

1 **Title: Positive feedback of elevated CO<sub>2</sub> on soil respiration in late autumn and**  
2 **winter**

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23 **Abstract**

24 Soil respiration of terrestrial ecosystems, a major component in the global carbon cycle is  
25 affected by elevated atmospheric CO<sub>2</sub> concentrations. However, seasonal differences of  
26 feedback effects of elevated CO<sub>2</sub> have rarely been studied. At the Giessen Free-Air CO<sub>2</sub>  
27 Enrichment (GiFACE) site, the effects of +20 % above ambient CO<sub>2</sub> concentration have been  
28 investigated since 1998 in a temperate grassland ecosystem. We defined five distinct annual  
29 seasons, with respect to management practices and phenological cycles. For a period of three  
30 years (2008-2010), weekly measurements of soil respiration were carried out with a survey  
31 chamber on vegetation-free subplots. The results revealed a pronounced and repeated increase  
32 of soil respiration under elevated CO<sub>2</sub> during late autumn and winter dormancy. Increased  
33 CO<sub>2</sub> losses during the autumn season (September-October) were 15.7 % higher and during the  
34 winter season (November – March) were 17.4 % higher compared to respiration from ambient  
35 CO<sub>2</sub> plots.

36 However, during spring time and summer, which are characterized by strong above- and  
37 below-ground plant growth, no significant change in soil respiration was observed at the  
38 FACE site under elevated CO<sub>2</sub>. This suggests (i) that soil respiration measurements, carried  
39 out only during the growing season under elevated CO<sub>2</sub> may underestimate the true soil-  
40 respiratory CO<sub>2</sub> loss (i.e. overestimate the C sequestered) and (ii) that additional C assimilated  
41 by plants during the growing season and transferred below-ground will quickly be lost via  
42 enhanced heterotrophic respiration outside the main growing season.

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46 **1 Introduction**

47 The atmospheric concentration of CO<sub>2</sub> has increased from pre-industrial values of 275 - 285  
48 ppm (Raynaud and Barnola, 1985) to 400 ppm in 2013 (Monastersky, 2013). Projections of  
49 future atmospheric CO<sub>2</sub> concentration in the year 2100 range between 490 and 1370 ppm  
50 depending on representative concentration pathways (Moss et al., 2010). As the major  
51 radiative forcing component (IPCC, 2013), atmospheric CO<sub>2</sub> is positively correlated with air  
52 temperature and is therefore an important component for global warming. Additionally,  
53 indirect effects of elevated atmospheric CO<sub>2</sub> (*e*CO<sub>2</sub>), which are altering carbon (C) fluxes in  
54 ecosystems, may impose a feedback to climate change. About half of photosynthetically  
55 assimilated C returns immediately to the atmosphere as plant-respired CO<sub>2</sub> (autotrophic  
56 respiration) (Chapin et al., 2002). Portions of the net carbon gain (net primary production) are  
57 transferred to the soil via root exudates, fine root growth and -turnover or other litter,  
58 providing the substrate for soil organic carbon (SOC) buildup (Kirschbaum, 2000).

59 Soil functions as an important C reservoir within the global carbon cycle and stores about  
60 1500 Gt of C (Amundson, 2001;Lal, 2004;Batjes, 1996), which is about twice the amount of  
61 C in the atmosphere (Schils et al., 2008).

62 Soil respiration, the sum of autotrophic root respiration and heterotrophic respiration from  
63 microorganisms and soil meso- and macrofauna, accounts for two thirds of the total C loss from  
64 terrestrial ecosystems (Luo, 2006). Enhanced net C losses under *e*CO<sub>2</sub> cause a positive feedback.

65 Many past studies focused on soil-atmosphere CO<sub>2</sub> exchange during the growing season.

66 However, soil respiration during vegetation dormancy may represent a significant component  
67 of the annual C budget and contributes to the observed winter CO<sub>2</sub> maximum in the  
68 atmosphere (Raich and Potter, 1995). Accordingly, analysis of CO<sub>2</sub> data from an air sampling  
69 network identified seasonal oscillation with highest concentrations occurring each winter

70 when respiration exceeds photosynthesis (Keeling et al., 1996). This emphasizes the necessity  
71 to study seasonal dynamics of soil respiration under future CO<sub>2</sub> conditions to gain a better  
72 understanding of how soil respiration responds to changing atmospheric CO<sub>2</sub> concentrations.

73 A meta-analysis of Zak et al. (2000) revealed a 51 % increase of soil respiration as a mean  
74 response in a grassland ecosystem under elevated CO<sub>2</sub>, Janssens & Ceulemans (2000) provided  
75 evidence for consistent stimulation of soil respiration under a variety of tree species. However, the  
76 majority of studies, to date, are based on short-term exposure (less than five years) with *e*CO<sub>2</sub>,  
77 often using open-top chamber experiments (Zak et al., 2000). Results from these experiments  
78 should be analyzed with appropriate caution because of the known “chamber effect” on the  
79 microclimate (Leadley and Drake, 1993) and their relevance to natural ecosystems in which  
80 longer-term biogeochemical feedbacks operate (Rastetter et al., 1991). Since soil respiration is a  
81 product of several rhizospheric processes i.e. root exudation, root respiration, and root turnover, as  
82 well as decomposition of litter and bulk soil organic matter from various pools with different  
83 characteristic turnover times, short- and long-term responses to *e*CO<sub>2</sub> may be quite different (Luo  
84 et al., 2001).

85 The most suitable approach for conducting ecosystem CO<sub>2</sub> experiments under natural conditions  
86 are FACE experiments, where intact ecosystems are exposed in-situ to a higher atmospheric CO<sub>2</sub>  
87 concentration. However, it has been reported that the sudden increase in atmospheric CO<sub>2</sub> (CO<sub>2</sub>  
88 step increase) at the beginning of a CO<sub>2</sub>-enrichment, may cause certain short-term responses of  
89 the ecosystem that differ from long-term responses (Luo, 2001; Newton et al., 2001). Accordingly,  
90 Kammann et al. (2005) showed that yield responses to *e*CO<sub>2</sub>, in the Giessen Free-Air CO<sub>2</sub>  
91 Enrichment (GiFACE), were different in the initial compared to the subsequent years. Moreover,  
92 plants may undergo micro-evolutionary changes in response to *e*CO<sub>2</sub> (Ward and Kelly, 2004),  
93 which may also be reflected in belowground processes (Klironomos et al., 2005).  
94 Consequently, to avoid misinterpretations due to insufficient experimental durations, results  
95 from long-term exposure studies are required. In the GiFACE this was after approximately 5-

96 6 years (Kammann et al., 2005). In the following we use the expression “short-term” for CO<sub>2</sub>  
97 enrichment durations <5 years and “long-term” for durations >5 years.

98 Based on a literature overview, we found 13 other FACE studies, from a wide variety of  
99 ecosystems, where in-situ soil respiration under *e*CO<sub>2</sub> has been investigated. All of these  
100 FACE studies operated at higher CO<sub>2</sub> enrichment concentrations than the GiFACE  
101 experiment (with +20 % CO<sub>2</sub> above ambient), i.e. they imposed larger initial step increases  
102 (Klironomos et al., 2005). Klironomos et al.(2005) have demonstrated that ecosystem responses  
103 to *e*CO<sub>2</sub> may differ between using a sudden step increase and a gradual rise in the CO<sub>2</sub>  
104 concentration. However, in any CO<sub>2</sub> enrichment study a step increase – also if lower than usual –  
105 cannot be avoided. Thus, experimental FACE results are more indicative for future predictions.  
106 However; experimental studies with duration of > 10 years are scarce (Carol Adair et al.,  
107 2011;Jackson et al., 2009). To our knowledge, 10 of the 16 investigations on soil respiration  
108 across these 13 FACE studies were carried out within the first five years of exposure, thus  
109 reporting short-term responses (Craine et al., 2001;King et al., 2001;Allen et al., 2000;Andrews  
110 and Schlesinger, 2001;Selsted et al., 2012;Masyagina and Koike, 2012;Soe et al.,  
111 2004;Lagomarsino et al., 2013;Liu et al., 2006;Nakayama et al., 1994). All short-term study  
112 results pointed towards a consistent stimulatory effect of *e*CO<sub>2</sub> on soil respiration. The average  
113 increase ranged from 12 % under a sweetgum plantation (King et al., 2004) to 70 % under a mixed  
114 plantation of *Populus* species (Lagomarsino et al., 2013). In two of the short-term studies,  
115 significant effects were only observed on days with high photosynthetic activity (Masyagina and  
116 Koike, 2012;Soe et al., 2004); measurements during dormancy were not carried out.

117 Three of the short-term studies conducted measurements during winter dormancy with contrasting  
118 results (Allen et al., 2000;Andrews and Schlesinger, 2001;Selsted et al., 2012;Lagomarsino et al.,  
119 2013). In a temperate heathland (CLIMAITE study), soil respiration was significantly increased  
120 under *e*CO<sub>2</sub> during three consecutive winter seasons (Selsted et al., 2012). Allen et al. (2000)  
121 detected a significant effect of *e*CO<sub>2</sub> on soil respiration during December 1997 in the Duke Forest

122 FACE study but not during the previous growing season beneath the loblolly pine forest. Andrews  
123 and Schlesinger (2001) reported from the same site greater increases of soil respiration during  
124 fumigation periods (26-59 %) than during non-fumigated periods (8-15 %). Fumigation was  
125 stopped when ambient air temperature dropped below 5 °C for more than one hour. In line with  
126 these results, much larger percentage enhancements of the soil CO<sub>2</sub> efflux were observed during  
127 the growing season (up to 111 %) than during dormant season (40 %) from a mixed plantation of  
128 *Populus* species exposed to eCO<sub>2</sub> (EuroFACE) (Lagomarsino et al., 2013). CO<sub>2</sub> enrichment was  
129 provided from bud burst to leaf fall at this site.

130 Out of six long-term studies on soil respiration (Carol Adair et al., 2011; Pregitzer et al.,  
131 2008; Jackson et al., 2009; Pendall et al., 2001; Bader and Körner, 2010; Dawes et al., 2013), only  
132 one study reported measurements throughout the dormant season, showing that after 10 years of  
133 eCO<sub>2</sub> during the growing season at a loblolly pine forest (Duke FACE) soil respiration was  
134 consistently higher in midsummer to early fall and diminished or disappeared in winter (Jackson  
135 et al., 2009). This was explained by a reduction in assimilation and hence available root exudate  
136 during dormancy. If the fumigation may continue during the dormant season in an ecosystem with  
137 a green canopy e.g. in a permanent grassland, the stimulation may theoretically continue on a  
138 higher level.

139 Reports from other long-term FACE studies in temperate ecosystems (disregarding the dormant  
140 season) were consistent by reporting an increase in soil respiration under eCO<sub>2</sub>, with the exception  
141 of the Swiss Canopy Crane experiment in an old-growth, mixed deciduous forest. Bader & Körner  
142 (2010) reported that soil respiration from the site was only stimulated when volumetric water  
143 content was ≤ 40 % at soil temperatures above 15 °C.

144 In summary, only fragmented information is available on how soil respiration responds to eCO<sub>2</sub>  
145 during vegetation as well as dormant periods after long-term eCO<sub>2</sub>. To our knowledge, no long-  
146 term FACE study in a grassland ecosystem exists which has investigated soil CO<sub>2</sub> fluxes across

147 several years. Consequently, it is difficult to generalize temporal patterns of soil respiration under  
148  $e\text{CO}_2$ , and thus the soil respiratory response to  $e\text{CO}_2$  at all.

149 Based on the available studies and earlier observations at our site, where whole-ecosystem  
150 respiration including the green canopy was increased under  $e\text{CO}_2$ , mainly during non-growing  
151 season (Lenhart, 2008), we hypothesized that (i) long-term (>10 years) moderate  $\text{CO}_2$   
152 enrichment causes increased soil respiration, (ii) soil respiration is more enhanced in the  
153 growing season than during vegetation dormancy (*winter*) and (iii) soil respiration is  
154 significantly enhanced in *winter* under  $e\text{CO}_2$  in the GiFACE where the  $\text{CO}_2$  enrichment is  
155 continuing during *winter*.

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171 **2 Materials and methods**

172 2.1 Study site and design

173 The Giessen Free Air Carbon Enrichment (GiFACE) experiment is located on permanent  
174 semi-natural grassland. It is situated near Giessen, Germany (50°32'N and 8°41.3'E) at an  
175 elevation of 172 m above sea level.

176 The set-up and performance of the GiFACE system has been described in detail by Jäger *et al.*  
177 (2003). In brief, from May 1998 until present, atmospheric CO<sub>2</sub> concentrations were enriched  
178 by 20 % above ambient, all-year-round during daylight hours. At present the GiFACE  
179 experiment is still ongoing.

180 The CO<sub>2</sub> enrichment was applied in three rings, each eight meter in diameter (E plots). Three  
181 equally sized control plots were maintained at ambient atmospheric CO<sub>2</sub> levels (A plots). The  
182 experimental design was a randomized block design. A block consisted of two plots to which  
183 ambient and eCO<sub>2</sub> treatments were randomly assigned. A characteristic attribute of the study  
184 site is a soil moisture gradient, resulting from a gradual terrain slope (2-3°) and varying depths  
185 of a subsoil clay layer. Within each of the three blocks, soil moisture conditions were  
186 relatively homogeneous (Jäger *et al.*, 2003).

187 The vegetation is an Arrhenatheretum elatioris Br.Bl. Filipendula ulmaria subcommunity,  
188 dominated by *Arrhenaterum elatium*, *Galium mollugo* and *Geranium pratense*. At least 12  
189 grass species, 15 non-leguminous herbs and 2 legumes are present within a single ring. For at  
190 least 100 years, the grassland has not been ploughed. Since several decades, it was managed  
191 as a hay meadow with two cuts per year, and fertilized in mid-April with granular mineral



192 calcium-ammonium-nitrate fertilizer at the rate of 40 kg N ha<sup>-1</sup> yr<sup>-1</sup>. Before 1996, fertilizer  
193 was applied at a rate of 50–100 kg N ha<sup>-1</sup> yr<sup>-1</sup> (Kammann et al., 2008).

194 The soil of the study site is classified as a Fluvisol (FAO classification) with a texture  
195 of sandy clay loam over a clay layer (Jäger et al., 2003).

196 Observations in this study were carried out from January 2008 - December 2010 (i.e. more  
197 than 9 years after the onset of CO<sub>2</sub> enrichment). During the observation period the mean  
198 annual temperature was 9.2 °C and mean annual precipitation was 562 mm which was  
199 identical to the average rainfall since the beginning of recording in 1995. Rainfall was  
200 recorded at the site in 30-min intervals with 20 randomly distributed “Hellmann” samplers.  
201 Air temperature was recorded continuously at two locations at the site in 2 m height and  
202 averaged 9.5 °C since 1995.

## 203 2.2 Measurement of soil CO<sub>2</sub> fluxes at the field site

204 In each of the six FACE plots, soil respiration rates were measured using an automated closed  
205 dynamic chamber system with an infrared gas analyzer (LI-COR 8100, LI-COR, Inc.,  
206 Lincoln, Nebraska, USA) with a patented vent for pressure equilibration between the closed  
207 chamber and the atmosphere (McDermitt et al., 2005). Carbon dioxide fluxes were reported in  
208 μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>. The measurements were performed at four permanently installed PVC soil  
209 collars per FACE ring, to cover the spatial heterogeneity within each ring. The soil collars had  
210 a diameter of 20.3 cm (8 inch) and were about 11 cm high. A beveled edge at one end  
211 facilitated the insertion into the soil, which took place on 9<sup>th</sup> May 2006 and the vegetation  
212 cover, including surficial rhizomes, was removed manually. Subsequently, the surface was  
213 held vegetation-free by removing germinated seedlings weekly. Due to uneven soil  
214 conditions, soil collars varied +/- 1 cm in their insertion depth. Generally, the insertion was  
215 chosen to be as shallow as possible, minimizing the trenching effect (Heinemeyer et al., 2011)

216 while maintaining an airtight connection between soil and chamber. A foam gasket and rubber  
217 seal between the bottom of the chamber and the top of the soil collar minimized leaks between  
218 the collar and the chamber. Before each measurement, the distance between the soil surface  
219 and the top of each soil collar (i.e. chamber offset) was measured and entered into the LICOR-  
220 software to enable correct flux calculations (= total chamber volume). After installation in  
221 May 2006, soil CO<sub>2</sub> efflux measurements were carried out over a period of one month to  
222 record the insertion and disturbance effects (Fig. S1). The investigation period spanned over  
223 three years (January 2008 until December 2010), after the collars were well established and  
224 held vegetation free for 1.5 years, allowing a die-back and decomposition of trenched roots,  
225 and in-growth of new roots from the outside vegetation. This ensured that soil respiration  
226 measurements in a dense, closed grassland canopy were taken as unbiased as possible.  
227 Measurements of soil respiration were carried out weekly in the evening, except in July 2009.  
228 From May to July 2010 and from October to December 2010, measurements were carried out  
229 every second week. No measurements were carried out in November and December 2008.  
230 During the measurement, a pump provided circulating air flow from the closed chamber on its  
231 collar to the infrared gas analyzer for thorough mixing of the systems' inner volume. Chamber  
232 closure time was between 1 and 3 min., depending on the season (i.e. the strength of the CO<sub>2</sub>  
233 efflux and thus the detection limit). CO<sub>2</sub> and H<sub>2</sub>O concentrations were measured  
234 simultaneously. The software calculated soil respiration rates by using the changes in CO<sub>2</sub>  
235 concentration over a period of time, taking the dilution of water vapor into account. Rates  
236 were calculated either by linear regression (lin\_flux) or as the efflux rate at time t<sub>0</sub> at chamber  
237 closure using an exponential CO<sub>2</sub> efflux function (exp\_flux) (LI-COR, 2007). The latter takes  
238 the diminishing CO<sub>2</sub> concentration gradient between the soil and the chamber headspace into  
239 account (Hutchinson and Mosier, 1981) and is implemented by LI-COR in the LI-8100 to  
240 avoid underestimations of the CO<sub>2</sub> efflux. We used the following algorithm to choose between

241 these two types of flux calculation for the subsequent processing of all obtained flux data. The  
242 use of the exp\_flux calculation was only allowed when (1) the  $R^2$  of the exp\_flux calculation  
243 was better than that of the lin\_flux calculation, and (2) when the number of iterations  
244 necessary for the exp\_flux calculation was lower than 5. By applying these comparatively  
245 strict criteria (stricter than those that are inbuilt by the manufacturer) we minimized  
246 miscalculations caused either by large initial  $CO_2$  concentration fluctuations at chamber  
247 closure (when the exp\_flux calculation is used) or underestimations of the true soil  $CO_2$  efflux  
248 (when only the lin\_flux calculation is used). The algorithm was applied to each measurement  
249 with the same settings. In general,  $CO_2$  flux rates with an  $R^2$  below 0.90 were excluded. This  
250 was the case in 0.6 % of all measurements taken in this study throughout the three year  
251 investigation period.

252 Soil moisture was measured in each FACE plot as the volumetric water content (VWC) with  
253 time-domain-reflectometric (TDR) probes (Imko, Ettlingen, Germany, type P2G). The probes  
254 were permanently installed (in March 1998) within the top 15 cm. The probes were monitored  
255 manually once a day, except on weekends or holidays. Soil temperature was logged in every  
256 plot at 10 cm depth as 15-min means (Imko, Ettlingen, Germany, Pt-100 sensors).

### 257 2.3 Data analyses

258 In order to describe changes in soil respiration during different seasons and to test for  
259 differences in soil respiration between ambient and elevated  $CO_2$ , we performed a linear  
260 mixed-effect model analysis with SPSS version 18. We used all measured data of three years  
261 for the linear mixed-effect model analysis to obtain seasonal estimates of soil respiration.  $CO_2$   
262 treatment was considered as a fixed effect in the model. Coding variables were introduced to  
263 indicate the hierarchical order of the data. The six mean fluxes taken in one measurement  
264 cycle received the same numerical code; this variable (“measurement cycle”) was considered

265 as a random effect in the linear mixed effect model. A further variable (“ringreplicate”) was  
266 introduced to define the ring where the measurement was taken (1-6). ”Ringreplicate” was  
267 selected as a repeated measure in the SPSS software using linear mixed effect model analysis.  
268 Maximum likelihood was used as the estimation method for the parameters in the model. The  
269 total observational data set was split by season to analyze seasonal CO<sub>2</sub>-response patterns.  
270 Therefore, we distinguished the following five seasons (1 – 5), depending on major dates of  
271 phenology and management practices at the grassland study site (Fig. 1): **1** = *winter*  
272 (November – March); **2** = *start of vegetation period* up to the date of spring fertilizer  
273 application (March – middle of April); **3** = *spring* until first biomass harvest (middle of April  
274 – end of May); **4** = regrowth and *summer* growing season (end of May – beginning of  
275 September); **5** = regrowth and *autumn* growing season (beginning of September – end of  
276 October).

277 The start of the vegetation period for the grassland ecosystem was identified according to the  
278 calculations defined by Wasshausen (1987). The date of leaf discoloration of *Quercus robur*  
279 in the nearby phenological garden was used to identify the beginning of winter dormancy. All  
280 other dates were chosen according to the management practices at the study site (Fig. 1); the  
281 exact dates varied by a few days between the years.

## 282 2.4 Soil respiration model

283 We applied a temperature response model to fill gaps in the measured data set. Therefore a  
284 function was fitted according to Lloyd & Taylor (1994) (Eq. 1) to 20 % of the data that were  
285 randomly selected. We defined values for coefficients E0 (= 62.16), T0 (= 262.47) and R10 (= 2.85)  
286 for the first run of the model. Subsequently, E0, T0 and R10 were fitted for each  
287 treatment (ambient and eCO<sub>2</sub>) by using the dynamic fit function in the SigmaPlot 11.0

288 software package (Systat Software, San Jose, CA, 2008). Mean soil temperature values were  
289 converted from °C to K.

$$290 \quad f = R10e^{E0\left(\frac{1}{(283.15-T0)} - \frac{1}{(x-T0)}\right)} \quad \text{Eq. ( 1)}$$

291 with  $E0$  = activation-energy-type empirical coefficient

292  $T0$  = lower temperature limit for soil respiration in K

293  $R10$  = respiration rate at 10 °C

294 Consequently, the quality of the soil respiration model was evaluated by plotting modelled  
295 soil respiration rates against the remaining 80% of the observed respiration values to test if the  
296 linear trend line meets the requested slope of 1 (Fig. 5).

## 297 2.5 Annual estimates of soil respiration

298 To obtain annual sums of soil respiration measured data was used whenever available and  
299 modelled data for data gaps. Modelled soil respiration rates were calculated, based on the  
300 almost continuous data set of soil temperature in 10 cm depth measured at 2-3 positions per  
301 ring. We received modelled fluxes for every 15 minutes over the three year period for all gaps  
302 where no observational data were available. Estimates of annual sums were then calculated  
303 with the observational data and the modelled data per ring and averaged between treatments  
304 as true steps (n=3). Differences in annual soil respiration between the CO<sub>2</sub> treatments were  
305 tested by using a paired t-test. Further, the absolute difference and relative change of monthly  
306 mean soil respiration rates under eCO<sub>2</sub> were calculated in comparison to soil respiration under  
307 ambient CO<sub>2</sub>, based on observational and modelled data . For calculating the relative change  
308 ambient soil respiration was set to 0 %.

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## 310 3 Results

### 311 3.1 Annual variability of soil respiration

312 From 2008 to 2010, soil respiration rates at the GiFACE experiment showed distinct annual  
313 dynamics, following the seasonal temperature cycle with lowest soil respiration effluxes  
314 during winter months and highest effluxes during mid-summer (Fig. 2c and 2g). Thus, soil  
315 respiration rates responded to abiotic factors in particular temperature and moisture. This is  
316 exemplified by the high CO<sub>2</sub> efflux rates in June 2009 which occurred shortly after a period of  
317 high precipitation while soil temperatures were > 20 °C (Fig. 2g).

318 The relative and absolute change of soil respiration under *e*CO<sub>2</sub> (Fig 2d and 2e) followed a  
319 seasonal pattern with greatest increases under *e*CO<sub>2</sub> during *autumn* and *winter*. During  
320 midsummer, when the largest absolute soil respiration rates occurred, the relative increase due  
321 to the CO<sub>2</sub> enrichment was lowest or non-existent. A linear mixed effect model analysis  
322 confirmed that soil respiration rates under *e*CO<sub>2</sub> were significantly higher compared to rates  
323 under ambient CO<sub>2</sub> during *autumn* (15.7 %) and *winter* (17.4 %) (Fig. 3). During all other  
324 seasons (*beginning of vegetation period* (season 2), *spring* (season 3) and *summer* (season 4)),  
325 covering most of the vegetation period, a trend towards higher soil respiration, but no  
326 significant CO<sub>2</sub> effect was observed with *e*CO<sub>2</sub> (Fig. 3).

### 327 3.2 Model performance and parameter estimation

328 By comparing modelled soil respiration with observed soil respiration for all observation  
329 dates from 2008 – 2010 a significant linear relationship was observed with a slope of 1.02  
330 (Fig. 5).

331 Based on the temperature-respiration function by Taylor & Lloyd (1994), soil respiration was  
332 significantly correlated to soil temperature under ambient as well as  $e\text{CO}_2$  ( $p = <0.0001$ ).  
333 From 2008 to 2010, 75 % of the variability of soil respiration rates was explained by soil  
334 temperature under ambient  $\text{CO}_2$  and 82 % under  $e\text{CO}_2$  (Fig. 4, Table 1). Soil respiration rates  
335 did not differ in their relationship to soil temperature between the treatments (Fig. 4).

### 336 3.3 Annual sums of soil respiration

337 Comparing annual sums of soil respiration, no mean treatment effect of elevated  $\text{CO}_2$  (over all  
338 seasons) was observed in any of the observation years (Table 2). Mean annual estimates of  
339 soil respiration under ambient  $\text{CO}_2$  ranged from 1283 to 1344  $\text{g C} [\text{CO}_2] \text{ m}^{-2} \text{ yr}^{-1}$  and under  
340  $e\text{CO}_2$  from 1300 to 1352  $\text{g C} [\text{CO}_2] \text{ m}^{-2} \text{ yr}^{-1}$  (Table 2).

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## 350 4 Discussion

### 351 4.1 Annual sums of soil respiration

352 In contrast to our initial hypotheses, annual estimates of soil respiration were not different  
353 between the CO<sub>2</sub> treatments (Table 2). Mean annual sums of soil respiration were  $1317 \pm 18$  g  
354 C m<sup>-2</sup> yr<sup>-1</sup> under ambient CO<sub>2</sub> and  $1331 \pm 16$  g C m<sup>-2</sup> yr<sup>-1</sup> under elevated CO<sub>2</sub>. Raich and  
355 Schlesinger (1992) estimated much lower rates of annual soil respiration, reporting 400 to 500  
356 g C m<sup>-2</sup> yr<sup>-1</sup> for temperate grasslands. Annual soil respiration sums from a sandstone and  
357 serpentine grassland were 485 and 346 g C m<sup>-2</sup> yr<sup>-1</sup> (Luo et al., 1996). These soil respiration  
358 rates were lower than those from the wet grassland site investigated here due to the larger net  
359 primary productivity of the wet temperate grassland with a year-round more or less moist  
360 climate, compared e.g. to a seasonally dry Mediterranean-type grassland. A lower net  
361 ecosystem productivity (NEP) will automatically result in lower overall soil respiratory C  
362 losses. Methodological differences may have been to a lesser extent been responsible, because  
363 the studies of Luo et al. (1996) and Raich and Schlesinger (1992) may have overestimated  
364 rather than underestimated the annual soil respiration. Their measurements did not exceed 2  
365 years in duration and soil respiration was less frequently measured for a portion of the year.  
366 Other recent studies reported higher rates of annual soil respiration which are closer to our  
367 estimates; however climatic factors are different from our site: In a tallgrass prairie of  
368 Oklahoma annual soil respiration rates were 1131 and 877 g C m<sup>-2</sup> yr<sup>-1</sup> in 2002 and 2003  
369 respectively (Zhou et al., 2006). In a Texas grassland annual soil respiration rates increased  
370 with annual precipitation and were 1600, 1300, 1200, 1000, 2100 and 1500 g C m<sup>-2</sup> yr<sup>-1</sup> in  
371 1993 through 1998 respectively (Mielnick and Dugas, 2000). At the Texas grassland site  
372 measurements were conducted year-round with a high time resolution. Consequently annual  
373 rates could be estimated by more measured (than gap-filled) data compared to other studies.



374 However the most important factors were likely the annual precipitation, its distribution over  
375 the year, and the annual mean temperature: High annual rainfall, a long growing season and  
376 large soil organic C contents explained the higher soil respiration rates (as a consequence of a  
377 higher NEP) at the Texas study site. Mean annual precipitation at the GiFACE study site (562  
378 mm) was close to the mean precipitation reached in 1995 at the Texas grassland with 657 mm,  
379 when annual soil respiration averaged  $1200 \text{ g C m}^{-2} \text{ yr}^{-1}$  at the Texas grassland.

#### 380 4.2 Seasonality of soil respiration

381 Also, contrary to our initial hypotheses is the observation that soil respiration was not  
382 significantly affected during the growing season (*start of vegetation period, spring and*  
383 *summer*) by the moderate long-term  $\text{CO}_2$  enrichment. This indicates that any increase in the  
384 ecosystem respiration (Lenhart, 2008) during this season will not have been due to enhanced  
385 soil (root-derived) respiration but rather to increases in the respiration of the green canopy.

386 The majority of long-term FACE studies reported significantly increased soil respiration  
387 under  $e\text{CO}_2$  during the growing season (Pregitzer et al., 2008; Jackson et al., 2009; Pendall et  
388 al., 2001; Dawes et al., 2013; Carol Adair et al., 2011), whereas Bader & Körner (2010)  
389 reported that seven years of  $e\text{CO}_2$  failed to stimulate cumulative soil respiration significantly  
390 during the growing season. Among the mentioned long-term FACE experiments, the GiFACE  
391 operates at the lowest  $\text{CO}_2$  enrichment step increase (20 % above ambient  $\text{CO}_2$ ), which may  
392 have contributed to this result.

393 However, in line with our hypotheses, the results revealed that 10 years of moderate  $\text{CO}_2$   
394 enrichment increased soil respiration during *winter* and *autumn* (Fig. 3). These seasonal  
395 stimulations of soil respiration under  $e\text{CO}_2$  were not observed by comparing the annual sums  
396 of soil respiration (Table 2). This may be because soil respiration fluxes were lower in *winter*

397 and *autumn* compared to fluxes from the other seasons where no differences in soil respiration  
398 between the CO<sub>2</sub> treatments were observed. However, within the *winter* and *autumn* season  
399 differences in soil respiration may play an important role concerning the global C balance.  
400 Increased rates of winter soil respiration under *e*CO<sub>2</sub> may increase the observed winter CO<sub>2</sub>  
401 maximum in the atmosphere (Raich and Potter, 1995; Keeling et al., 1996) when respiration  
402 exceeds photosynthesis. Another reason why annual sums of soil respiration were not  
403 different between the CO<sub>2</sub> treatments may be that our model underestimated high soil  
404 respiration fluxes (>10 μmol m<sup>-2</sup> s<sup>-1</sup>). However these fluxes occurred only in 1.72 % of all  
405 observations. Our model did not take soil moisture into account. The high variability of  
406 observed soil respiration during summer may be partly due to differing soil moisture  
407 conditions, which were not significantly different between ambient and *e*CO<sub>2</sub> plots  
408 (Kammann et al., 2005;2008).

409 In most FACE studies which reported the effect of *e*CO<sub>2</sub> on soil respiration, the winter was  
410 excluded since fumigation during this period was mostly switched off (often in response to  
411 sub-zero freezing temperatures or deciduous forest ecosystems). This was the case in the  
412 Swiss FACE study, where seeded grassland was exposed to 600 ppm CO<sub>2</sub> (de Graaff et al.,  
413 2004), the BioCON FACE, also a grassland study (Craine et al., 2001; Carol Adair et al.,  
414 2011), the Aspen FACE, an aspen forest enriched with *e*CO<sub>2</sub> (Pregitzer et al., 2008; King et  
415 al., 2001), a Japanese model forest ecosystem exposed to 550 ppm CO<sub>2</sub> (Masyagina and  
416 Koike, 2012) and in a 9-year FACE study of an alpine treeline ecosystem (Dawes et al.,  
417 2013). In the Swiss Canopy Crane study soil respiration was measured during the beginning  
418 of the dormant season but not over the complete dormant season while fumigation was  
419 switched off (Bader and Körner, 2010). In the Maricopa FACE, where a wheat field was  
420 exposed to *e*CO<sub>2</sub>, no winter measurements were carried out because this season was a fallow

421 season (Pendall et al., 2001). Outside the cultivation period no soil respiration measurements  
422 were made on a cotton plantation exposed to  $e\text{CO}_2$  (Nakayama et al., 1994).

423 Increased winter soil  $\text{CO}_2$  fluxes are in line with results from Selsted et al.(2012), who  
424 reported stimulated rates during three consecutive winter periods in a Danish N-limited  
425 *Calluna-Deschampsia*-heathland exposed to FACE at 510 ppm (CLIMAITE study).  
426 Fumigation was carried out all year-round except during periods with full snow cover.  
427 Contrary to our results, in the CLIMAITE study, the stimulatory effect of  $e\text{CO}_2$  on soil  
428 respiration persisted throughout most of the year, i.e. also in summer and not only during  
429 winter. However, in the CLIMAITE study, monthly soil respiration measurements were  
430 carried out within the first three years after the experimental start and may therefore reflect  
431 short-term responses, driven by the initial  $\text{CO}_2$  step increase (Klironomos et al., 2005). Thus  
432 the results are not completely comparable to this study where measurements were carried out  
433 in the 11<sup>th</sup> – 13<sup>th</sup> year of  $\text{CO}_2$  enrichment.

434 To our knowledge, the Duke Forest FACE is the only other FACE experiment where soil  
435 respiration was measured in an evergreen ecosystem year-round for several years and after  
436 long-term fumigation with  $e\text{CO}_2$  (+200 ppm). On average, soil respiration was significantly  
437 higher by 23 % under  $e\text{CO}_2$ . Jackson et al. (2009) summarized, after 10 years of  $\text{CO}_2$   
438 enrichment, that the greatest stimulation of soil respiration under  $e\text{CO}_2$  occurred from  
439 midsummer to early fall, in contrast to our observations, during winter the  $\text{CO}_2$  response of  
440 soil respiration was weakest. However, fumigation was stopped at the Duke Forest FACE  
441 when ambient air temperature dropped below 5°C for more than one hour.

442 After short-term enrichment with  $e\text{CO}_2$  (550 ppm) on a mixed plantation of *Populus* species  
443 (EuroFACE; in the 4<sup>th</sup> and 5<sup>th</sup> year of enrichment), Lagomarsino et al. (2013) recorded much

444 larger stimulation of soil respiration during the vegetation (up to 111 % enhancement) than  
445 dormant season (40 % enhancement), when fumigation was stopped, which is also contrary to  
446 our results. However, experimental setup and climate differed from our site. While minimum  
447 soil temperatures reached -1.7 °C in the GiFACE experiment during winter (Fig. 2b),  
448 comparably warm and mild winters without sub-zero temperatures were typical at the  
449 EUROFACE site located in Italy. Moreover, the *Populus* plantation was a fertilized agro-  
450 ecosystem, where coppicing was carried out every three years, while the GiFACE was an old  
451 established, species-rich ecosystem where N-supply was limited.

452 In line with results from the EuroFACE but in contrast to our findings, Volk & Niklaus (2002)  
453 did not observe any wintertime increase in the ecosystem CO<sub>2</sub> efflux from a calcareous  
454 grassland in response to three years of CO<sub>2</sub> enrichment (600 ppm) with a screen-aided CO<sub>2</sub>  
455 enrichment facility.

456 Investigations from the GiFACE experiment showed that N<sub>2</sub>O emissions also exhibited a  
457 “seasonality response”, with the greatest stimulation of N<sub>2</sub>O emission under *e*CO<sub>2</sub> being  
458 observed in late-summer and autumn (Kammann et al., 2008). These findings support the  
459 hypothesis that the driving mechanism of the *e*CO<sub>2</sub> seasonality responses of enhanced  
460 microbial activity may have been related to the mineralization of previously accumulated  
461 organic matter, fuelling denitrification (Kammann et al., 2008).

#### 462 4.3 Root derived soil respiration

463 Increased root biomass was frequently recorded under *e*CO<sub>2</sub> (Rogers et al., 1994; Jastrow et  
464 al., 2000; Lukac et al., 2009), potentially affecting soil respiration rates (Zak et al., 2000).  
465 However, at the GiFACE, root biomass, picked with forceps (for set time intervals per  
466 sample, n=3 per FACE ring), was only different in December 2005 between the CO<sub>2</sub>

467 treatments but not at other dates during 2004 – 2007 (Lenhart, 2008) or in November 2011  
468 (unpublished results). Lenhart (2008) observed in the GiFACE  $e\text{CO}_2$  plots, using Keeling  
469 plots and two-component mixing models that the fraction of root-derived  $\text{CO}_2$  (root- and root-  
470 exudate respiration and fine root decay), as part of the total soil  $\text{CO}_2$  efflux was lower in  
471 winter than during the growing season. Accordingly, during *winter*, the soil  $\text{CO}_2$  efflux  
472 originated mainly from microbial soil respiration.

473 Higher fine root turnover under  $e\text{CO}_2$ , resulting in higher C input via root necromass could  
474 explain increased *autumn* soil respiration but unlikely the *winter* increase in soil  $\text{CO}_2$  efflux at  
475 the GiFACE since root necromass was not changed under  $e\text{CO}_2$  in November 2011  
476 (unpublished results). Alternatively, differences in the root necromass could already have  
477 been decomposed at this time of sampling or may be observed later in the year, so that  
478 “enhanced fine root decomposition” as cause of the *autumn* and *winter* soil respiration  
479 increase under  $e\text{CO}_2$  cannot be ruled out.

#### 480 4.5 N availability

481 Since soil microorganisms require C as well as N for maintenance and growth (De Graaff et  
482 al., 2006; Zak et al., 1993), N availability plays an important role in determining soil  $\text{CO}_2$   
483 efflux. Root respiration rates were observed to correlate with tissue nitrogen concentration  
484 (Burton et al., 1996, 1998). In the Giessen-FACE,  $e\text{CO}_2$  caused reduced tissue N  
485 concentrations and higher C:N-ratios of aboveground plant biomass (Kammann et al.,  
486 2008). Through freezing effects in winter, mineral N, which was immobilized into the  
487 microbial biomass shortly after fertilizer application in spring, became partly available again  
488 (Müller et al., 2003). It is possible that N, as a limiting factor in the temperate grassland, may

489 partly be responsible for the increase in soil C loss during the *autumn* and *winter* season under  
490  $e\text{CO}_2$ .

#### 491 4.6 Microbial community

492 Multiple observations from the GiFACE indicated that increases in winter soil respiration  
493 under  $e\text{CO}_2$  were largely associated with microbial respiration (including rhizosphere  
494 microbiota). Recent studies from other FACE sites detected differences between microbial  
495 communities at  $e\text{CO}_2$  compared to ambient  $\text{CO}_2$  (Drigo et al., 2008; Drigo et al., 2009). At the  
496 GiFACE, stimulated rhizosphere-C utilization by arbuscular mycorrhizal fungi were found  
497 under  $e\text{CO}_2$  by a  $^{13}\text{C}$ -PLFA study (Denef et al., 2007), which may have contributed to altered  
498 soil respiration. Recent measurements in 2013 did not indicate any differences in the  
499 abundance of bacteria and archaea between the ambient and  $e\text{CO}_2$  plots (K. Brenzinger,  
500 personal communication) so that this can be ruled out as a cause for differed soil respiration  
501 between the  $\text{CO}_2$  treatments if this observation persists throughout *autumn* and *winter*.

#### 502 4.7 Soil moisture

503 Several studies showed that  $e\text{CO}_2$  can affect soil moisture (Niklaus et al., 1998; Field et al.,  
504 1995; Hungate et al., 1997), which in turn regulates soil respiration. However, large effects are  
505 only expected and were detected at the dry end of the spectrum (Moyano et al., 2012; Guntinas  
506 et al., 2013; Rodrigo et al., 1997). During the investigation period, the volumetric water content  
507 ranged from 20 to 80 vol.% at the GiFACE site, with an average of 44% during 2008-2010,  
508 and 39% over the vegetation periods of these years. Thus, the soil moisture effect is likely not  
509 to be large. Moreover, no significant effect of  $e\text{CO}_2$  on the soil water content was observed  
510 either during the first 5 years of enrichment (Kammann et al., 2005) or after 13 years of

511 enrichment (Meine, 2013). Consequently, a CO<sub>2</sub>-induced soil moisture effect is unlikely  
512 governing increased soil respiration rates.

513 However, it can be assumed that annual dynamics of soil moisture with wettest conditions in  
514 winter, i.e. close to saturation, and driest conditions in summer (Fig. 2a) contributed to the  
515 seasonal dynamics of soil respiration under *e*CO<sub>2</sub> due to diffusion limitations. Previous results  
516 from the GiFACE site show that in periods when soil moisture in the main rooting zone was  
517 low (0.3 m<sup>3</sup> m<sup>-3</sup>), soil continued to produce N<sub>2</sub>O from deeper soil layers (20 – 50 cm), where  
518 soil moisture remained high (c. 0.6 m<sup>3</sup> m<sup>-3</sup>) (Müller et al., 2004). The production of N<sub>2</sub>O at  
519 deep soil layers seemed to coincide with the production of CO<sub>2</sub> during summer, which was  
520 also characterized by a homogenous δ <sup>13</sup>CO<sub>2</sub> profile during vegetation period at our study site  
521 (Lenhart, 2008). However, a detailed investigation on layer-specific CO<sub>2</sub> production was  
522 beyond the scope of this study. At times of high soil moisture CO<sub>2</sub> diffusion was slowed  
523 down, coinciding with limited oxygen supply (Skopp et al., 1990). At these times, soil  
524 respiration was likely originating to a major part from the topsoil. However, increased *autumn*  
525 soil respiration under *e*CO<sub>2</sub> cannot be attributed to this phenomenon since soil water content is  
526 relatively low at this season (Fig. 2a). We suggest that increased substrate supply under *e*CO<sub>2</sub>  
527 from end-of-season dieback of roots and enhanced root-associated microbiome activity may  
528 explain stimulated soil respiration rates in *autumn*.

#### 529 4.9 Plant community

530 Another aspect which may have contributed to altered soil respiration rates under *e*CO<sub>2</sub> is a  
531 shift in the plant community composition. Grütters et al. (2006) observed that summer-greens  
532 decreased, whereas evergreens increased under *e*CO<sub>2</sub> in the GiFACE experiment. Since soil  
533 respiration is controlled by substrate supply via rhizodeposition (Verburg et al., 2004; Wan

534 and Luo, 2003;Craine et al., 1999), higher photosynthetic activity in  $e\text{CO}_2$  plots during mild  
535 *winter* may have contributed to the observed increase in soil respiration. In addition, since the  
536 vegetative aboveground growth is dormant and does not provide an assimilate sink, the  
537 relative proportion of assimilate partitioned below-ground towards the root-associated micro-  
538 biota may increase, contributing to the relative increase under  $e\text{CO}_2$  during *winter*. The higher  
539 abundance of evergreens at  $e\text{CO}_2$  also underlines the importance of a year-round  $\text{CO}_2$   
540 enrichment strategy in such ecosystems with the respective climatic conditions. To date,  
541 increased winter soil respiration at  $e\text{CO}_2$  was only found in FACE experiments with year-  
542 round fumigation and a photosynthesizing at least partly green canopy, i.e. in the CLIMAITE  
543 study (Selsted et al., 2012) and in this study.

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## 554 **5 Conclusions**

555 In conclusion, our results demonstrate the importance of winter soil respiration measurements,  
556 by showing that soil respiration was increased during *autumn* and *winter* after moderate long-  
557 term *eCO*<sub>2</sub>. Measurements and year-round CO<sub>2</sub> enrichment should not be neglected, at least in  
558 winter-green temperate ecosystems. Studies in such ecosystems excluding measurements  
559 during the dormant season may thus underestimate the effect of *eCO*<sub>2</sub> on annual soil-  
560 respiratory CO<sub>2</sub> losses (i.e. leading to an overestimation of C sequestered). Consequently,  
561 winter soil CO<sub>2</sub> fluxes may play a crucial role in determining the carbon balance and  
562 dynamics of temperate grassland ecosystems. Our results indicate that temperate European  
563 grasslands which are characterized by a greenhouse gas balance near zero (Soussana et al.,  
564 2007) may gradually turn into greenhouse gas sources with rising atmospheric CO<sub>2</sub> due to  
565 enhanced CO<sub>2</sub> losses during *autumn* and *winter*, in particular if N<sub>2</sub>O emissions are  
566 significantly increased as well as observed in the GiFACE (Kammann et al., 2008;Regan et  
567 al., 2011).

568 To generalize and explain the variation in the temporal dynamics of soil respiration under  
569 *eCO*<sub>2</sub> more studies of winter C dynamics under long-term *eCO*<sub>2</sub> are required. For such future  
570 studies it is advisable to include frequent samplings of root biomass, including the fine root  
571 fraction and necromass, in particular during the *autumn/winter* period under *eCO*<sub>2</sub>. Another  
572 beneficial research strategy may be combined (pulse) labelling of <sup>15</sup>N and <sup>13</sup>C to elucidate  
573 gross C and N turnover processes after long-term (>10 years) of CO<sub>2</sub> enrichment to study the  
574 C-N gross dynamics and associated carbonaceous gas losses.

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600 **References**

- 601 Allen, A. S., Andrews, J. A., Finzi, A. C., Matamala, R., Richter, D. D., and Schlesinger, W.  
602 H.: Effects of free-air CO<sub>2</sub> enrichment (FACE) on belowground processes in a *Pinus taeda*  
603 forest, *Ecol. Appl.*, 10, 437-448, 10.2307/2641105, 2000.
- 604 Amundson, R.: The carbon budget in soils, *Annual Review of Earth and Planetary Sciences*,  
605 29, 535-562, 2001.
- 606 Andrews, J. A., and Schlesinger, W. H.: Soil CO<sub>2</sub> dynamics, acidification, and chemical  
607 weathering in a temperate forest with experimental CO<sub>2</sub> enrichment, *Global Biogeochem.*  
608 *Cycles*, 15, 149-162, 10.1029/2000gb001278, 2001.
- 609 Bader, M. K. F., and Körner, C.: No overall stimulation of soil respiration under mature  
610 deciduous forest trees after 7 years of CO<sub>2</sub> enrichment, *Global Change Biol.*, 16, 2830-2843,  
611 10.1111/j.1365-2486.2010.02159.x, 2010.
- 612 Batjes, N. H.: Total carbon and nitrogen in the soils of the world, *Eur. J. Soil Sci.*, 47, 151-  
613 163, 10.1111/j.1365-2389.1996.tb01386.x, 1996.
- 614 Burton, A. J., Pregitzer, K. S., Zogg, G. P., and Zak, D. R.: Latitudinal variation in sugar  
615 maple fine root respiration, *Can. J. For. Res.*, 26, 1761-1768, 10.1139/x26-200, 1996.
- 616 Burton, A. J., Pregitzer, K. S., Zogg, G. P., and Zak, D. R.: Drought reduces root respiration  
617 in sugar maple forests, *Ecol. Appl.*, 8, 771-778, 10.1890/1051-  
618 0761(1998)008[0771:drrris]2.0.co;2, 1998.
- 619 Carol Adair, E., Reich, P. B., Trost, J. J., and Hobbie, S. E.: Elevated CO<sub>2</sub> stimulates  
620 grassland soil respiration by increasing carbon inputs rather than by enhancing soil moisture,  
621 *Global Change Biol.*, 17, 3546-3563, 10.1111/j.1365-2486.2011.02484.x, 2011.
- 622 Chapin, F. S., III, Matson, P. A., and Mooney, H. A.: *Principles of terrestrial ecosystem*  
623 *ecology*, Springer, New York, 436 pp., 2002.
- 624 Craine, J. M., Wedin, D. A., and Chapin, F. S.: Predominance of ecophysiological controls on  
625 soil CO<sub>2</sub> flux in a Minnesota grassland, *Plant Soil*, 207, 77-86, 1999.
- 626 Craine, J. M., Wedin, D. A., and Reich, P. B.: The response of soil CO<sub>2</sub> flux to changes in  
627 atmospheric CO<sub>2</sub>, nitrogen supply and plant diversity, *Global Change Biol.*, 7, 947-953, 2001.
- 628 Dawes, M. A., Hagedorn, F., Handa, I. T., Streit, K., Ekblad, A., Rixen, C., Korner, C., and  
629 Hattenschwiler, S.: An alpine treeline in a carbon dioxide-rich world: synthesis of a nine-year  
630 free-air carbon dioxide enrichment study, *Oecologia*, 171, 623-637, 10.1007/s00442-012-  
631 2576-5, 2013.
- 632 De Graaff, M.-A., Van Groenigen, K.-J., Six, J., Hungate, B. A., and Van Kessel, C.:  
633 Interactions between plant growth and soil nutrient cycling under elevated CO<sub>2</sub>: a meta-  
634 analysis, *Global Change Biol.*, 12, 2077-2091, 2006.

635 de Graaff, M. A., Six, J., Harris, D., Blum, H., and van Kessel, C.: Decomposition of soil and  
636 plant carbon from pasture systems after 9 years of exposure to elevated CO<sub>2</sub>: impact on C  
637 cycling and modeling, *Global Change Biol.*, 10, 1922-1935, 10.1111/j.1365-  
638 2486.2004.00862.x, 2004.

639 Deneff, K., Bubenheim, H., Lenhart, K., Vermeulen, J., van Cleemput, O., Boeckx, P., and  
640 Müller, C.: Community shifts and carbon translocation within metabolically-active  
641 rhizosphere microorganisms in grasslands under elevated CO<sub>2</sub>, *Biogeosciences*, 4, 769-779,  
642 2007.

643 Drigo, B., Kowalchuk, G. A., and van Veen, J. A.: Climate change goes underground: effects  
644 of elevated atmospheric CO<sub>2</sub> on microbial community structure and activities in the  
645 rhizosphere, *Biol. Fertil. Soils*, 44, 667-679, 10.1007/s00374-008-0277-3, 2008.

646 Drigo, B., Van Veen, J. A., and Kowalchuk, G. A.: Specific rhizosphere bacterial and fungal  
647 groups respond differently to elevated atmospheric CO<sub>2</sub>, *Isme J.*, 3, 1204-1217,  
648 10.1038/ismej.2009.65, 2009.

649 Field, C. B., Jackson, R. B., and Mooney, H. A.: Stomatal response to increased CO<sub>2</sub>:  
650 implications from the plant to the global scale, *Plant Cell Environ.*, 18, 1214-1225, 1995.

651 Grüters, U., Janze, S., Kammann, C., and Jäger, H.-J.: Plant functional types and elevated  
652 CO<sub>2</sub>: a method of scanning for causes of community alteration, *Journal of Applied Botany  
653 and Food Quality*, 80, 116-128, 2006.

654 Guntinas, M. E., Gil-Sotres, F., Leiros, M. C., and Trasar-Cepeda, C.: Sensitivity of soil  
655 respiration to moisture and temperature, *J. Soil Sci. Plant Nutr.*, 13, 445-461, 10.4067/s0718-  
656 95162013005000035, 2013.

657 Heinemeyer, A., Di Bene, C., Lloyd, A. R., Tortorella, D., Baxter, R., Huntley, B.,  
658 Gelsomino, A., and Ineson, P.: Soil respiration: implications of the plant-soil continuum and  
659 respiration chamber collar-insertion depth on measurement and modelling of soil CO<sub>2</sub> efflux  
660 rates in three ecosystems, *Eur. J. Soil Sci.*, 62, 82-94, 10.1111/j.1365-2389.2010.01331.x,  
661 2011.

662 Hungate, B. A., Chapin, F. S., III, Zhong, H., Holland, E. A., and Field, C. B.: Stimulation of  
663 grassland nitrogen cycling under carbon dioxide enrichment, *Oecologia*, 109, 149-153, 1997.

664 Hutchinson, G. L., and Mosier, A. R.: Improved soil cover method for field measurement of  
665 nitrous oxide fluxes, *Soil Science Society of America Journal*, 45, 311-316, 1981.

666 Jackson, R. B., Cook, C. W., Phippen, J. S., and Palmer, S. M.: Increased belowground  
667 biomass and soil CO<sub>2</sub> fluxes after a decade of carbon dioxide enrichment in a warm-  
668 temperate forest, *Ecology*, 90, 3352-3366, 10.1890/08-1609.1, 2009.

669 Jäger, H.-J., Schmidt, S. W., Kammann, C., Grünhage, L., Müller, C., and Hanewald, K.: The  
670 University of Giessen Free-Air Carbon Dioxide Enrichment Study: Description of the  
671 experimental site and of a new enrichment system, *J. Appl. Bot.*, 77, 117-127, 2003.

- 672 Janssens, I. A., and Ceulemans, R.: The response of soil CO<sub>2</sub> efflux under trees grown in  
673 elevated atmospheric CO<sub>2</sub>: A literature review, *Phyton-Ann. REI Bot.*, 40, 97-101, 2000.
- 674 Jastrow, J. D., Miller, R. M., and Owensby, C. E.: Long-term effects of elevated atmospheric  
675 CO<sub>2</sub> on below-ground biomass and transformation to soil organic matter in grassland, *Plant*  
676 *Soil*, 224, 85-97, 2000.
- 677 Kammann, C., Grünhage, L., Grüters, U., Janze, S., and Jäger, H.-J.: Response of  
678 aboveground grassland biomass and soil moisture to moderate long-term CO<sub>2</sub> enrichment,  
679 *Basic and Applied Ecology*, 6, 351-365, 2005.
- 680 Kammann, C., Müller, C., Grünhage, L., and Jäger, H.-J.: Elevated CO<sub>2</sub> stimulates N<sub>2</sub>O  
681 emissions in permanent grassland, *Soil Biol. Biochem.*, 40, 2194-2205, 2008.
- 682 Keeling, C. D., Chin, J. F. S., and Whorf, T. P.: Increased activity of northern vegetation  
683 inferred from atmospheric CO<sub>2</sub> measurements, *Nature*, 382, 146-149, 1996.
- 684 King, J. S., Pregitzer, K. S., Zak, D. R., Sober, J., Isebrands, J. G., Dickson, R. E., Hendrey,  
685 G. R., and Karnosky, D. F.: Fine-root biomass and fluxes of soil carbon in young stands of  
686 paper birch and trembling aspen as affected by elevated atmospheric CO<sub>2</sub> and tropospheric  
687 O<sub>3</sub>, *Oecologia*, 128, 237-250, 2001.
- 688 King, J. S., Hanson, P. J., Bernhardt, E., DeAngelis, P., Norby, R. J., and Pregitzer, K. S.: A  
689 multiyear synthesis of soil respiration responses to elevated atmospheric CO<sub>2</sub> from four forest  
690 FACE experiments, *Global Change Biol.*, 10, 1027-1042, 10.1111/j.1529-8817.2003.00789.x,  
691 2004.
- 692 Kirschbaum, M. U. F.: Will changes in soil organic carbon act as a positive or negative  
693 feedback on global warming?, *Biogeochem.*, 48, 21-51, 10.1023/a:1006238902976, 2000.
- 694 Klironomos, J. N., Allen, M. F., Rillig, M. C., Piotrowski, J., Makvandi-Nejad, S., Wolfe, B.  
695 E., and Powell, J. R.: Abrupt rise in atmospheric CO<sub>2</sub> overestimates community response in a  
696 model-plant soil system, *Nature*, 433, 621-624, 2005.
- 697 Lagomarsino, A., Lukac, M., Godbold, D. L., Marinari, S., and De Angelis, P.: Drivers of  
698 increased soil respiration in a poplar coppice exposed to elevated CO<sub>2</sub>, *Plant Soil*, 362, 93-  
699 106, 10.1007/s11104-012-1261-0, 2013.
- 700 Lal, R.: Soil carbon sequestration impacts on global climate change and food security,  
701 *Science*, 304, 1623-1627, 2004.
- 702 Leadley, P. W., and Drake, B. G.: Open top chambers for exposing plant canopies to elevated  
703 CO<sub>2</sub> concentration and for measuring net gas-exchange *Vegetatio*, 104, 3-15,  
704 10.1007/bf00048141, 1993.
- 705 Lenhart, K.: The effects of long-term Free Air CO<sub>2</sub> Enrichment (FACE) on soil aggregation,  
706 soil carbon input, and ecosystem CO<sub>2</sub> dynamics in a temperate grassland ecosystem,  
707 Department of Plant Ecology, Justus-Liebig University, Giessen, 134 pp., 2008.

- 708 LI-COR: LI-8100 Instruction Manual, LI-8100 automated soil CO<sub>2</sub> flux system., Li-COR,  
709 Inc, Lincoln, NE, USA 68504, 2007.
- 710 Liu, Q., Edwards, N. T., Post, W. M., Gu, L., Ledford, J., and Lenhart, S.: Temperature-  
711 independent diel variation in soil respiration observed from a temperate deciduous forest,  
712 *Global Change Biol.*, 12, 2136-2145, 2006.
- 713 Lloyd, J., and Taylor, J. A.: On the temperature-dependence of soil respiration, *Funct. Ecol.*,  
714 8, 315-323, 10.2307/2389824, 1994.
- 715 Lukac, M., Lagomarsino, A., Moscatelli, M. C., De Angelis, P., Cotrufo, M. F., and Godbold,  
716 D. L.: Forest soil carbon cycle under elevated CO<sub>2</sub> – a case of increased throughput?,  
717 *Forestry*, 82, 75-86, 10.1093/forestry/cpn041, 2009.
- 718 Luo, Y., Jackson, R. B., Field, C. B., and Mooney, H. A.: Elevated CO<sub>2</sub> increases  
719 belowground respiration in California grasslands, *Oecologia*, 108, 130-137,  
720 10.1007/bf00333224, 1996.
- 721 Luo, Y.: Transient ecosystem responses to free-air CO<sub>2</sub> enrichment (FACE): experimental  
722 evidence and methods of analysis, *New Phytol.*, 152, 3-8, 2001.
- 723 Luo, Y., Wu, L., Andrews, J. A., White, L., Matamala, R., Schäfer, K. V. R., and Schlesinger,  
724 W. H.: Elevated CO<sub>2</sub> differentiates ecosystem carbon processes: deconvolution analysis of  
725 Duke forest data, *Ecol. Monogr.*, 71, 357-376, 2001.
- 726 Luo, Y. a. Z., Z: *Soil Respiration and the Environment*, Academic/Elsevier, San Diego,  
727 328 pp., 2006.
- 728 Masyagina, O. V., and Koike, T.: Soil Respiration in Model Plantations under Conditions of  
729 Elevated CO<sub>2</sub> in the Atmosphere (Hokkaido Island, Japan), *Russ. J. Ecol.*, 43, 24-28,  
730 10.1134/s1067413611060099, 2012.
- 731 McDermitt, D., Xu, L., Gracia, R., Madsen, R., and Anderson, D.: On equalizing pressure in a  
732 soil respiration chamber with pressure in the ambient air under windy conditions, *Geophysical*  
733 *Research Abstracts*, 7 05841, 2005.
- 734 Meine, M.: *Charakterisierung und Quantifizierung der mikrobiellen Bodenrespiration eines*  
735 *Grünlandbodens unter erhöhten atmosphärischen CO<sub>2</sub>-Konzentrationen.*, diploma,  
736 *Geography, Philipps-Universität Marburg, Marburg*, 101 pp., 2013.
- 737 Mielnick, P. C., and Dugas, W. A.: Soil CO<sub>2</sub> flux in a tallgrass prairie, *Soil Biology and*  
738 *Biochemistry*, 32, 221-228, [http://dx.doi.org/10.1016/S0038-0717\(99\)00150-9](http://dx.doi.org/10.1016/S0038-0717(99)00150-9), 2000.
- 739 Monastersky, R.: Global carbon dioxide levels near worrisome milestone, *Nature*, 497, 13-14,  
740 2013.
- 741 Moss, R. H., Edmonds, J. A., Hibbard, K. A., Manning, M. R., Rose, S. K., van Vuuren, D. P.,  
742 Carter, T. R., Emori, S., Kainuma, M., Kram, T., Meehl, G. A., Mitchell, J. F. B.,

- 743 Nakicenovic, N., Riahi, K., Smith, S. J., Stouffer, R. J., Thomson, A. M., Weyant, J. P., and  
 744 Wilbanks, T. J.: The next generation of scenarios for climate change research and assessment,  
 745 *Nature*, 463, 747-756,  
 746 [http://www.nature.com/nature/journal/v463/n7282/supinfo/nature08823\\_S1.html](http://www.nature.com/nature/journal/v463/n7282/supinfo/nature08823_S1.html), 2010.
- 747 Moyano, F. E., Vasilyeva, N., Bouckaert, L., Cook, F., Craine, J., Curiel Yuste, J., Don, A.,  
 748 Epron, D., Formanek, P., Franzluebbers, A., Ilstedt, U., Kätterer, T., Orchard, V., Reichstein,  
 749 M., Rey, A., Ruamps, L., Subke, J. A., Thomsen, I. K., and Chenu, C.: The moisture response  
 750 of soil heterotrophic respiration: interaction with soil properties, *Biogeosciences*, 9, 1173-  
 751 1182, 10.5194/bg-9-1173-2012, 2012.
- 752 Müller, C., Kammann, C., Ottow, J. C. G., and Jäger, H.-J.: Nitrous oxide emission from  
 753 frozen grassland soil and during thawing periods, *Z. Pflanzenern. Bodenk.*, 166, 46-53, 2003.
- 754 Müller, C., Stevens, R. J., Laughlin, R. J., and Jäger, H.-J.: Microbial processes and the site of  
 755 N<sub>2</sub>O production in a temperate grassland soil, *Soil Biol. Biochem.*, 36, 453-461, 2004.
- 756 Nakayama, F. S., Huluka, G., Kimball, B. A., Lewin, K. F., Nagy, J., and Hendrey, G. R.: Soil  
 757 carbon dioxide fluxes in natural and CO<sub>2</sub>-enriched systems, *Agric. For. Met.*, 70, 131-140,  
 758 10.1016/0168-1923(94)90052-3, 1994.
- 759 Newton, P. C. D., Clark, H., Edwards, G. R., and Ross, D. J.: Experimental confirmation of  
 760 ecosystem model predictions comparing transient and equilibrium plant responses to elevated  
 761 atmospheric CO<sub>2</sub>, *Ecol. Lett.*, 4, 344-347, 2001.
- 762 Niklaus, P. A., Spinnler, D., and Korner, C.: Soil moisture dynamics of calcareous grassland  
 763 under elevated CO<sub>2</sub>, *Oecologia*, 117, 201-208, 10.1007/s004420050649, 1998.
- 764 Pendall, E., Leavitt, S. W., Brookes, T., Kimball, B. A., Pinter, P. J., Jr, Wall, G. W.,  
 765 LaMorte, R. L., Wechsung, G., Wechsung, F., Adamsen, F., Matthias, A. D., and Thompson,  
 766 T. L.: Elevated CO<sub>2</sub> stimulates soil respiration in a FACE wheat field, *Bas. App. Ecol.*, 2,  
 767 193-201, 2001.
- 768 Pregitzer, K. S., Burton, A. J., King, J. S., and Zak, D. R.: Soil respiration, root biomass, and  
 769 root turnover following long-term exposure of northern forests to elevated atmospheric CO<sub>2</sub>  
 770 and tropospheric O<sub>3</sub>, *New Phytol.*, 180, 153-161, 10.1111/j.1469-8137.2008.02564.x, 2008.
- 771 Raich, J. W., and Schlesinger, W. H.: The global carbon dioxide flux in soil respiration and its  
 772 relationship to vegetation and climate, *Tellus*, 44B, 81-99, 1992.
- 773 Raich, J. W., and Potter, C. S.: Global patterns of carbon dioxide emissions from soils, *Global*  
 774 *Biogeochem. Cyc.*, 9, 23-36, 1995.
- 775 Rastetter, E. B., Ryan, M. G., Shaver, G. R., Melillo, J. M., Nadelhoffer, K. J., Hobbie, J. E.,  
 776 and Aber, J. D.: A general biogeochemical model describing the response of the C and N  
 777 cycles in terrestrial ecosystems to changes in CO<sub>2</sub>, climate, and N deposition, *Tree Phys.*, 9,  
 778 101-126, 1991.

- 779 Raynaud, D., and Barnola, J. M.: An Antarctic ice core reveals atmospheric CO<sub>2</sub> variations  
780 over the past few centuries, *Nature*, 315, 309-311, 1985.
- 781 Regan, K., Kammann, C., Hartung, K., Lenhart, K., Muller, C., Philippot, L., Kandeler, E.,  
782 and Marhan, S.: Can differences in microbial abundances help explain enhanced N<sub>2</sub>O  
783 emissions in a permanent grassland under elevated atmospheric CO<sub>2</sub>?, *Global Change Biol.*,  
784 17, 3176-3186, 10.1111/j.1365-2486.2011.02470.x, 2011.
- 785 Rodrigo, A., Recous, S., Neel, C., and Mary, B.: Modelling temperature and moisture effects  
786 on C-N transformations in soils: comparison of nine models, *Ecol. Mod.*, 102, 325-339, 1997.
- 787 Rogers, H. H., Runion, G. B., and Krupa, S. V.: Plant responses to atmospheric CO<sub>2</sub>  
788 enrichment with emphasis on roots and the rhizosphere, *Environ. Pollut.*, 83, 155-189, 1994.
- 789 Schils, R. L. M., Kuikman, P., Liski, J., van Oijen, M., Smith, P., Webb, J., Alm, J., Somogyi,  
790 Z., van den Akker, J., Billett, M., Emmett, B. A., Evans, C. D., Lindner, M., Palosuo, T.,  
791 Bellamy, P. H., Jandl, R., and Hiederer, R.: Review of existing information on the  
792 interrelations between soil and climate change, *Alterra*, Wageningen, 208, 2008.
- 793 Selsted, M. B., van der Linden, L., Ibrom, A., Michelsen, A., Larsen, K. S., Pedersen, J. K.,  
794 Mikkelsen, T. N., Pilegaard, K., Beier, C., and Ambus, P.: Soil respiration is stimulated by  
795 elevated CO<sub>2</sub> and reduced by summer drought: three years of measurements in a multifactor  
796 ecosystem manipulation experiment in a temperate heathland (CLIMAITE), *Global Change  
797 Biology*, 18, 1216-1230, 10.1111/j.1365-2486.2011.02634.x, 2012.
- 798 Skopp, J., Jawson, M. D., and Doran, J. W.: Steady-State Aerobic Microbial Activity as a  
799 Function of Soil Water Content, *Soil Sci. Soc. Am. J.*, 54, 1619-1625,  
800 10.2136/sssaj1990.03615995005400060018x, 1990.
- 801 Soe, A. R. B., Gieseemann, A., Anderson, T. H., Weigel, H. J., and Buchmann, N.: Soil  
802 respiration under elevated CO<sub>2</sub> and its partitioning into recently assimilated and older carbon  
803 sources, *Plant Soil*, 262, 85-94, 10.1023/B:PLSO.0000037025.78016.9b, 2004.
- 804 Soussana, J. F., Fuhrer, J., Jones, M. B., and Van Amstel, A. R.: The greenhouse gas balance  
805 of grasslands in Europe, *Agric. Ecosyst. Environ.*, 121, 1-4, 2007.
- 806 Verburg, P. J., Arnone, J. A., III, Obrist, D., Schorran, D. E., Evans, R. D., Leroux-Swarthout,  
807 D., Johnson, D. W., Luo, Y., and Coleman, J. S.: Net ecosystem carbon exchange in two  
808 experimental grassland ecosystems, *Global Change Biol.*, 10, 498-508, 2004.
- 809 Volk, M., and Niklaus, P. A.: Respiratory carbon loss of calcareous grasslands in winter  
810 shows no effects of 4 years' CO<sub>2</sub> enrichment, *Funct. Ecol.*, 16, 162-166, 2002.
- 811 Wan, S. Q., and Luo, Y. Q.: Substrate regulation of soil respiration in a tallgrass prairie:  
812 Results of a clipping and shading experiment, *Global Biogeochem. Cyc.*, 17, 1054  
813 10.1029/2002gb001971, 2003.



814 Ward, J. K., and Kelly, J. K.: Scaling up evolutionary responses to elevated CO<sub>2</sub>: lessons from  
815 *Arabidopsis*, *Ecol. Lett.*, 7, 427-440, 2004.

816 Wasshausen, W.: Frühjahrspflege auf dem Grünland:Zehn Punkte beachten,  
817 *Landwirtschaftsblatt Weser-Ems*, 8, 6-8, 1987.

818 Zak, D. R., Pregitzer, K. S., Curtis, P. S., Teeri, J. A., Fogel, R., and Randlett, D. L.: Elevated  
819 atmospheric CO<sub>2</sub> and feedback between carbon and nitrogen cycles, *Plant Soil*, 151, 105-117,  
820 1993.

821 Zak, D. R., Pregitzer, K. S., King, J. S., and Holmes, W. E.: Elevated atmospheric CO<sub>2</sub>, fine  
822 roots and the response of soil microorganisms: a review and hypothesis, *New Phytol.*, 147,  
823 201-222, 2000.

824 Zhou, X., Sherry, R. A., An, Y., Wallace, L. L., and Luo, Y.: Main and interactive effects of  
825 warming, clipping, and doubled precipitation on soil CO<sub>2</sub> efflux in a grassland ecosystem,  
826 *Global Biogeochem. Cyc.*, 20, GB1003, doi:10.1029/2005GB002526, 2006.

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843 **Tables**

844 **Table 1**

845 Results of fitting the temperature-dependence model after Lloyd and Taylor (Lloyd and  
846 Taylor, 1994) to 20% of our observation data under ambient and elevated CO<sub>2</sub>.

CO <sub>2</sub> treatment	R	R <sup>2</sup>	Adjusted R <sup>2</sup>	Standard Error of Estimate
Ambient CO <sub>2</sub>	0.87	0.75	0.75	1.35
Elevated CO <sub>2</sub>	0.91	0.82	0.82	1.19

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862 **Table 2**  
 863 Annual sums of soil respiration under ambient and  $e\text{CO}_2$  from 2008 – 2010. Data are  
 864 presented as averages ( $n=3$ )  $\pm$  standard error (SE). P-values indicate the difference between  
 865 treatments per year obtained by a paired t-test.  
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Year	CO <sub>2</sub> treatment	Mean annual sum of soil respiration (g CO <sub>2</sub> m <sup>-2</sup> yr <sup>-1</sup> )	Mean annual sum of soil respiration (g C[CO <sub>2</sub> ] m <sup>-2</sup> yr <sup>-1</sup> )	Relative change to control (%)	P value
2008	Ambient CO <sub>2</sub>	4854 $\pm$ 34	1324 $\pm$ 9	1.22	0.17
	Elevated CO <sub>2</sub>	4913 $\pm$ 14	1340 $\pm$ 4		
2009	Ambient CO <sub>2</sub>	4928 $\pm$ 48	1344 $\pm$ 13	0.56	0.64
	Elevated CO <sub>2</sub>	4956 $\pm$ 39	1352 $\pm$ 11		
2010	Ambient CO <sub>2</sub>	4702 $\pm$ 37	1283 $\pm$ 10	1.38	0.23
	Elevated CO <sub>2</sub>	4767 $\pm$ 12	1300 $\pm$ 3		

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**Figure legends**

**Fig. 1** Seasonal patterns and the five defined seasons at the GiFACE grassland study site.

**Fig. 2** Volumetric water content under ambient and elevated CO<sub>2</sub> (a), daily sums of precipitation at the GiFACE (b), mean soil temperature during soil respiration measurements and minimum daily soil temperature at 10 cm depth (c), the relative mean monthly change of soil respiration under elevated CO<sub>2</sub> based on measured and modelled data (d), the absolute mean monthly difference in soil respiration under elevated CO<sub>2</sub> based on measured and modelled data (e), modelled soil respiration under ambient and elevated CO<sub>2</sub> from 2008 to 2010 (f) and measured soil respiration under ambient and elevated CO<sub>2</sub> from 2008 to 2010 (g). Data are presented as averages (n=3) ± 1 SE.

**Fig. 3** Mean soil respiration rates during the five defined seasons under ambient and elevated CO<sub>2</sub> averaged over three years from 2008 – 2010. Error bars show ± 1 SE associated by averaging across the three replicates per treatment (n=3) (1) = *winter dormancy*; (2) = *start of vegetation period*; (3) = *spring*; (4) = *summer*; (5) = *autumn* (for details see methods). P-values indicate the difference between treatments obtained by a linear mixed-effect model analysis.

911 **Fig. 4** Relationship between soil respiration rate and soil temperature under ambient and  
912 elevated CO<sub>2</sub>. Equation of dynamic fit (Lloyd and Taylor, 1994):  
913  $f = R_{10}e^{E_0\left(\frac{1}{(283.15-T_0)} - \frac{1}{(x-T_0)}\right)}$

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915 **Fig. 5** Observed versus modelled soil respiration rates under ambient and elevated CO<sub>2</sub>.

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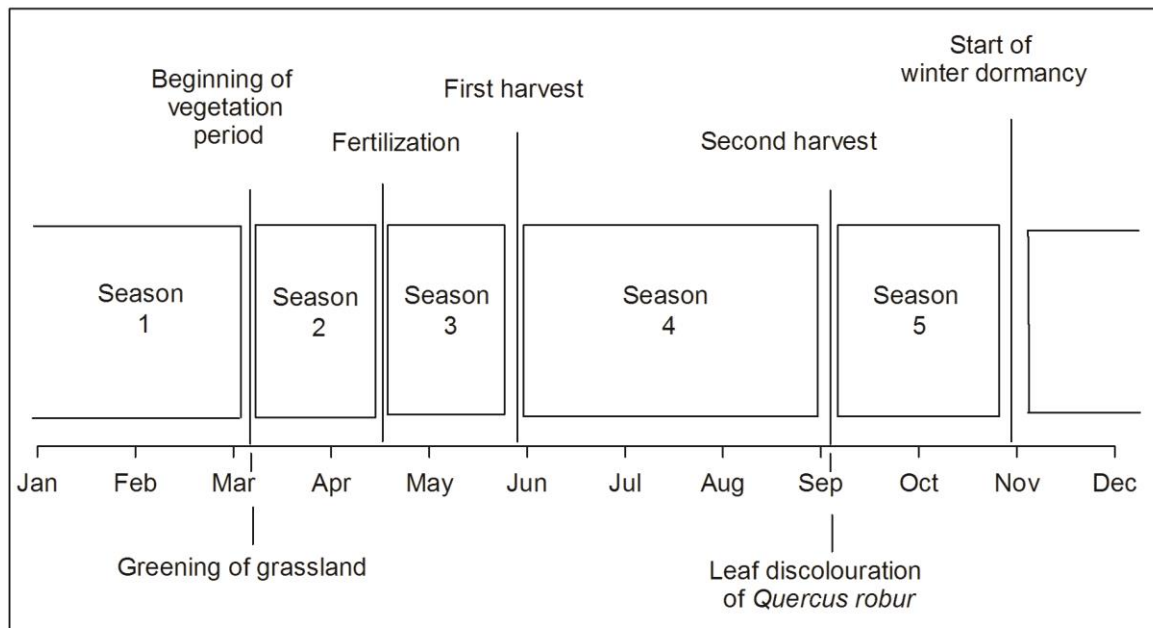
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925 **Figures**

926 Fig. 1



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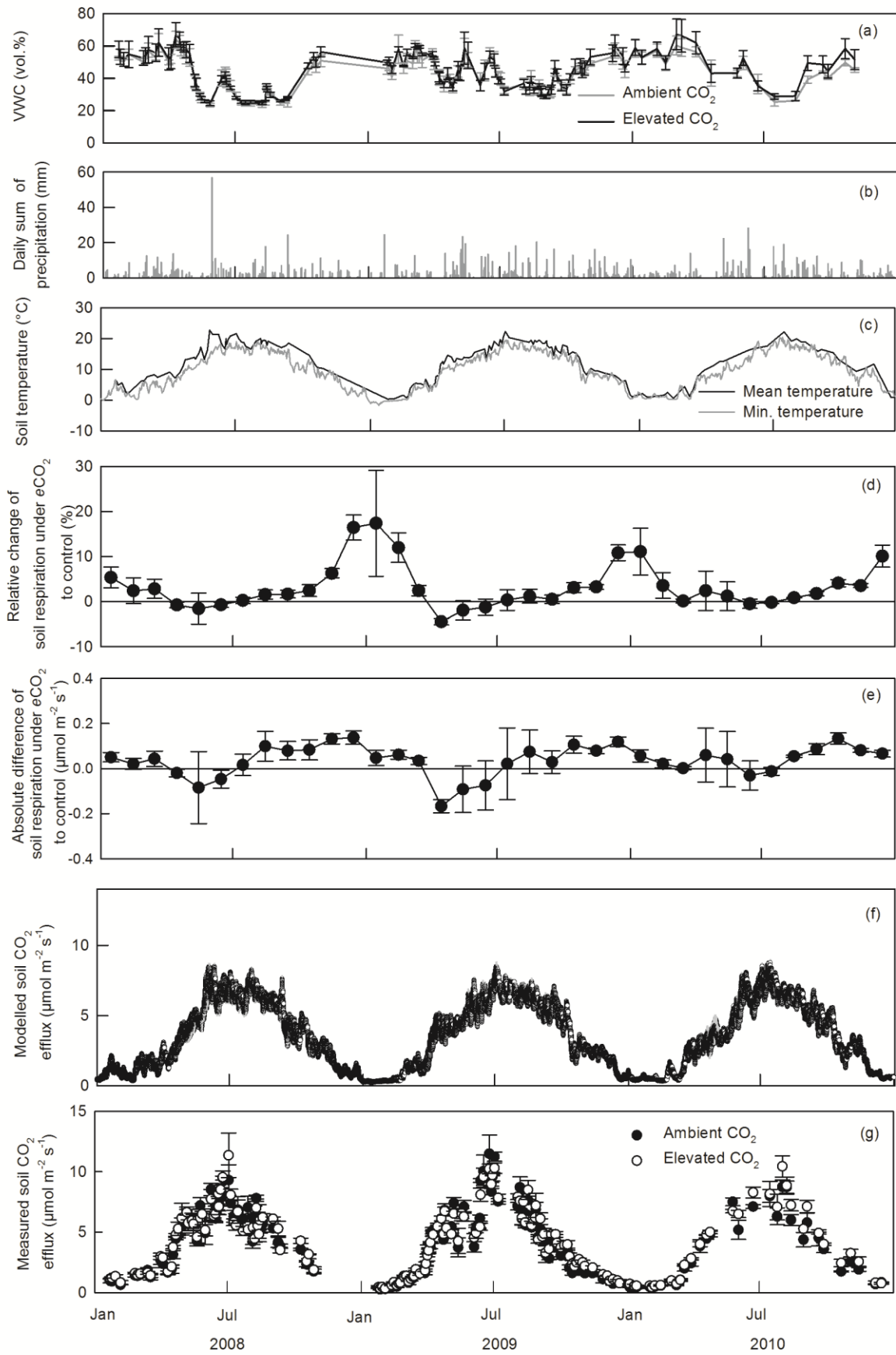
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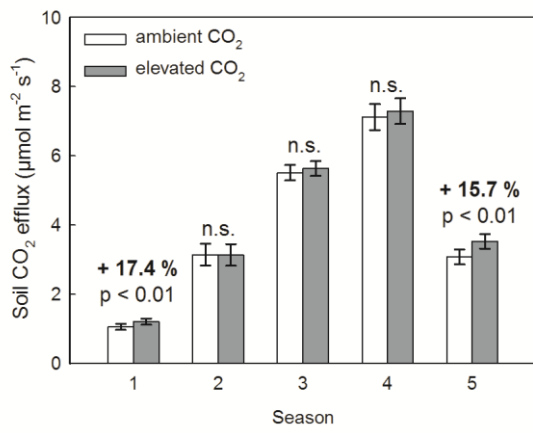
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939 Fig. 2





941 Fig. 3



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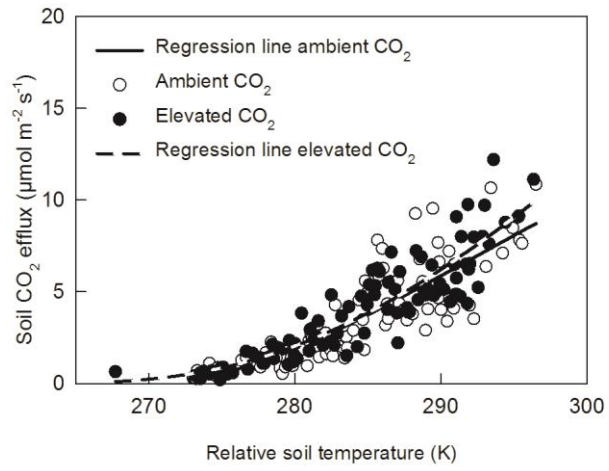
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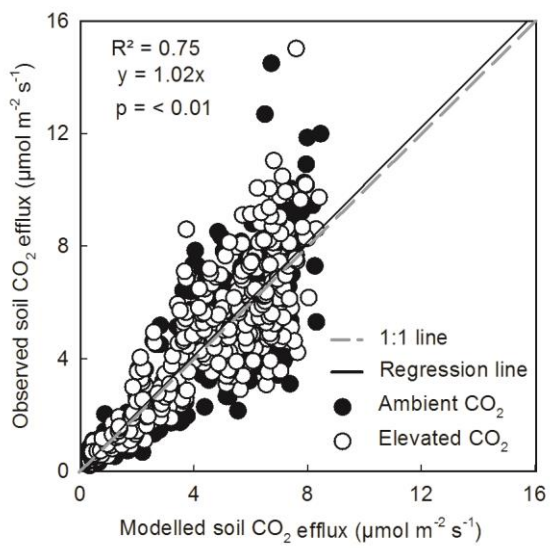
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976 Fig. 5

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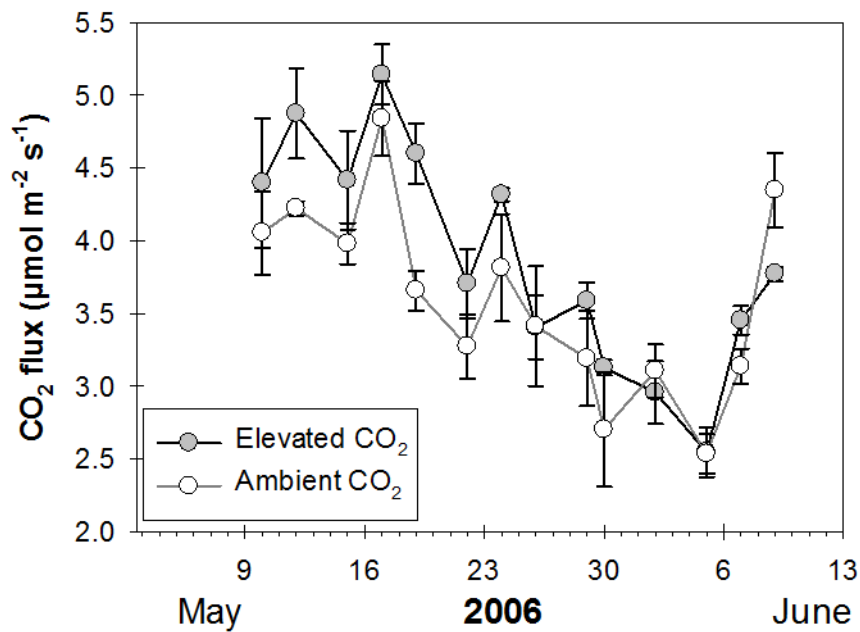
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993 **Supporting Information**

994 **Fig. S1**



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996 **Fig S1:** Mean CO<sub>2</sub> efflux +/- standard error (n=3) after installation of the frames and removal  
997 of the aboveground biomass on 9<sup>th</sup> May 2006.

998 On 11 out of 14 measurement occasions all three E-plot fluxes were higher than those of  
999 their corresponding A-plot partner. A mixed Model analysis (SPSS version 18) with the  
1000 factors CO<sub>2</sub>-treatment and time revealed that the soil CO<sub>2</sub> efflux was significantly increased  
1001 by CO<sub>2</sub> enrichment.

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1012 **Table S1**

1013 Parameter estimates of the temperature-dependence model after Lloyd and Taylor (1994)

CO <sub>2</sub> treatment	Model parameter	Coefficient	P value
	E0	61.92 ± 33.59	0.07
Ambient CO <sub>2</sub>	R10	3.00 ± 0.19	< 0.001
	T0	261.18 ± 6.53	< 0.001
	E0	143.68 ± 103.57	0.17
Elevated CO <sub>2</sub>	R10	3.11 ± 0.17	< 0.001
	T0	248.72 ± 13.35	< 0.001

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