at Site 690, and the latter to early Paleogene records at other
994 locations is problematic, at least with any detail. For example, using Hole 690B records, Cramer et
995 al. (2003) estimated that 1.4 Myr occurred between the PETM and the H-1 event. This is incorrect,

996 as the duration is close to 1.8 Myr (Westerhold et al., 2008). Beyond the aforementioned core gaps,
997 there are major issues with the paleomagnetic record of early Paleogene sediments in Hole 690B
998 (Ali et al., 2000). Indeed, Ali et al. (2000) recommend using calcareous nannofossil records for
999 correlation purposes of this interval.

1000 Records of bulk carbonate $\delta^{13}\text{C}$ (Cramer et al., 2003) and calcareous nannofossil relative
1001 abundances (Pospichal and Wise, 1990) have been generated using sediment at Hole 690B. When
1002 coupled together (**Figure S4**), these records show similarities to those at Cicogna (**Figure 11**). There
1003 is the long-term late Paleocene-early Eocene drop in $\delta^{13}\text{C}$ and several superimposed short-term
1004 negative CIEs. There are also closely coeval changes in calcareous nannofossil abundances, such as
1005 the peak in *D. multiradiatus* across the C event, the subsequent peak in *Fasciculithus* spp., and the
1006 cross-over of *T. contortus* and *T. orthostylus* just before the H-1 event. One can also see the problem
1007 with examining nannofossils at low depth/time resolution and qualitatively. We suggest here a
1008 “frosty glass” hypothesis, where details of Earth system change in the distant past are blurred
1009 presently by poorly resolved stratigraphy. This includes basic problems with aligning sections in
1010 depth and time, as well as interpretable quantification of data at high spatial resolution. Despite the
1011 need for additional work at Site 690, we suggest that available records at this location support the
1012 template offered in the main text.

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1050 **Supplementary figure captions** **Figure S1.** PCA plots of calcareous nannofossil data from the Cicogna
1051 section (Italy). A) Loading plot of Component 1; B) Loading plot of Component 2; C) Biplot.

1052 **Figure S2.** Non-metric multidimensional scaling (NMS) plot of calcareous nannofossil data from the
1053 Cicogna section (Italy). Grey dots = barren to virtually barren samples.

1054 **Figure S3.** Abundance patterns of *D. multiradiatus* and *D. Iodoensis* from the Cicogna section, ODP
1055 Site 1262 and DSDP Site 550. For these three successions paleomagnetic (Ali and Hailwood, 1998;
1056 Dallanave et al., 2009; Westerhold et al., 2008), carbon isotope (Cramer et al., 2003; Zachos et al.,
1057 2004; this study) and calcareous nannofossil data (Müller, 1985; Agnini et al., 2007, this study) are
1058 available. Top *D. multiradiatus* and Base *D. Iodoensis* are clearly recognizable at Cicogna and ODP
1059 Site 1262, where quantitative counts have been performed. By contrast, qualitative data from DSDP
1060 Site 550 do not provide reliable biostratigraphic data P= present; R=rare; F=few; C=common;
1061 A=abundant; V=very abundant.

1062 **Figure S4.** Carbon isotope data from ODP Site 690 (Cramer et al., 2003) plotted against qualitative
1063 abundance estimates of selected calcareous nannofossil taxa (Pospichal and Wei, 1990). Top
1064 *Fasciculithus* spp. (Aubry et al., 1996)

1065 **Table S1.** Bulk carbonate stable isotopes and carbonate content of samples from the Cicogna
1066 section.

1067 **Table S2.** Calcareous nannofossil assemblage counts for samples from the Cicogna section.

1068 **Table S3.** Dataset used to perform the principal component analysis for calcareous nannofossil
1069 assemblages from the Cicogna section. Calcareous nannofossils are subdivided in 15 subgroups
1070 (*Chiasmolithus*, *Coccolithus*, *Ellipsolithus*, *Discoaster*, *Ericsonia*, *Fasciculithus*, *Girgisia*, *Octolithus*,
1071 *Prinsius*, *Sphenolithus*, *Toweius*, *Rhomboaster/Tribrachiatus*, *Zyghrablithus*, reworking, others). In
1072 order to avoid the closed-sum effect that derives from the use of percentage data, we apply a log
1073 transformation of raw data.

1074 **Table S4.** Dataset used to perform the non-metric multidimensional scaling (MDS) for calcareous
1075 nannofossil assemblages from the Cicogna section. Calcareous nannofossils are subdivided in 15
1076 subgroups (*Chiasmolithus*, *Coccolithus*, *Ellipsolithus*, *Discoaster*, *Ericsonia*, *Fasciculithus*, *Girgisia*,
1077 *Octolithus*, *Prinsius*, *Sphenolithus*, *Toweius*, *Rhomboaster/Tribrachiatus*, *Zyghrablithus*, reworking,
1078 others). A square root transformation was used to minimize the influence of dominant taxa on the
1079 ordination.

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