

1 **Soil Respiration on an Aging Managed Heathland: Can Empirical Model Selection**
2 **Procedures Contribute to our Understanding of Ecosystem Processes?**

3

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9

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11 respiration modeling, model selection procedure

12

13 **Abstract**

14 Heathlands are cultural landscapes which are managed through cyclical cutting, burning or
15 grazing practices. Understanding the carbon (C) fluxes from these ecosystems provides
16 information on the optimal management cycle-time to maximize C uptake and minimize C
17 output. Soil respiration studies are increasingly undertaken with the aim of quantifying C
18 fluxes and predicting changes for the future. The interpretation of field data into annual C loss
19 values requires the use of soil respiration models. These generally include model variables
20 related to the underlying drivers of soil respiration, such as soil temperature, soil moisture and
21 plant activity. Very few studies have reported using model selection procedures in which
22 structurally different models are calibrated, then validated on separate observation datasets and
23 the outcomes critically compared. This study utilized thorough model selection procedures to
24 determine soil heterotrophic (microbial) and autotrophic (root) respiration for a heathland
25 chronosequence. During the model selection process, it became clear that measures of
26 photosynthesis, plant biomass, photosynthetically active radiation, root biomass, and microbial

27 biomass did not significantly improve model fit when included with soil temperature. This
28 contradicts many current studies in which these plant variables are used (although are not often
29 tested for parameter significance). The process indicated that one or more ecosystem processes
30 apart from the measured variables were important when trying to explain soil respiration in this
31 system. In the absence of these important covarying variables, soil temperature sufficed to
32 explain the seasonal variation observed in the ecosystem. The best predictive model used a
33 generalized linear multi-level model with soil temperature as the only variable. Total annual
34 soil C loss from the youngest and oldest communities was calculated to be $650 \text{ gC m}^{-2} \text{ year}^{-1}$
35 and $435 \text{ gC m}^{-2} \text{ year}^{-1}$, respectively. There were no heterotrophic respiration differences
36 between the community ages. In contrast, autotrophic respiration was significantly greater on
37 the youngest vegetation (50% of annual total soil C loss) and decreased as the plants aged
38 (oldest vegetation: 26% of total soil C loss).

39

40 1.0 Introduction

41 Soil respiration represents an important source of CO_2 in the biosphere as it is the second
42 largest flux after gross primary productivity in the global carbon (C) cycle, contributing 20 –
43 40% of atmospheric annual C input (Raich and Schlesinger 1992, Schlesinger and Andrews
44 2000). Soil respired CO_2 originates from a number of partitioned belowground sources and
45 total soil respiration (R_S) can be broadly categorized into autotrophic respiration (R_A : the
46 activity of roots and rhizosphere organisms) and heterotrophic respiration (R_H : bacteria and
47 fungi decomposition of organic matter and soil faunal activity in the organic and mineral
48 horizons) (Hanson et al. 2000). There has been increasing research attention directed towards
49 quantifying C losses from soil respiration, both at a local ecosystem scale and at a global scale,
50 with the aim of quantifying C balances and predicting C flux changes for the future.

51

52 These changes to soil C fluxes have been linked to anthropogenically induced conditions, such
53 as the IPCC predicted climate change (IPCC 2007), where increased soil warming has resulted
54 in increased C efflux rates (eg. Davidson and Janssens 2006, Rustad et al. 2001, Schindlbacher
55 et al. 2012), and prolonged drought periods have resulted in reduced C efflux rates (eg. Selsted
56 et al. 2012, Sowerby et al. 2008, Suseela et al. 2012). Changes in C fluxes can also be
57 associated with anthropogenic land management regimes, such as the selected land use (eg.
58 grazing; Peichl et al. (2012)), any subsequent land use change (Perez-Quezada et al. 2012); soil
59 disturbances (Novara et al. 2012) and cyclical vegetation management practices like heathland
60 burning or plantation forest harvesting (Clark et al. 2004, Clay et al. 2010).

61
62 Changes in CO₂ efflux are often associated with changes to the major drivers of R_S activity in
63 an ecosystem. These include abiotic factors, such as temperature and soil moisture, and biotic
64 factors, such as gross primary productivity (Bahn et al. 2010a, Davidson and Janssens 2006,
65 Trumbore 2006). These factors can interact with each other or can independently affect soil
66 respiration from either R_H or R_A (Davidson et al. 2006). The R_H is proportionate to the
67 decomposition of soil carbon by microbial communities, which use mostly the recently
68 produced organic matter as an energy source (Ryan and Law 2005, Trumbore 2006). In
69 contrast, CO₂ lost from autotrophic activity is tied to the assimilation of organic compounds
70 supplied by plant metabolism with a part of this carbon rapidly released from the soil (Horwath
71 et al. 1994, Metcalfe et al. 2011, Ryan and Law 2005). Live root respiration is typically
72 quantified either by using an isotopic approach, such as repeated pulse labeling, continuous
73 labeling, natural abundance (following change of land use/species); by vegetation removal
74 techniques, such as tree girdling; or by using one of the root exclusion methods, such as root
75 removal, trenching and gap analysis (Chemidlin Prévost-Bouré et al. 2009, Díaz-Pinés et al.
76 2010, Gomez-Casanovas et al. 2012, Graham et al. 2012, Hanson et al. 2000, Jassal and Black
77 2006).

78

79 Once field measurements have been collected, respiration data has generally been interpreted
80 through statistical analysis to determine any treatment effects. Many studies then additionally
81 processed their observations using the known exponential relationship between organic matter
82 decomposition and temperature (Davidson and Janssens 2006, Sierra et al. 2011) to determine
83 Q_{10} values and investigate the sensitivity of R_S to temperature within their studied treatments
84 (eg. Sowerby et al. 2008, Suseela et al. 2012, Webster et al. 2009, Xiang and Freeman 2009).
85 However, a much fewer number of studies calculated a continuous CO_2 efflux time series for
86 either the length of the study period or predicted for a projection into the future, to allow the
87 annual C loss from R_S (or R_H and R_A) to be estimated. Where these continuous efflux series
88 were modeled, other ecosystem measures have, in some cases, been included in the equations.
89 These have included soil water content or precipitation, as organic matter decomposition and
90 plant activity are affected by moisture availability (Davidson et al. 2006, Raich and
91 Schlesinger 1992). Increasingly, measures of plant activity, such as plant metabolism or litter
92 production, have also been included within the soil respiration models to link the aboveground
93 processes with the belowground processes that occur within ecosystems (Bahn et al. 2010b,
94 Metcalfe et al. 2011, Ryan and Law 2005). The degree to which soil respiration models could
95 be modeled with apriori parameter values or required calibration was often dependent on both
96 the spatio-temporal scale at which the models were to be applied and the available
97 environmental data (Keenan et al. 2012).

98

99 Where modeling was used to generate annual C estimates (rather than to generalize the results
100 of an experiment or survey), most studies assessed their selected model using measures of fit
101 for the calibration-data (eg. Kutsch et al. 2010, Selsted et al. 2012), with many fewer studies
102 evaluating models through a (cross-)validation procedure on separate observation data sets
103 (Caquet et al. 2012, Webster et al. 2009). Furthermore, relatively few studies considered the

104 evaluation of structurally different models and a complete variable selection procedure (Chen
105 et al. 2011, Webster et al. 2009). Recently, several review studies have discussed progress in
106 the modeling of soil respiration and proposed better model-data integration with more rigorous
107 and critical procedures to test respiration models (Keenan et al. 2012, Vargas et al. 2011).
108 Interestingly, soil respiration trial measurements have often been collected repeatedly in time
109 (i.e. longitudinal) and clustered in space but this method has generally not been discussed
110 within the context of soil respiration models. This type of data should ideally be analyzed by
111 hierarchical (multi-level) model framework. In this framework, the measurements which are
112 collected for the same observation unit are explicitly assumed to be dependent, which leads to a
113 more realistic estimate of the effective degrees of freedom (and consequently more realistic
114 confidence bounds) than when assuming independent observations. However, only a few soil
115 respiration studies adopt a multi-level modeling approach (Bernhardt et al. 2006), whereas
116 multi-level modeling is commonplace in many other areas of ecology and the environmental
117 sciences (Qian et al. 2010). In this study, we aimed to follow these guidelines to implement
118 good modeling practices and build predictive models for R_S , R_A and R_H for a managed
119 heathland site. The ultimate goal of this research was to evaluate soil respiration fluxes for the
120 heathland at different vegetation development phases, which would allow for future calculation
121 of a C balance.

122
123 Heathlands are cultural landscapes in which cyclical management practices, such as cutting,
124 burning or grazing are undertaken (Webb 1998). It is known that the structure of the dominant
125 heathland plant (*Calluna vulgaris*) changes with increasing plant age, from a 'net biomass
126 gain' phase up until 15 years of age, to a 'net biomass loss' phase after this time (Gimingham
127 1985). It was hypothesized that the younger vegetation community would have the highest
128 plant activity, resulting in greater allocation of C to the roots and therefore a greater R_A (and
129 subsequently greater R_S) than on the older communities. Community age was not expected to

130 influence R_H as there was no significant difference in the quantity of microbial energy source
131 (carbon stock) between the vegetation ages prior to treatment application (Kopittke et al.
132 2012). Therefore, based on the known relationships between microbial respiration, soil
133 temperature and soil moisture, it was hypothesized that soil temperature and soil moisture
134 would be significant variables for the R_H model. In addition, based on the contribution of plant
135 metabolism to root processes, it was hypothesized that soil temperature, soil moisture and a
136 measure of plant activity would contribute significantly to the R_S models for all three
137 community ages.

138

139 2.0 Materials and Methods

140 2.1 Study Site

141 The investigation was undertaken at a dry heathland, located approximately 25 meters a.s.l. at
142 Oldebroek, the Netherlands. The dominant vascular species at the site is *Calluna vulgaris* (L.)
143 Hull which grows to a maximum height of 75 cm and provides approximately 95% of the
144 groundcover, with some *Deschampsia flexuosa* and *Molinia caerulea*. The dominant non-
145 vascular species is *Hypnum cupressiforme* Hedw. with two ecological phenotypes, one
146 growing under *Calluna* protection and the other adapted to more light between *Calluna* plants.

147

148 The trial was established within a 50 m x 50 m area, at the convergence of three *Calluna*
149 communities of different ages. Each community age was considered to be a treatment.

150 Replication of these treatments was not possible due to the inherent nature of the site.

151 Therefore, a quasi-experimental design was used, in which groups were selected upon which
152 the variables were tested but where randomization and replication processes were not possible
153 (Campbell and Stanley 1966).

154

155 The oldest heathland area (the Old community) was approximately 28 years of age at the
156 conclusion of the investigation, while the vegetation on the south-eastern third of the research
157 site was approximately 19 years of age (the Middle community). The southern portion of the
158 site was last cut in the year 2000 as part of the creation of a fire break and was 12 years old (the
159 Young community) at the conclusion of the study.

160
161 The site is relatively flat in the west and rises in the east and north-east onto a gentle slope with
162 a south-western aspect. The soil is a nutrient-poor, well drained, acid sandy haplic podzol (van
163 Meeteren et al. 2008). The soil has an organic horizon which ranged between 1.4 and 8 cm
164 thick, with the mean thickness of $3.9 \text{ cm} \pm 0.04$ (Kopittke et al. 2012). The carbon stock of the
165 soil (organic layer and to 25cm depth of mineral soil) was $8.01 \pm 0.6 \text{ kg m}^{-2}$ on the Young
166 community, $7.61 \pm 0.5 \text{ kg m}^{-2}$ on the Middle community and $6.18 \pm 0.4 \text{ kg m}^{-2}$ on the Old
167 community and were not significantly different to each other (Kopittke et al. 2012). Further
168 information about the site location, species composition and climate is provided in Table 1.

169

170 *2.2 Experimental Design*

171 To measure soil respiration and calibrate soil respiration models, eight experimental plots
172 (60cm x 60cm) were established within each heathland age in April 2011 (n=24). Four of these
173 plots were used to measure heterotrophic respiration on each community age (henceforth called
174 ‘Trenched’ plots; n=12) and the other four were used to measure total respiration on each
175 community age (‘Untrenched’ plots; n=12). In this study, the terminology ‘total soil
176 respiration’ and ‘heterotrophic soil respiration’ refers to the observed field data from the
177 Untrenched plots and Trenched plots, respectively. Due to the inherent nature of the site,
178 randomization of the factor ‘community age’ is not possible in our experiment. However,
179 colinearity of weather data with the distribution of the three age classes is highly unlikely since

180 the area is small compared to variations in weather-variables. Furthermore, soil data (including
181 soil temperature and soil moisture) appear not to vary much between the age classes.

182 The terminology ‘ R_S ’, ‘ R_H ’ and ‘ R_A ’ refers to the modeled total soil respiration, modeled
183 heterotrophic soil respiration and modeled autotrophic respiration, respectively.

184
185 The plots were placed in pairs (one Trenched in combination with one Untrenched plot) that
186 were 1.5 m apart, but the exact location of the individual plot as well as the location of the
187 pairs were randomly allocated within each vegetation age (Figure 1). In May 2011, the
188 aboveground biomass was harvested from the four Trenched plots within each age group and a
189 narrow trench was excavated to 50cm depth around the 60 x 60 cm plot area. This depth
190 extended below the main rooting zone, but was above the water table and did not encounter
191 any impermeable layers, all of which may have affected CO_2 concentration productions at
192 depths (Jassal and Black 2006). A nylon mesh (Plastok Associated Ltd, Birkenhead, Wirral,
193 UK) of 41 μ m was placed in the trench to prevent the new roots growing into the plots during
194 the experiment. The soil horizons were backfilled in the order of removal to keep soil
195 disturbance to a minimum. Any subsequent vegetation regrowth was periodically removed but
196 the remains left in the plot on the soil surface. The remaining four Untrenched plots in each
197 vegetation age were not disturbed and were used as a control treatment.

198
199 For the purposes of soil respiration model validation, an additional four plots (‘Trenched
200 Validation’ plots) in each heathland age group were trenched using the described method
201 (n=12) and data collected for the purposes of validating the derived R_H model. A further nine
202 untrenched plots (‘Untrenched Validation’ plots) were established in the Old vegetation and the
203 collected data was used for validation of the derived R_S model.

204

205 *2.3 Site Meteorological and Treatment Soil Conditions*

206 Site meteorological conditions were recorded on an hourly basis (Decagon Devices Inc.; DC,
207 USA). Air temperature and relative humidity measurements were obtained from 20 cm above
208 ground surface at a central location on the site. Rainfall was measured using a Vaisala tipping
209 bucket rain gauge (Vaisala; Vantaa, Finland) connected to a Decagon datalogger.

210
211 Treatment soil conditions were recorded on an hourly basis (Decagon Devices Inc.; DC, USA).
212 Soil moisture ($\text{m}^3 \text{m}^{-3}$) and soil temperature ($^{\circ}\text{C}$) measurements were obtained from 4 – 7 cm
213 below ground surface in two Trenched plots, two Untrenched plots, and two Trenched
214 Validation plots in each heathland age group (5TM Sensor, Decagon Devices Inc., DC, USA).
215 The same measurements were obtained from the three Untrenched Validation plots on the Old
216 community. In total, 21 soil probes were installed, with six being in the Young community, six
217 in the Middle community and nine in the Old community.

218

219 *2.4 Soil Respiration Measurements*

220 Respiration collars of 10 cm diameter and 6 cm height were inserted approximately one
221 centimeter into the soil surface in each plot, maintaining a buffer zone of 10 cm from the plot
222 boundary. In the Untrenched plots, moss was removed from inside these collars, to ensure that
223 only soil respiration was measured. Moss was not present on the Trenched plots as it had been
224 removed during trenching activities. Soil respiration measurements were obtained using a
225 Portable Gas Exchange and Fluorescence System (LI-6400XT; LICOR Biosciences, Lincoln,
226 NE USA) in combination with a soil CO_2 flux chamber (LI-6400-09; LICOR Biosciences)
227 which fitted onto the collars.

228

229 Soil respiration measurements using this methodology commenced in May 2011, three days
230 after trenching occurred, and continued until August 2012. A total of 29 measurement events
231 occurred post-trenching on the three ages of vegetation. A common effect of a trenching

232 methodology is a flush of CO₂ within the first weeks or months after trenching which
233 originates from decomposing roots (Hanson et al. 2000). To minimize this effect of root
234 decomposition, the first four months of CO₂ efflux measurements were excluded from the
235 study and only observations after 21 September 2011 are included in the analyses. In addition,
236 to determine if there had been significant root biomass loss from the Trenched plots (ie
237 decomposition) during the study period, the root biomass in the Trenched and the Untrenched
238 plots was assessed one year after trenching activities. There were 19 soil respiration
239 measurement events from September 2011 until August 2012.

240
241 Soil respiration measurements using the above methodology were also obtained from the
242 Trenched Validation plots to validate the R_H model and from the Untrenched Validation plots
243 to validate the Old vegetation R_S model.

244 245 *2.5 Photosynthesis Measurements*

246 The gross photosynthetic rate provided a measure of plant activity for the three heathland ages.
247 This gross photosynthetic rate (P_G) was calculated as the Net Ecosystem Exchange (NEE) rate
248 of CO₂ flux minus the Ecosystem Respiration (ER) rate of CO₂ flux ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). This
249 photosynthetic rate has a negative sign. A loess smoother curve was applied to the P_G data to
250 obtain daily estimates of plant activity for use in the soil respiration models.

251
252 Three permanent sampling locations were selected in each vegetation age. A metal base frame
253 (60cm x 60cm) was permanently installed using small, narrow sandbags to provide a seal
254 between the frame and the soil surface and fixed with metal pins. The CO₂ fluxes of the
255 vegetation were measured with the same LI-6400 infrared gas analyzer as used for the soil
256 respiration measurements (LI-COR, Lincoln, NE, USA) but in this case attached to a 288 L

257 ultra-violet light transparent Perspex chamber (60cm x 60cm x 80cm). Full details of the NEE
258 and ER method used at this site are provided in Appendix A.

259

260 *2.6 Plant and Microbial Biomass*

261 The biomass harvested from the Trenched plots in April 2011 was separated into Calluna and
262 moss layers. These components were oven dried at 70°C and the dry weight recorded (n=12).

263

264 Microbial biomass and root biomass were sampled in May 2012, approximately one year after
265 trenching activities. Soil sampling was undertaken using a soil corer of 5cm diameter and intact
266 soil samples were obtained from the organic horizon and 0-5cm mineral soil. Three cores were
267 obtained and were bulked by soil horizon from each Trenched plot (n=12) and Untrenched plot
268 (n=12). The soils were kept refrigerated during preparation. All the soil was sieved and roots
269 were separated, washed, oven dried at 70°C and the root dry weight calculated for the organic
270 and the mineral horizon.

271

272 In the organic horizon, each sample was divided into three subsamples of each 10 g. One part
273 was analyzed for water content by drying at 70°C and bulk density was then calculated.

274 Samples were ground and the carbon concentrations were analyzed on a CNS analyzer (Vario
275 EL Analyzer, Elementar). Another subsample was fumigated with the chloroform-fumigation
276 method and extracted for 1 h in 50 ml 0.5 M K₂SO₄ (Jonasson et al. 1996) . The third soil
277 fraction was extracted for 1 h without prior fumigation for initial content of carbon and
278 nutrients. The extractions were frozen until shortly before analysis. Upon defrosting, analysis
279 of total organic C (TOC) was undertaken on a vario TOC cube (Elementar). Microbial C was
280 estimated as the difference between the concentration of TOC in the fumigated and

281 unfumigated extract. An extractability constant of $K_{EC} = 0.45$ was used for microbial C (Jensen
282 et al. 2003). Microbial C (mg) of the organic horizon is reported per gram of substrate C.

283

284 *2.7 Data Analysis*

285 The data analysis workflow approach is described in the following sections and is summarized
286 in Figure 2. Initially, the observational data was analyzed to determine if there were
287 statistically significant differences between community ages (an age effect) or between
288 Trenched and Untrenched plots (a methodological effect). This indicated how the datasets
289 should be grouped in the later modeling phase; for example, if there was no soil respiration
290 difference between Trenched plots on the three community ages and there was no hypothesized
291 environmental reason as to why there should be a R_H difference, then the three age datasets
292 were grouped for the modeling phase.

293

294 Once the observation data had been statistically analyzed, a number of plausible model formats
295 and explanatory variables were chosen for calibration and validation (Jørgensen and
296 Bendoricchio 2001). The explanatory variables were chosen around the major drivers of R_S
297 and R_H , being abiotic factors, such as temperature and soil moisture, and biotic factors, such as
298 gross primary productivity (Bahn et al. 2010a, Davidson and Janssens 2006, Trumbore 2006).
299 A number of drivers were considered for inclusion as explanatory variables but the final
300 decision was based on the observation data available, the outcome of the statistical analysis, the
301 variables used in other studies and the outcome of a preliminary fitting of the models.

302

303 Preliminary model fitting indicated that no model could account for the extreme values
304 recorded on 21 March 2012, which were associated with an extreme meteorological episode
305 (freeze followed by thaw). In addition, the misfit on this day dominated the overall
306 performance criterion. These extreme values are most likely associated with the death of fine

307 roots and microbial populations during a late winter, extreme freeze period (-20°C), followed
308 by the rapid recovery of microbial populations as daytime air temperatures reached >15 °C
309 which all lead to short term fluxes of CO₂ from the soil (Matzner and Borcken 2008, Sulkava
310 and Huhta 2003). Although these CO₂ releases occur, there is strong evidence that these events
311 have little effect on soil C losses at an annual time scale (Matzner and Borcken 2008), therefore
312 it was decided to omit this specific extreme event in the modeling process. This allowed the
313 model to be calibrated and validated more accurately on the observations in which non-extreme
314 processes are believed to be dominant.

315
316 The models were calibrated and validated, using the procedures described in the following
317 sections. Based on these results, a model was selected and soil respiration rates were predicted.
318 These values were used to estimate annual C losses for R_S, R_H and R_A for each community.

319 320 2.7.1 Observational Data Analysis

321 The effect of community age on the single occasion measurements observed biomass (plant
322 leaves/shoots, plant roots and microbes) was investigated by a linear model ANOVA. If a
323 treatment effect was identified, then a pairwise t tests (using the Bonferroni correction factor)
324 was undertaken whereby an effect is considered as significant if it's associated p-value is
325 smaller than 0.05. The effect of vegetation age on the repeated measurements (soil respiration
326 and on photosynthetic activity) was investigated using a linear multi-level model (Pinheiro and
327 Bates 2000). Where the response variable in the linear multi-level model was the CO₂ efflux
328 measurement (a repeated measurement per location), the vegetation ages formed the fixed
329 effects and the measurement locations formed the random effects.

330

331 Where mean results are referenced, the standard errors of the mean (SEM) are provided in both
332 text and graphics. For all statistical analyses, the R statistical computing program was used (R
333 Development Core Team 2008).

334

335 *2.7.2 Soil Moisture Model*

336 A zero-dimensional finite difference soil moisture model (i.e. a 'bucket model'), with a daily
337 time resolution and model inputs of rainfall plus air temperature, was constructed and
338 calibrated on the observed soil moisture data. When compared to observed data, the soil model
339 gives an unbiased prediction and explains approximately 70% of the variance (the details of
340 this model are given in Appendix B). The soil moisture information in this study is used as a
341 potential explanatory variable for respiration. A soil moisture model, rather than observed soil
342 moisture, was used for two reasons. Firstly, a dynamic model is an appropriate method to
343 integrate the soil moisture values per sensor to an average soil moisture value per treatment and
344 this integration is necessary because not all plots were equipped with a soil moisture sensor.
345 Secondly, it overcomes problems of missing data, such as when a respiration model is used at
346 other sites for predictive purposes: in these cases, the soil moisture data is usually not
347 available, whereas daily rainfall and temperature are commonly present.

348

349 *2.7.3 Soil Respiration Model Calibration and Validation*

350 A model comparison framework was used to assess the R_S models and R_H models (Burnham
351 and Anderson 2002). A number of plausible models were calibrated and only the models with
352 significant parameter values were retained. These models were ranked according to the root
353 mean squared error for the calibration data ($RMSE_C$) and the models with low $RMSE_C$ were
354 considered suitable for further validation and discussion.

355

356 Validation of the suitable models was done with soil respiration data obtained from the
357 validation plots. The models were fitted and validated to data in accordance with Table 2,
358 where validation was conducted on different observation data over the calibration time period
359 (Validation Type I) and for a different time period (Validation Type II). A third validation
360 method (cross-validation) was also used in the model selection procedure. The cross-validation
361 results did not alter the outcomes of the model selection procedure; therefore, the full details on
362 method and results are not discussed further here but are provided in Appendix C.

363
364 For each of the validation data sets, a root mean squared error ($RMSE_V$) was calculated. The
365 $RMSE$ is specified in equation 1.

$$366 \quad RMSE = \sqrt{\frac{\sum_{i=1}^n (\hat{R}_i - R_i)^2}{n}} \quad (1)$$

367 where \hat{R}_i is the predicted respiration at time i , R_i is the observed respiration at time i and n is
368 total number the number of observations. The general equation is identical when applied to
369 calibration or validation data, as well as for R_S and R_H .

370
371 The group of plausible models was built-up as follows. First, an existing soil respiration model
372 was selected from a study undertaken on a comparable *Calluna vulgaris* heathland located in
373 Denmark (Selsted et al. 2012). This model (henceforth denoted as the Selsted model) is used in
374 this study as a null model for both R_S and R_H . It is a non-linear model with three explanatory
375 variables (temperature, soil moisture and biomass) and four parameters that need to be
376 calibrated (further details follow below). The model selection procedure calibrated and
377 validated not only the full model with three explanatory variables, but also the more
378 parsimonious variants with two variables (temperature and soil moisture or temperature and
379 biomass) and with one variable (temperature).

380

381 Next, a linear multi-level model (LMM) with the same variables as the Selsted model was
382 calibrated and validated. The multi-level structure is required to deal with the repeated
383 measurements on individual locations. Furthermore, a generalized linear multi-level model
384 (GLMM) with a Gamma-distributed error and a log link function (again with the same
385 variables) was calibrated and validated. Generalized linear models extend linear models that
386 involve non-normal error distributions or heteroscedasticity and may also require a
387 transformation to become linear. Linear functions of the predictor variables are obtained by
388 transforming the right side of the equation by a so-called link function. In this case the shape of
389 the relationship is exponential, so by taking its logarithm it becomes linear. The data are then
390 fit in this transformed scale (using an iterative routine based on least squares), but the expected
391 variance is calculated on the original scale of the predictor variables. The Gamma distribution
392 describes that the error is right-skewed at low values of the predictor variable and becomes
393 symmetric at higher values. In our case, the mean and variance of the model error are equal
394 (McCullagh and Nelder 1989).

395
396 In a next step, the soil moisture and biomass variables were transformed into quadratic
397 variables and the LMM and GLMM models using these variables were also calibrated and
398 validated (these models are denoted by LMM2 and GLMM2). These quadratic forms of the
399 models were successfully applied in the study by Khomik et al. (2009).

400
401 Following the approach by Selsted et al. (2012), soil moisture as well as biomass was scaled to
402 represent relative soil moisture and relative biomass. Equations 2 and 3, respectively, provide
403 the details of these transformed variables.

404
405
$$M = \frac{\theta}{\theta_{fc}} \quad (2)$$

406 where M is the relative soil moisture content (a fraction between approximately 0.1 and 1), θ is
407 the volumetric soil moisture content (in this study output from a dynamic soil moisture model,
408 Section 2.7.2), θ_{fc} is the soil moisture content at field capacity. An estimate for θ_{fc} was
409 available per treatment from the soil moisture model (section 2.7.2).

410

$$411 \quad B = \frac{\text{Biomass}}{\text{Max Biomass}} \quad (3)$$

412 where B is the relative biomass (a fraction between approximately 0.3 and 1), ‘Biomass’ is the
413 aboveground Calluna biomass in g m^{-2} for a given observation plot and ‘Max Biomass’ (a value
414 of 2.2) for the plot with the greatest quantity of aboveground biomass. Moss was also harvested
415 from the plots, however only the Calluna biomass was used in this calculation as the Calluna
416 root systems were expected to contribute to R_A but the moss layer lacks a rooting system and
417 would not contribute to R_A . For the model developed by Selsted *et al.* (2012), peak biomass
418 was estimated using non-destructive techniques. In the current study, the biomass initially
419 harvested from the Trenched plots within each nested replicate was used as an estimate of
420 aboveground biomass for the Untrenched plots in the same nested replicate.

421

422 However, as harvested biomass does not give a dynamic measure of plant activity throughout
423 the year and the changes of seasons, a measure of photosynthetic activity (Section 2.5) was
424 included in the model testing process as an alternative variable for Calluna biomass. A value
425 for relative photosynthetic activity was calculated as follows.

$$426 \quad P = \frac{P_G}{\text{Maximum } P_G} \quad (4)$$

427 where P_G is the gross photosynthesis measured per plot in $\mu\text{mol CO}_2\text{m}^{-2}\text{s}^{-1}$, and Maximum P_G
428 is the maximum CO_2 consumption rate measured during the study, as described in section 2.5.
429 The absolute values for Maximum P_G were 23.2, 12.2 and 8.1, respectively, for Young, Middle
430 and Old communities.

431
432 In the first modeling cycle, the soil temperature at 5 cm depth (T_{soil}) was used, as it is a
433 common component of soil respiration models. However, in a second modeling cycle, air
434 temperature (T_{air}) was also tested as a substitute for soil temperature, as it is often a more
435 commonly recorded variable across ecosystems. The equations for the Selsted, LMM, LMM2,
436 GLMM and GLMM2 models using the T, M, B and P variables (see equations 2 to 4) as
437 predictor variables are shown in Table 3.

438
439 In addition to the variables detailed above, a number of other variables were tested in an early
440 explorative phase that occurred prior to the formal model identification process. These other
441 variables included Photosynthetically Active Radiation (PAR) values used as a substitute for
442 the P variable, the microbial biomass as a substitute for the B variable and the root biomass as
443 a substitute for the B variable in both R_S and R_H models. In that explorative phase, it was found
444 that the $RMSE_C$ and $RMSE_V$ values for the models involving these variables were higher or
445 close to those variables shown in Table 3. Therefore, the results of these alternative variable
446 combinations were not tested further.

447

448 *2.7.4 Soil Respiration Model Selection and Generation of Predictions*

449 The final models for R_S and R_H were selected using the following rationale. Firstly, the
450 calibrated models in which all coefficients were significant were identified and retained for
451 further consideration. Secondly, only models in which parameters were feasible according to
452 literature values and experience were retained. The reasonableness of these parameters were
453 defined for basal respiration rate: (R_0 0 to 0.5 for the Selsted and GLMM models), $a > 0$ and c
454 > 0 (GLMM models) or $a < 0$ and $c < 0$ (LMM models). For the R_H models, a complete set of
455 validation data for each vegetation age was available. Therefore, the subset of R_H models with

456 significant parameter values were further assessed by their $RMSE_{V1}$ values, and those with the
457 lowest values were considered most suitable.

458

459 In the R_S models, the validation data and therefore, the $RMSE_{V1}$ and $RMSE_{V2}$'s, were only
460 available for the Old community. Consequently, the $RMSE_C$ provided a better measure of
461 model performance across each age of vegetation. Hence, the R_S models with significant
462 parameter values and the lowest $RMSE_C$ were selected while the values for $RMSE_{V1}$ and
463 $RMSE_{V2}$ were of secondary importance (these should lie in the lower to middle-range of all
464 $RMSE$ values).

465

466 Following the selection of the model, R_S and R_H were predicted for the length of the study
467 period using a single hourly soil temperature dataset from the Untrenched treatment. The mean
468 annual C loss from R_S and the 95% confidence intervals of model predictions were calculated
469 using a bootstrap procedure with 1000 replications (Davison and Hinkley 1997).

470

471 3.0 Results

472 3.1 *Vegetation Characteristics*

473 Destructive vegetation sampling indicated that mean *Calluna* aboveground biomass was lowest
474 on the Young community and greatest on the Middle community. This difference was just
475 above the 0.05 significance level after the Bonferroni correction ($p=0.059$; Figure 3a). The
476 biomass of the moss layer was almost double on the Young community ($0.43 \pm 0.09 \text{ kg m}^{-2}$)
477 than the moss biomass on either the Middle community ($0.27 \pm 0.04 \text{ kg m}^{-2}$) or Old community
478 ($0.26 \pm 0.04 \text{ kg m}^{-2}$; results not shown).

479

480 Photosynthesis, as a measure of plant activity throughout the year, was greatest in the summer
481 months, least in the winter months and was significantly different between communities

482 ($F=25.1$, $p<0.001$;Figure 3b). In winter months, there was no significant difference between
483 mean photosynthesis on the Young ($-2.1 \pm 0.7 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$), Middle ($-1.0 \pm 0.3 \mu\text{mol CO}_2$
484 $\text{m}^{-2} \text{ s}^{-1}$) or Old ($-1.8 \pm 0.5 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) communities. However, in summer months there
485 was significantly greater photosynthesis on the Young community ($-16.0 \pm 1.4 \mu\text{mol CO}_2 \text{ m}^{-2}$
486 s^{-1}) than on either the Middle community ($-5.7 \pm 1.5 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) or the Old community
487 ($-5.2 \pm 1.0 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). The Old community was significantly different to the Middle
488 community in summer ($F=48$, $p=0.049$) but there were no other seasonal differences between
489 the mean photosynthetic rates of the Middle and Old communities during the study period.

490

491 3.2 Soil Respiration

492 The age of the vegetation had a significant effect on soil respiration ($F=5$, $p=0.035$) and in
493 every season of the year, total soil respiration was significantly greater on the Young
494 community than on the Old community (winter $p=0.034$, spring $p = 0.0144$, summer $p = 0.007$,
495 autumn $p=0.006$). The greatest mean total soil respiration was recorded in summer months on
496 all three communities, ranging from a mean of $2.8 \pm 0.2 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ on the Young
497 community to $2.1 \pm 1.9 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ on the Old community (Figure 4a). The lowest mean
498 soil respiration values were recorded in winter, although soil respiration was still significantly
499 greater than zero ($t=14.7$, $p<0.001$) in these colder conditions. The differences between the
500 communities were greatest in spring with total soil respiration on the Young community ($1.9 \pm$
501 $0.2 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) exceeding respiration on the Middle community by a factor of 1.6 and
502 exceeding the Old community by a factor of 1.7.

503

504 There was no effect of community age in any season for heterotrophic soil respiration on the
505 Trenched plots (Figure 4b). Therefore, the heterotrophic data was not split into age treatments
506 for further analyses, but was treated as a single dataset. Mean heterotrophic soil respiration was

507 least in winter months ($0.4 \pm 0.05 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) and greatest in summer months ($1.7 \pm$
508 $0.09 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$).

509
510 A peak was observed in both total soil respiration and heterotrophic soil respiration on 21
511 March 2012. The elevated respiration results were observed on both Trenched and Untrenched
512 plots and, although the CO_2 flux was variable between measurement locations, the largest
513 fluxes were generally observed on the Young community. The maximum respiration observed
514 on this day for the Trenched plots was $10.28 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (Young community) and for the
515 Untrenched plots was $5.11 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (also Young community).

516
517 *3.3 Treatment Effect*
518 Soil temperature at 5 cm below ground surface was significantly different between the
519 Trenched plots and the Untrenched plots over the study period (Figure 5a). The mean soil
520 temperature in winter was significantly lower on the Trenched plots ($3.8 \pm 0.03 \text{ }^\circ\text{C}$) than on the
521 Untrenched plots ($4.8 \pm 0.03 \text{ }^\circ\text{C}$). However, the reverse occurred in summer, where mean soil
522 temperature was significantly greater on the Trenched plots ($16.9 \pm 0.03 \text{ }^\circ\text{C}$) than on the
523 Untrenched plot ($15.5 \pm 0.02 \text{ }^\circ\text{C}$). Mean air temperature at 20 cm above ground surface $3.0 \pm$
524 $0.03 \text{ }^\circ\text{C}$ in winter and $15.7 \pm 0.11 \text{ }^\circ\text{C}$ in summer. Soil moisture was significantly different
525 between the Trenched and Untrenched plots with lower soil moisture values observed on the
526 Trenched plots than the Untrenched plots in non-rainfall periods (Figure 5b).

527
528 Microbial C was not significantly different between the Trenched plots and the Untrenched
529 plots in the organic horizons of the in either the Young, Middle or in the Old vegetation (Figure
530 6). On Untrenched plots, the organic horizon microbial C was significantly greater in the

531 Young vegetation than in the Middle or the Old vegetation but there was no significant
532 difference between the Middle and the Old vegetation.

533
534 Root biomass (summed for organic horizon and 0-5 cm mineral horizon) was not significantly
535 different between the Trenched and Untrenched plots on the Young , Middle or the Old
536 vegetation (Figure 6). Additionally, the root biomass in the Untrenched plots was not
537 significantly affected by the vegetation age. There was however, a significantly greater root
538 biomass in the organic horizon than in the 0-5cm mineral horizon for all vegetation ages (data
539 not shown).

540

541 3.4 Calibration of the Model for Total Soil Respiration (R_S)

542 All model predictions of soil respiration generally followed the seasonal soil temperature
543 patterns, where the lowest respiration was recorded in winter (in February). However, not all
544 models predicted the highest respiration equally, with some models predicting peak values in
545 June, while others predicted peak values in August.

546

547 Step-wise application of variables into the different models using the Untrenched datasets
548 produced models with absolute $RMSE_C$ values that ranged from 0.30 to 2.32 (Figure 7, left
549 panel). When soil temperature (T_{soil}) was assigned as the T variable, the $RMSE$'s were
550 generally lower than when air temperature was used (T_{air}). The lowest $RMSE_C$ values were
551 obtained using the Selsted, GLMM and GLMM2 models and therefore, the LMM and LMM2
552 models are not further discussed within this results section. A selection of models and the
553 $RMSE_C$ values are provided in Table 4.

554

555 Within the GLMM and GLMM2 model formats the use of the explanatory variable T_{soil}
556 resulted in lower mean $RMSE_C$ values (0.31 to 0.49) than where T_{air} (0.35 to 0.68) was

557 included, with the exception of the T+M+P models. When all three variables T + M + P were
558 used in the GLMM format, the model over-predicted soil efflux and resulted in very high errors
559 (0.68 to 2.32); thus these were excluded from further consideration.. This did not occur with
560 the GLMM2 format. When T (both for T_{soil} and T_{air}) was the only variable used, the model
561 parameters were significant for all three Young, Middle and Old dataset.

562 The GLMM models in which all parameters were considered significant occurred on 18
563 occasions. However, the GLMM model was only significant for all three vegetation
564 communities when the T variable (either T_{soil} and T_{air}) was used alone (Figure 7, left panel).
565 The parameters in all of these significant models were considered reasonable.

566
567 Table 5 lists the parameter values for the GLMM models T_{soil} , $T_{\text{soil}} + M$ and $T_{\text{soil}} + P$. It
568 appears that adding soil moisture to a model with only temperature especially lowers R_0 while
569 hardly influencing the parameter value associated with temperature (k), whereas adding
570 photosynthesis has the reverse effect (it lowers the k -parameter, associated with temperature,
571 and does not influence R_0).

572
573 The stepwise addition of the Selsted equation resulted in $RMSE_C$ and NSE values that were
574 very similar to the GLMM and GLMM2 models. However, there were fewer occasions for the
575 Selsted models (10 occasions) than for the GLMM models where all parameters were
576 significant (Figure 7, left panel).

577

578 3.5 Calibration of the Model for Heterotrophic Soil Respiration (R_H)

579 Step-wise application of variables into the different models using the Trenched data produced
580 models with absolute $RMSE_C$ values that ranged from 0.3 to 0.44 (Figure 7, right panel). The
581 $RMSE_C$ values were lower on the heterotrophic models in which T_{soil} was used as the T
582 variable, rather than T_{air} . Similarly, the GLMM models and Selsted models resulted in RMSE

583 values that were lower than the LMM models. Therefore, only Selsted, GLMM and GLMM2
584 T_{soil} models are further within this results section.

585
586 The GLMM model which applied T_{soil} variable singly and also when applied in combination
587 with M were significant. This was not the case for the Selsted model, where only T_{soil} applied
588 alone resulted in a model in which all parameters were significant. The GLMM $T_{\text{soil}}+M$ model
589 had the highest NSE and lowest $RMSE_C$, while the Selsted and GLMM T_{soil} models had the
590 second highest NSE and second lowest $RMSE_C$. All parameters were considered to be
591 reasonable for these significant models.

592
593 Table 5 lists the parameter values for the R_H GLMM models T_{soil} and $T_{\text{soil}} + M$. Similar to the
594 models for R_S , adding soil moisture to a model with only temperature lowers R_0 while not
595 influencing the parameter value associated with temperature (k).

596

597 3.6 Model Validation

598 The calibrated models were used on the validation data for period one (September 2011 –
599 August 2012) and period two (November 2010 – August 2011, see Table 2). The resulting
600 RMSE validation values ($RMSE_{V1}$ and $RMSE_{V2}$) were then compared to the RMSE calibration
601 values ($RMSE_C$). A selection of these results, are shown in Table 4. The R_S models which had
602 the lowest $RMSE_C$ and $RMSE_V$ values used the GLMM format with T_{soil} as a single variable,
603 with $T_{\text{soil}} + P$ and with $T_{\text{air}} + P$. Of these, the GLMM T_{soil} model and the GLMM $T_{\text{air}} + P$
604 model were the only ones where all parameters were significant for all vegetation ages. The R_S
605 models which performed the worst in the validation phase also used the GLMM format and
606 included the T variable (both T_{soil} and T_{air}) in combination with M + P. A large part of the
607 unexplained variance in the models with T+M+P appears to be due to location-effects (when

608 the error of the multi-level models models are evaluated with location as fixed effects, the
609 misfit is in fact quite small). .

610
611 The average ratio of $RMSE_{V1} : RMSE_C$ in the R_S models was 1.5 and the ratio of $RMSE_{V2} :$
612 $RMSE_C$ was 1.3. The ratio validation error:calibration error measures the degree at which a
613 model can generalize the results for a specific site (or experiment) to other locations or
614 conditions. If the ratio is large, it indicates that the calibration data is unrepresentative or that
615 the model for which the ratio is calculated is over-parameterized. In our experience, ratio's
616 smaller than 2 are quite acceptable and we therefor think that the calibration data set is
617 representative and that the models that were applied are not over-parameterized. The ranges for
618 $RMSE_{V1}$ (approx. 0.5 to 2.8) and $RMSE_{V2}$ (approx. 0.45 to 3.5) were comparable, with the
619 same four of the fifty-seven R_S models (LMM and GLMM, using T,M and P, for both air and
620 soil temperature) leading to very high values for $RMSE_{V1}$ as well as for $RMSE_{V2}$. For the R_S
621 models, there was very a high correlation (>0.99) between $RMSE_C$, $RMSE_{V1}$ and $RMSE_{V2}$. It
622 should be noted that the validation was done only for the old vegetation.

623
624 The R_H models produced relatively low $RMSE_V$ values for all combinations and formats
625 (<0.49).The R_H models which produced the lowest $RMSE_V$ values were the GLMM format
626 with $T_{soil} + M$, the GLMM and Selsted format with T_{soil} alone, and the LMM format with T_{soil}
627 + M. The ratio of $RMSE_{V1} : RMSE_C$ in the R_H models was, on average, 1.15. There was a
628 very strong correlation between $RMSE_C$ and $RMSE_{V1}$ (Pearson correlation coef. of 0.997).

629
630 *3.7 Model Selection*

631 Following the rationale described in the methodology to select the best predictive models, both
632 the Selsted and GLMM models using only T_{soil} or using $T_{soil} + M$ are selected as the best
633 predictive models for R_H . However, the GLMM models provide more realistic confidence

634 bounds (by taking the property of repeated measurements in our data into account). Therefore
635 we prefer to use the GLMM model for predictive purposes. Furthermore, the RMSE,
636 parameter values and predictions of the T_{soil} and $T_{\text{soil}} + M$ models were similar. So the most
637 parsimonious model (using only T_{soil} as predictor) was selected to be used for further
638 predictions.

639
640 For the R_S models, the best predictive models were the Selsted and GLMM models, using only
641 T_{soil} or using $T_{\text{soil}} + P$. As with the R_H models, we have selected the GLMM models for
642 prediction rather than the Selsted model. The differences between the GLMM T_{soil} and GLMM
643 $T_{\text{soil}} + P$ models (with respect to $RMSE_C$, $RMSE_{V1}$ and $RMSE_{V2}$) were minor. So choosing the
644 most parsimonious model for R_S also leads to a model with T_{soil} as only predictor variable. The
645 GLMM T_{soil} models for R_H and R_S were used to predict soil respiration over the length of the
646 study period (Table 5 and Figure 8).

647
648 *3.8 Autotrophic Soil Respiration*
649 Autotrophic soil respiration was determined by subtracting the model predicted heterotrophic
650 soil respiration results from the total soil respiration results in each vegetation community (R_S
651 $- R_H = R_A$; Figure 9). Soil R_A was approximately zero on the Middle and Old communities in
652 winter. The greatest R_A was predicted to occur on the Young community in the summer
653 months, with a maximum in July when approximately 55% of soil respiration was attributable
654 to autotrophic sources. In this same time period, approximately 45% and 37% of soil
655 respiration on the Middle and Old communities, respectively, was attributable to autotrophic
656 sources.

657
658 *3.9 Annual Carbon Loss Estimates*

659 Based on model predictions, annual C loss from R_S was significantly greater on the Young
660 community ($650 \text{ gC m}^{-2} \text{ year}^{-1}$) than either the Middle ($462 \text{ gC m}^{-2} \text{ year}^{-1}$; $p=0.048$) or the Old
661 ($435 \text{ gC m}^{-2} \text{ year}^{-1}$; $p=0.029$) communities (Figure 11). There was no significant difference
662 between R_S annual C loss on the Middle and Old communities ($p=0.39$). The annual losses
663 from R_A and R_H were approximately equal in the Young vegetation (50% was R_A), but it was
664 calculated that there was greater C loss from R_H than from R_A sources in both the Middle and
665 the Old communities (30% and 26% was R_A , respectively). The soil C loss was plotted against
666 community age, using a 'time for space' chronosequence approach to approximate changes in
667 C loss over a 30 year period. Year zero represents the bare soil which would be expected
668 following a vegetation cutting cycle. In this case, all soil respiration would be expected to be
669 from R_H , as there are no plant roots respiring and the lack of vegetation cover would result in
670 more variable soil temperatures, as observed in the bare Trenching plots. Therefore, C loss in
671 year zero was predicted using the more variable Trenched soil temperatures ($350 \text{ gC m}^{-2} \text{ year}^{-1}$)
672 ¹). Soil temperatures were less variable under plant cover and so the Untrenched temperatures
673 were used in the model to predict annual R_H C loss ($322 \text{ gC m}^{-2} \text{ year}^{-1}$) where plant cover was
674 present.

675

676 4.0 Discussion

677 Carbon loss from soil respiration was greatest on the Young community and root-associated
678 respiration contributed approximately equally to the annual C sum as was contributed by
679 microbial respiration. As the community age increased, the annual C loss from soil respiration
680 decreased and this change was driven by the decreasing contribution of root respiration.

681

682 The following sections have been grouped around discussion of the soil respiration, of the
683 trenching effects, the modeling process and finally a discussion of the annual model
684 predictions.

685

686 *4.1 Soil Respiration*

687 Heterotrophic respiration rates were not statistically different between the three communities
688 and this was consistent with the original hypothesis. In general, CO₂ effluxes from microbial
689 decomposition are determined by the quantity and quality of available substrate, the soil
690 temperature and other conditions that control decomposer activity (Kirschbaum 2006). This
691 was consistent with trial observations, as there was no difference between the quantity of
692 available substrate in the different communities prior to trenching, that is, soil C stocks to
693 10cm soil depth (Kopittke et al. 2012), and no soil temperature or soil moisture pattern
694 differences between the Trenched plots. However, the quality of the organic matter and
695 recently deposited litter (prior to trenching) was not known. The proportion of lignin in the
696 litter could be expected to increase with increasing community age, as woody stem growth
697 increases with increasing plant age (Gimingham 1985). Increasing the lignified material in
698 organic matter results in slower decomposition rates (Filley et al. 2008, Kalbitz et al. 2003).
699 However, as no differences in respiration were observed, it is possible that the rapid
700 decomposition of the labile organic matter masked any underlying differences (if indeed
701 present) in the more recalcitrant pools.

702

703 The differences observed between total soil respiration on the community ages was not
704 associated with heterotrophic respiration and therefore by elimination ($R_S - R_H = R_A$), was
705 associated with autotrophic respiration. The greater total soil respiration on the Young
706 community indicated that the Young Calluna plant roots were more actively respiring than on
707 the Middle or Old communities. These higher rates corresponded to a higher P_G and supported
708 the hypothesis that the youngest plants, which were in a 'net biomass gain' phase of growth
709 (Gimingham 1985), had the highest plant activity with greater allocation of carbon to the roots.

710

711 However, *Calluna* biomass was not the only contributor to P_G . Mosses also contributed to P_G ,
712 with almost double the moss biomass on the Young community than on the Middle or Old
713 communities. Although moss did not contribute directly to R_A , as it lacks a root system, this
714 mismatch in aboveground and belowground rates is likely to have introduced additional bias
715 when including P_G as a variable in the R_S models. This study did not quantify the separate P_G
716 contributions of moss and *Calluna*. However, based on the preliminary data from in a trial in
717 May 2012, the Young *Calluna* plants were approximately 2.5 times more photosynthetically
718 active than the Middle and Old *Calluna*; therefore, P_G would still provide a measure of the
719 plant activity for each community.

720
721 The peak respiration values recorded in March 2012 corresponded to the first warm period in
722 which air temperatures exceeded 15 °C, following from a severe frost (-20 °C) in February
723 2012. These extreme values were most likely associated with the death of fine roots and
724 microbial populations, followed by the rapid recovery of microbial populations which lead to
725 short term fluxes of CO₂ from the soil (Matzner and Borken 2008, Sulkava and Huhta 2003). In
726 addition, *Calluna* litter fall measurements on the Old vegetation have shown peak fall rates
727 occur approximately in January and old flowers are the dominant litter type (unpublished data
728 from the adjacent long term trial). This unignified litter is likely to provide a rapidly
729 decomposable energy source for microbial populations and may have contributed to the general
730 CO₂ efflux peak that was observed in spring.

731
732 The observed total soil respiration rates were comparable to other *Calluna* heathland
733 communities, such as in Brandbjerg, Denmark and a hydric *Calluna* heathlands in the Northern
734 Pennines, England (Heinemeyer et al. 2011, Selsted et al. 2012). The mean summer total soil
735 respiration rates in Brandbjerg ranged between 1.2 and 2.9 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (2008 and 2006,
736 respectively) and this was within the same range observed at Oldebroek in the summer of 2012.

737
738 Total soil respiration of other heathlands far exceeded the observations recorded at the
739 Oldebroek study site. In the mesic heathland at Mols in Denmark, mean summer total soil
740 respiration rates were $16 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ in 2003 (Sowerby et al. 2008), which was
741 approximately 5.8 times the mean summer respiration observed on the Young community at
742 Oldebroek in 2012. This large difference is most likely associated with the age of the
743 vegetation and possibly differences in vegetation composition rather than soil differences. The
744 soil type at Mols was similar, but the heathland experienced a heather beetle attack in 1999,
745 which mainly resulted in *Deschampsia* regrowth of young *Calluna* plants (four years old).
746 Similarly, total soil respiration on a hydric *Calluna* heathland at Clocaenog in Wales was also
747 consistently greater in every season than the Young community, even when the peak values of
748 $5.6 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (Young community) and $7.6 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ (Clocaenog) were
749 compared (Emmett et al. 2004).

750

751 *4.2 Trenching Effect*

752 The soil temperature difference observed between Trenched and Untrenched plots is likely to
753 be a function of the *Calluna* plants providing shade and the thick moss layer providing
754 insulation at the soil surface. These two factors are hypothesized to have regulated soil
755 temperature in the Untrenched plots but not in the Trenched plots where the aboveground
756 vegetation had been removed. Temperature determines the rate of organic matter
757 decomposition and CO_2 production (Kirschbaum 2006). Therefore, the Trenched plots
758 (producing autotrophic respiration) and Untrenched plots (producing total soil respiration) were
759 exposed to different temperature regimes. In principle, R_A is the difference between the R_S and
760 R_H , however, under these differing temperature conditions, the observed results cannot be
761 directly used to calculate autotrophic respiration. This temperature difference confirmed the

762 necessity of using soil respiration models to predict both total soil respiration and autotrophic
763 respiration for the same temperature range.

764
765 Soil moisture patterns were also observed to differ between the Trenched and Untrenched
766 plots, where the Trenched plots were drier than the Untrenched plots in non-rainfall periods.
767 This is contrary to other studies in which trenching was observed to result in higher soil
768 moisture than the control plots (Hanson et al. 2000). It is hypothesized that vegetation removal
769 led to a loss of shade cover and this resulted in the organic layer and litter layer being exposed
770 to greater evaporation rates. This hypothesis is supported by visual observations of a drier and
771 cracked organic layer on the Trenched plots. The respiration models being tested incorporated
772 a soil moisture parameter so that any moisture effect could be assessed.

773

774 *4.3 Model Evaluation*

775 All models followed generally the same pattern in the prediction of minimum effluxes in the
776 winter, maximum effluxes in the summer and the highest autotrophic respiration for the Young
777 community (see Figure 8, showing only the results for GLMM). However, the specific fit to the
778 observations (as summarized by RMSE varied between the different models (see Figure 7).

779

780 The RMSE values for all models using T_{soil} were consistently lower than those using T_{air} .
781 Additionally, the Selsted and GLMM models led to lower RMSE values and a lower spread in
782 RMSE between the different vegetation ages than the other models. However, only the models
783 using T_{soil} alone were significant for all community ages. These results indicated that the
784 complex parameterization of soil moisture and biomass effects in the Selsted model were not
785 suitable for our site. This difference in model fitness may be due to site differences, such as the
786 % grass cover or topsoil thickness, between the Brandbjerg heathland in Denmark (for which
787 the Selsted model was developed) and Oldebroek. In future research it would be interesting to

788 pool the two data sets and re-evaluate the models to see whether a model can be identified that
789 is adequate for both sites.

790
791 Both for the R_H and R_S models, the RMSE values were very similar and highly correlated
792 between the calibration and validation phases. Therefore, these models were considered stable
793 and it can be assumed that the model predictive uncertainty was mainly due to parametric
794 uncertainty. Also, the very high correlation between model prediction errors for calibration and
795 validation indicates that the calibration and validation data contain data with a very similar
796 information content. When the model misfit is analysed in greater detail (see Appendix D),
797 several structural deviations of the residual are seen over time (the model residual is not
798 uncorrelated but contains information which is not captured by the model). This misfit is not
799 apparent with regard to temperature. In our view, the most plausible explanation for the
800 structure in the model residuals is that one or more important covarying variables are lacking in
801 the models that were parameterized so far.

802
803 From the variables assessed and available for inclusion in our model selection process, very
804 similar fits of the observed data was provided by models using soil temperature (possibly in
805 combination with soil moisture or plant activity). However, the model with soil temperature as
806 only variable contained less parameters and was therefore preferred for predictive purposes. The
807 application of only a temperature function to model soil respiration data has previously been
808 questioned since, as already discussed, other factors such as soil moisture limitation of
809 microbial processes and the C allocation via plant roots are all reported to influence soil
810 respiration rates (Davidson et al. 2006, Rustad et al. 2000). However, our results indicated that
811 soil moisture and plant activity (Calluna biomass, P_G , microbial biomass and root biomass)
812 were not significant variables for our site. To examine this further, it is first considered if it is
813 possible that some of the measured variables would have been significant, if the data had been

814 measured differently. Secondly, other variables are considered that have been used in similar
815 soil respiration studies and may have improved model fit.

816
817 Soil moisture was measured but was not a significant parameter. For the R_H model, soil
818 moisture has been shown to impact microbial respiration (and therefore R_H) only at extremely
819 low water contents when desiccation stress becomes important for microbial substrate supply
820 (Davidson et al. 2006). It is possible that in our study the soil did not reach these desiccation
821 stress levels, thus resulting in a non-significant soil moisture parameter for the R_H models. For
822 the R_S model, *Calluna* plants appear to be resilient to water stress and heathlands can withstand
823 quite severe summer droughts, if annual rainfall is high enough to compensate for the drought
824 (Loidi et al. 2010). Additionally, the Oldebroek heathland is established on a free-draining,
825 sandy soil that has relatively low stored soil moisture in the mineral soil. The majority of the
826 *Calluna* roots were identified within the nutrient-rich, organic layer of the soil and this is also
827 where the largest proportion of the soil moisture is stored (see Table 1). However, continuous
828 soil moisture measurements in the organic layer are very difficult due to instrumentation
829 constraints (Schaap et al. 1997). Because of this, it is likely that a large proportion of the soil
830 respiration response to reductions in soil moisture occurred in the organic horizon, and this was
831 not able to be quantified with the current technology. Therefore, continuous soil moisture
832 measurements in the organic horizon may have improved model fit.

833
834 Other variables from published soil respiration models that could be considered have included
835 using relative PAR with soil temperature and soil moisture (Caquet et al. 2012). In our study,
836 PAR was included in the initial model screening process as a single predictor variable and as a
837 predictor variable together with temperature. However, neither of these models resulted in a
838 better fit than soil temperature alone and therefore, PAR was not included in further model
839 testing.

840

841 Alternatively, another plant variable which has been considered in other respiration models is
842 the rate of litter decomposition (Kutsch and Kappen 1997, Kutsch et al. 2010). However, it is
843 unclear from these studies whether the addition of litter decomposition to the soil temperature
844 and moisture model resulted in a better model fit, as the parameter significance was not
845 reported. Soil temperature has been found to generally have a good relationship with organic
846 matter decomposition rates (Davidson and Janssens 2006) and therefore it is hypothesized that
847 a litter decomposition variable would not explain significantly more variability than already
848 explained by soil temperature. Other plant litter variables, such as litter fall rates, are also often
849 included in dynamic models as they provide an important feedback into the carbon cycle and
850 substrate available for decomposition (Keenan et al. 2012). Litterfall results were not available
851 for the Young and Middle communities, although litter data was collected on the Old
852 community validation plots between March 2011 and February 2012 (unpublished results).
853 The maximum litterfall rate occurred in January ($8 \text{ g m}^{-2} \text{ month}^{-1}$) and the minimum in
854 February ($2 \text{ g m}^{-2} \text{ month}^{-1}$) with gradually declining rates recorded from March to November (7
855 to $5 \text{ g m}^{-2} \text{ month}^{-1}$). This pattern did not correspond to the observed soil respiration rates and
856 suggests R_H is more closely associated with the temperature than with litterfall patterns.
857 However, if sufficient litterfall data had been available for inclusion as a variable with soil
858 temperature, it may have improved R_H model fit by explaining additional data variation.

859

860 Root maintenance (as a function of root nitrogen concentrations) and root growth have also
861 been included in soil respiration models. In a study in Tennessee USA, this model with root
862 variables was able to describe more of the biological dynamics than the other models tested
863 although it was still not capable of capturing all the data variation across the different study
864 treatments (Chen et al. 2011). Root dynamics provide a direct measure of root activity and, if it

865 had been measured at the Oldebroek site, may have explained more variance than the
866 photosynthetic rates.

867
868 A further layer of complexity to the discussion is that model results may be influenced by a
869 suboptimal measurement integration volume or integration time, as well as the alignment in
870 space and time of different measurements. Problems of this kind ('scale problems') are
871 common in the natural sciences and are an important source of model error, thus are considered
872 as the most important challenge in ecology (Blöschl and Sivapalan 1995, Wiens 1989). An
873 example of a data alignment problem in our study is the collection of soil respiration
874 measurements on different days than the photosynthesis measurements, which required
875 intermediate data processing for photosynthesis (viz. Figure 3). Also, soil temperature was
876 measured at a depth (5 cm), whereas the soil respiration was an integral measurement over a
877 soil column (eg. Reichstein and Beer 2008). There may also be a lag time present within the
878 data, where plant growth on one day does not immediately correspond to root respiration
879 (Gomez-Casanovas et al. 2012, Kuzyakov and Gavrichkova 2010), which our non-continuous
880 data would not have been able to detect. These trial design aspects may have resulted in small
881 data mismatches and it is possible that the model calibration and validation results would have
882 improved if the resolution and alignment of the data had improved.

883
884 The model selection process resulted in a model that used T_{soil} alone, which is arguably the
885 simplest variable. However, this 'simple' result does not negate the use of a detailed selection
886 procedure, as the process also highlighted that the current variables measured were not
887 adequate to model all the variation observed in the R_S (and therefore R_A) data. This is an
888 important outcome of this study, as many studies include processes that are theoretically
889 associated with soil respiration but the model variables are not assessed for significance and
890 may not explain any additional data variation. This practice leads to a publication bias

891 (Dieleman and Janssens 2011). The use and reporting of a full data pre-processing and
892 modeling workflows that apply sound scientific procedures, which also report the ‘negative’ or
893 ‘less interesting’ results, helps to avoid such a publication bias.

894
895 It is not possible to measure all ecosystem processes on an experimental trial due to practical
896 constraints and it is not always possible to know which measured process would improve the
897 model fit, although pre-planning field experiments based on the models we wish to use may
898 assist in this process. This finding supports the discussion presented by Subke and Bahn (2010)
899 on the ability to use the immeasurable to predict the unknown. In this study, although one or
900 more important covarying variables were lacking (and model fit would have improved had
901 these been measured), it is worth considering that soil temperature was likely to also be related
902 to seasonal plant activity and may simply be the overwhelming driver of soil respiration in this
903 system. Therefore, in the absence of other variables, the T_{soil} variable was sufficient to explain
904 most of the seasonal variation of R_S . Similar findings have been reported in other studies,
905 where site differences in R_S were largely determined by plant productivity but since both R_S
906 and P_G fluxes increased with temperature, it was concluded that the soil temperature typically
907 sufficed to explain R_S in non-drought ecosystems (Bahn et al. 2010a, Janssens et al. 2001,
908 Reichstein et al. 2003).

909
910 We think that the findings from this empirical study (on the basis of static models) can also be
911 used to investigate or test dynamic soil respiration models (which are typically parameter-rich
912 and often model more than only soil respiration in isolation). First of all, there are many
913 dynamic soil respiration models which do effectively contain a respiration equation similar to
914 those used in this study (e.g. Keenan et al. 2012, Kutsch et al. 2010). In those models, a more
915 complex equation could simply be replaced by a simpler one, if that seems more appropriate
916 for the case at hand (e.g. because it leads to less parameters to calibrate or more stable model

917 behaviour). Otherwise, if the way of modeling respiration would be incompatible with the
918 static equations in this study (and results cannot directly be translated), we think that the point
919 of evaluating different models or model components on calibration and validation data sets (i.e.
920 the method promoted in this study), deserves attention – not because it is new, but because it is
921 currently uncommon in the area of soil respiration research. A lack of critical model evaluation
922 limits progress.

923

924 In nature, many interactions can occur and when our field trials don't test these interactions, it
925 is not possible to incorporate them into long-term model predictions. Therefore, it is necessary
926 to develop field trials which incorporate this increased complexity, as suggested by Dieleman
927 et al. (2012). However, if early consideration isn't given to the models that we later want to fit
928 to the data (and the data required to rigorously test the models), then increasing the complexity
929 of field experiments will not necessarily provide us with better predictions of these
930 interactions. Therefore, attention should be given to the trial layout, variable selection,
931 measurement intensity and model selection process prior to the start of a trial to determine if
932 they will provide the appropriate data for model predictions. Consideration also needs to be
933 given to the cost associated with obtaining the appropriate measurements, in terms of collection
934 frequency, method accuracy and overall outcomes of the project. In some cases, it may be that
935 using a proxy such as soil temperature (or even air temperature for rough estimations) with the
936 soil respiration observations is a suitable substitute in models in the absence of suitable and
937 significant variables.

938

939 *4.4 Annual C Loss and Links to Global Change*

940 Our model interpolations identified an annual C loss from R_S that was at the lower end of the
941 range identified on the Danish heathland ecosystem of 672 - 719 g C m⁻² year⁻¹ (Selsted et al.
942 2012). To place this within a broader European context, the heathland soil respiration is within

943 the same range as temperate forest ecosystems, which have been reported between 430 g C m⁻²
944 year⁻¹ (Belgium) and 859 g C m⁻² year⁻¹ (Germany) (Bahn et al. 2010a, Khomik et al. 2009,
945 Raich and Schlesinger 1992). In contrast, the heathland is at the lower end of the scale for
946 annual C loss in comparison to temperate grasslands, which ranged between 729 g C m⁻² year⁻¹
947 (Germany) and 1988 g C m⁻² year⁻¹ (Switzerland) (Bahn et al. 2010a).

948
949 The study also identified a change in soil respiration with an increasing age of heathlands. Soil
950 R_A provided the largest change over time, from a complete absence on bare soil to a maximum
951 at the 12 years and then decreasing up to the maximum studied age of 28 years. A similar
952 relationship between soil respiration and vegetation age has been previously found for forest
953 stands, where the younger stands had significantly higher respiration rates than the more
954 mature sites (Saiz et al. 2006, Wang et al. 2011).

955
956 Within the last 50 years, the cutting, burning and grazing cycles on heathlands have not
957 occurred as frequently or as regularly as during the intensive agricultural periods of past
958 centuries (Webb 1998). Management of heathlands is required to maintain these cultural
959 landscapes and in past times this management occurred on a 3 – 4 year cycle (Webb 1998).
960 Currently, this cycle length has extended or is non-existent (Diemont and Heil 1984, Wessel et
961 al. 2004). From the perspective of optimizing C uptake and minimizing C output, having an
962 understanding the C dynamics of these ecosystems allows us to determine the optimum time to
963 cut the vegetation, thus contributing to global C emission mitigation measures. Based on the
964 presented data and a preliminary assessment of other fluxes within the system, it is
965 hypothesized that the younger vegetation will be a C sink and the older vegetation will be a C
966 source, indicating management of these cultural landscapes should occur on relatively shorter
967 cycle lengths if C emission mitigation is of concern. This hypothesis is based on the relative
968 rates of P_G and R_S on the communities and could be further investigated by applying a similar

969 modeling approach to the ER and NEE data (used in this paper only to calculate P_G) to
970 determine annual sums of C uptake and C loss from the ecosystem.

971

972 **Conclusions**

973 The main aim of this paper was to investigate one year of total and heterotrophic soil
974 respiration results and, with the use of a model chosen through a modeling selection process, to
975 calculate annual soil C losses. Based on the known relationship between soil microbial
976 decomposition, soil temperature and soil moisture, it was hypothesized that soil temperature
977 and soil moisture would be significant variables for the R_H model. In addition, based on the
978 belowground allocation of plant metabolites to root processes, it was hypothesized that soil
979 temperature, soil moisture and a measure of plant activity would contribute significantly to the
980 R_S models for all three communities.

981

982 During the model selection process, it became clear that the hypotheses were not supported:
983 that is, soil moisture was not significant in the 'best' R_H models and neither were soil moisture
984 nor plant activity variables significant in the 'best' R_S model. However, a close look at the
985 residuals of the best models did show autocorrelated variability. This indicated that one or
986 more ecosystem variables besides photosynthesis, plant biomass, PAR, root biomass, or
987 microbial biomass were probably important when trying to explain soil respiration in this
988 system. In the absence of these covariates, soil temperature sufficed for soil respiration
989 prediction in the heathland ecosystem. of our study. For our future experimental work this
990 model based on soil temperature may act as a null model against which the performance of
991 other models can be compared. The annual sums of soil C loss were calculated using the
992 GLMM model with T_{soil} as the only variable for R_S (Young: $649 \text{ g C m}^{-2} \text{ year}^{-1}$; Old: 434 g C
993 $\text{m}^{-2} \text{ year}^{-1}$) and R_H ($322 \text{ g C m}^{-2} \text{ year}^{-1}$).

994

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1004 access to the field site.

1005

1006 **Captions**

1007 **Table 1** Description of the Oldebroek Trial Location

1008

1009 **Table 2** Description of the data used for model calibration and validation

1010

1011 **Table 3** The Models to estimate R_S and R_H ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). The explanatory variables are T
1012 (models using air temperature at 20 cm above ground surface and soil temperature at 5 cm
1013 below ground surface are evaluated) and M, B, P as defined in Eq. (2) to (4). The model
1014 parameters are R_0 , k , a , b and c and the units vary per model. R_0 is always in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$.
1015 Parameter k is in $^\circ\text{C}^{-1}$ for Selsted, GLMM and GLMM2 models, and in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1} \text{ }^\circ\text{C}^{-1}$
1016 for LMM and LMM2. The parameters a , b and c are dimensionless for Selsted, GLMM and
1017 GLMM2 models and are in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ for the LMM and LMM2 models.

1018

1019 **Table 4** The residual standard deviation with RMSE_C and RMSE_V (Validation Type I and II)
1020 values for a selection of GLMM models. Only models in which all variables were significant

1021 are shown. See Table 2 for definition of validation types, Table 3 for model definitions and
1022 Table 5 for parameter values of the models shown here. Results for cross-validation and with
1023 additional error metrics are provided in Appendix C..

1024
1025 **Table 5** Optimal parameter values of total and heterotrophic respiration models for the Young,
1026 Middle and Old communities (GLMM T_{soil} Model), with 95% confidence intervals for the
1027 parameters between brackets. See text and Table 3 for parameter explanations. For R_H , model
1028 $T_{\text{soil}+P}$ does not exist. The values of the different parameters are given with different numbers
1029 of significant digits to reflect the uncertainty in the corresponding variable. The shaded cells
1030 indicate parameter values that are not significant at the 0.05 significance level. Diagnostic plots
1031 for the models listed in this table are shown in Appendix D.

1032
1033 **Figure 1** The experimental layout showing the nested design of the Untrenched plots (“U”),
1034 Trenched plots (“T”) and the Trenched Validation plots (▨) in the Young, Middle and Old
1035 vegetation communities (not to scale). The Untrenched Validation plots (◻) are shown in the
1036 Old community. Gross photosynthesis measurement locations are shown with a “P_G”. The
1037 boundaries of the three communities are represented by a dotted grey line.

1038
1039 **Figure 2** Schematic Representation of the Data Analysis Workflow.

1040
1041 **Figure 3** Measures of Plant Activity for the Young, Middle and Old communities, showing (a)
1042 mean Calluna Biomass (kg m^{-2}) obtained in April 2011 during trenching activities ($n=12$); and
1043 (b) C uptake by Photosynthesis ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) obtained between August 2011 and August
1044 2012 ($n=9$) with observations represented by symbols and the mean curves (loess curves)
1045 represented by lines. On plot (a), the letters represent statistical significance and the SEM bars
1046 are shown for each mean value.

1047
1048 **Figure 4** Soil Respiration ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) on three ages of vegetation for the (a) total soil
1049 respiration as represented by the Untrenched plots; and (b) heterotrophic soil respiration, as
1050 represented by the Trenched plots from September 2011 until August 2012 (n = 4 per age per
1051 sampling event). For plot (b), the Young community SEM bar in March 2012 extends outside
1052 the graphical boundaries to $6.79 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$.

1053
1054 **Figure 5** Environmental parameters for September 2011 – August 2012, showing (a) hourly
1055 temperatures ($^{\circ}\text{C}$) of the air at 20cm above ground surface and of the soil at 5cm below ground
1056 surface; and (b) mean daily soil moisture ($\text{m}^3 \text{ m}^{-3}$) at 5cm below ground surface for the
1057 Trenched Plots and the Untrenched Plots. Periods of frozen soil moisture are indicated by
1058 shading (■).

1059
1060 **Figure 6** A comparison of the Trenched plots (n=12) and Untrenched plots (n=12) for (a) mean
1061 microbial C biomass (mg C g C^{-1}) in the organic horizon and (b) mean root biomass (g m^{-2}) in
1062 the summed (organic + 0-5cm mineral) horizons shown for the three ages of heathland
1063 vegetation. Different letters above the bars represent statistical significance and the SEM bars
1064 are shown for each mean value.

1065
1066 **Figure 7** Comparison of RMSE_C values for models of (a) total soil respiration data
1067 (Untrenched plots) and (b) heterotrophic soil respiration data (Trenched plots). The models
1068 tested are listed on the left side of the figure. The explanatory variables within each model are
1069 listed on the y axis and are abbreviated as: T = temperature (soil or air $^{\circ}\text{C}$ as indicated), M =
1070 soil moisture, B = relative biomass, P = relative photosynthesis. The “*” indicates that all
1071 model parameters were significant for one of either “Y” (Young), “M” (Middle) or “O” (Old)

1072 vegetation community models. The SEM bars on the total soil respiration means were
1073 calculated from the $RMSE_C$'s of the three community ages. SEM bars could not be calculated
1074 for the heterotrophic models.

1075
1076 **Figure 8** Predicted and observed soil respiration on the Young, Middle and Old community
1077 “Untrenched” plots (total soil respiration: $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) and the “Trenched” plots
1078 (heterotrophic soil respiration: $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) calculated with the GLMM model using the
1079 T_{soil} explanatory variable. The observed values from 21 March 2012 are excluded from these
1080 plots.

1081
1082 **Figure 9** Predictions of soil respiration for (a) Young community, (b) Middle community and
1083 (c) Old community calculated using the GLMM model with the T_{soil} explanatory variable.

1084
1085 **Figure 11** Estimated annual C loss from total soil respiration (R_S), heterotrophic soil
1086 respiration (R_H) and autotrophic soil respiration (R_A) as predicted by the GLMM T_{soil} model.
1087 Year 0 is represented by respiration from bare soil, Year 12 by the Young community, Year 19
1088 by the Middle community and Year 28 by the Old community. Mean prediction values are
1089 provided with the bars representing the 95% Confidence Intervals.

1090
1091 **Appendix A** Details of the P_G Measurements
1092 **Appendix B** Details of the Soil Moisture Model
1093 **Appendix C** Dealing with Additional Validation Methods and Error Metrics
1094 **Appendix D** Diagnostic plots for selected models
1095
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1309 decomposition in North peatlands: implications for thermal responses of carbon cycling to
1310 global warming, *Environmental Geology*, 58, 499-508, 2009.
1311

1312 **Table 1** Description of the Oldebroek Trial Location

Category	Description																																																																															
Location	ASK Oldebroek, Oldebroekse heide, Province of Gelderland, The Netherlands																																																																															
Co-ordinates	52°24'N 5°55'E																																																																															
Elevation	25 m ASL																																																																															
Slope	2%																																																																															
Climate	Temperate, humid.																																																																															
Rainfall	1018 mm																																																																															
Air Temperature	Average for January: 2.0 °C July: 17.8 °C Annual: 10.1 °C																																																																															
Plant Species	<i>Calluna vulgaris</i> , <i>Molinia caerulea</i> , <i>Deschampsia flexuosa</i> , <i>Pinus sylvestris</i> , <i>Betula pendula</i> , <i>Empetrum nigrum</i> , <i>Juniperus communis</i> , <i>Hypnum cupressiforme</i> Hedw, <i>Hypnum jutlandicum</i> Holmen et Warncke, <i>Dicranum scoparium</i> Hedw.																																																																															
Soil	Haplic Podzol with mormoder humus form																																																																															
Parent Material	Coversand, fluvioglacial deposits																																																																															
Soil Chemistry^a	<table border="1"> <thead> <tr> <th rowspan="2">Name</th> <th colspan="3">Organic Horizons</th> <th colspan="4">Mineral Horizons</th> </tr> <tr> <th>L+F</th> <th>H</th> <th>Ae</th> <th>Bs</th> <th>1BC</th> <th>2BC</th> <th>C</th> </tr> </thead> <tbody> <tr> <td>Depth (cm)</td> <td>+8.0 to +1.4</td> <td>+1.4 to 0</td> <td>0 to 5.5</td> <td>5.5 to 13</td> <td>13 to 21</td> <td>21 to 27</td> <td>>27</td> </tr> <tr> <td>pH</td> <td>3.7</td> <td>3.9</td> <td>3.9</td> <td>4.0</td> <td>4.5</td> <td>4.4</td> <td>4.9</td> </tr> <tr> <td>EC ($\mu\text{S cm}^{-1}$)</td> <td>197.9</td> <td>92.0</td> <td>88.7</td> <td>73.2</td> <td>32.3</td> <td>46.3</td> <td>30.8</td> </tr> <tr> <td>NO₃ ($\mu\text{mol kg}^{-1}$)</td> <td>646.6</td> <td>216.2</td> <td>20.2</td> <td>62.4</td> <td>22.1</td> <td>47.6</td> <td>13.1</td> </tr> <tr> <td>PO₄ ($\mu\text{mol kg}^{-1}$)</td> <td>1589</td> <td>126</td> <td>4.6</td> <td>1.4</td> <td>0.1</td> <td>0.1</td> <td>0.1</td> </tr> <tr> <td>C/N ratio</td> <td>40.4</td> <td>17.7</td> <td>27.7</td> <td>18.0</td> <td>16.7</td> <td>18.5</td> <td>11.7</td> </tr> <tr> <td>Soil Moisture^b %</td> <td>104.8</td> <td>47.1</td> <td>15.7</td> <td>14.9</td> <td>6.3</td> <td>6.3</td> <td>6.3</td> </tr> <tr> <td>Texture</td> <td colspan="2">-</td> <td colspan="2">fine grain sand</td> <td colspan="3">medium to coarse grain</td> </tr> </tbody> </table>	Name	Organic Horizons			Mineral Horizons				L+F	H	Ae	Bs	1BC	2BC	C	Depth (cm)	+8.0 to +1.4	+1.4 to 0	0 to 5.5	5.5 to 13	13 to 21	21 to 27	>27	pH	3.7	3.9	3.9	4.0	4.5	4.4	4.9	EC ($\mu\text{S cm}^{-1}$)	197.9	92.0	88.7	73.2	32.3	46.3	30.8	NO ₃ ($\mu\text{mol kg}^{-1}$)	646.6	216.2	20.2	62.4	22.1	47.6	13.1	PO ₄ ($\mu\text{mol kg}^{-1}$)	1589	126	4.6	1.4	0.1	0.1	0.1	C/N ratio	40.4	17.7	27.7	18.0	16.7	18.5	11.7	Soil Moisture ^b %	104.8	47.1	15.7	14.9	6.3	6.3	6.3	Texture	-		fine grain sand		medium to coarse grain		
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1313 ^a Water extraction of 1:5 for organic horizons and 1:1 for mineral horizons

1314 ^b obtained following a rainfall event and reported as a percentage (g per g dry weight soil)

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1317 **Table 2** Description of the data used for model calibration and validation

Modelling Stage	Total Soil Respiration Models (R_s)	Heterotrophic Respiration Models (R_H)
Calibration	<i>Data:</i> Untrenched plots <i>Dates:</i> September 2011 – August 2012	<i>Data:</i> Trenched plots <i>Dates:</i> September 2011 – August 2012
Validation (Type I)	<i>Data:</i> Untrenched Validation plots <i>Dates:</i> September 2011 – August 2012	<i>Data:</i> Trenched Validation plots <i>Dates:</i> September 2011 – August 2012
Validation (Type II)	<i>Data:</i> Untrenched Validation plots <i>Dates:</i> November 2010 – August 2011	—

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1320 **Table 3** The Models to estimate R_S and R_H in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$. The explanatory variables are T (models using air
1321 temperature at 20 cm above ground surface and soil temperature at 5 cm below ground surface are evaluated) and
1322 M, B, P as defined in Eq. (2) to (4). The model parameters are R_0 , k , a , b and c and the units vary per model. R_0 is
1323 always in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$. Parameter k is in $^\circ\text{C}^{-1}$ for Selsted, GLMM and GLMM2 models, and in $\mu\text{mol CO}_2 \text{ m}^{-2}$
1324 $\text{s}^{-1} \text{ }^\circ\text{C}^{-1}$ for LMM and LMM2. The parameters a , b and c are dimensionless for Selsted, GLMM and GLMM2
1325 models and are in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ for the LMM and LMM2 models.

Model Type	Variables	Equations for R_S Model	Equations for R_H Model
Selsted	T	$R_0 e^{kT}$	$R_0 e^{kT}$
	TM	$R_0 e^{kT} (1 - e^{a-b(1-M)^{-2}})$	$R_0 e^{kT} (1 - e^{a-b(1-M)^{-2}})$
	TB	$R_0 e^{kT} (B + c)$	-
	TP	$R_0 e^{kT} (P + c)$	-
	TMB	$R_0 e^{kT} (1 - e^{a-b(1-M)^{-2}}) (B + c)$	-
	TMP	$R_0 e^{kT} (1 - e^{a-b(1-M)^{-2}}) (P + c)$	-
LMM	T	$R_0 + kT$	$R_0 + kT$
	TM	$R_0 + kT + aM$	$R_0 + kT + aM$
	TB	$R_0 + kT + cB$	-
	TP	$R_0 + kT + cP$	-
	TMB	$R_0 + kT + aM + cB$	-
	TMP	$R_0 + kT + aM + cP$	-
LMM2 **	TM	$R_0 + kT + a(M - 1)^2$	$R_0 + kT + a(M - 1)^2$
	TB	$R_0 + kT + c(B - 1)^2$	-
	TP	$R_0 + kT + c(P - 1)^2$	-
	TMB	$R_0 + kT + a(M - 1)^2 + c(B - 1)^2$	-
	TMP	$R_0 + kT + a(M - 1)^2 + c(P - 1)^2$	-
GLMM *	T	$R_0 e^{kT}$ (identical to Selsted - T)	$R_0 e^{kT}$
	TM	$R_0 e^{kT} e^{aM}$	$R_0 e^{kT} e^{aM}$
	TB	$R_0 e^{kT} e^{cB}$	-
	TP	$R_0 e^{kT} e^{cP}$	-
	TMB	$R_0 e^{kT} e^{aM} e^{cB}$	-
	TMP	$R_0 e^{kT} e^{aM} e^{cP}$	-
GLMM2 **	TM	$R_0 e^{kT} e^{a(M-1)^2}$	$R_0 e^{kT} e^{a(M-1)^2}$
	TB	$R_0 e^{kT} e^{c(B-1)^2}$	-
	TP	$R_0 e^{kT} e^{c(P-1)^2}$	-
	TMB	$R_0 e^{kT} e^{a(M-1)^2} e^{c(B-1)^2}$	-
	TMP	$R_0 e^{kT} e^{a(M-1)^2} e^{c(P-1)^2}$	-

1326 * The equation for the GLMM-T model is identical to the Selsted-T equation. The GLMM-T model is still included as
1327 a separate model due to a different treatment of model residuals and different optimality criteria in the calibration of
1328 the Selsted and the GLMM models, which results in different optimal parameters for the two models.

1329 ** The equations and the optimal parameters for the LMM-T and GLMM-T models are identical to those of
1330 respectively LMM2-T and GLMM2-T. Therefore, LMM2-T and GLMM2-T are not included in the table.

1331

1332 **Table 4.** The residual standard deviation with $RMSE_C$ and $RMSE_V$ (Validation Type I and II) values for a selection of GLMM
 1333 models. Only models in which all variables were significant are shown. See Table 2 for definition of validation types, Table 3
 1334 for model definitions and Table 5 for parameter values of the models shown here. Results for cross-validation and with
 1335 additional error metrics are provided in Appendix C.

Variables	Residual standard deviation				
	random factors	unexplained	$RMSE_C$	$RMSE_{V1}$	$RMSE_{V2}$
R_S models (Old community)					
T _{soil}	0.08	0.07	0.37	0.51	0.57
T _{soil+M}	0.06	0.08	0.32	0.62	0.58
T _{soil+P}	0.06	0.08	0.32	0.51	0.48
R_H models					
T _{soil}	0.14	0.25	0.32	0.39	-
T _{soil+M}	0.15	0.25	0.31	0.37	-

1336

1337

1338 **Appendix A Details of the P_G Measurements**

1339 The gross photosynthetic rate provided a measure of photosynthetic activity for the three heathland
1340 ages. The gross photosynthetic rate (P_G) was calculated as the Net Ecosystem Exchange (NEE) rate of
1341 CO₂ flux minus the Ecosystem Respiration (ER) rate of CO₂ flux (μmol CO₂ m⁻² s⁻¹). This
1342 photosynthetic rate has a negative sign. A loess smoother curve was applied to the photosynthesis data
1343 to obtain daily estimates of plant activity.

1344 The CO₂ fluxes of the vegetation were measured a LI-6400 infrared gas analyzer (LI-COR, Lincoln,
1345 NE, USA) attached to a 288 L ultra-violet light transparent Perspex chamber (60cm x 60cm x 80cm)
1346 using the method described by Larsen et al. (2007). The chamber was installed with a fan as well as a
1347 soil temperature probe (LI-6400-09 temperature probe) and a PAR sensor (LI-COR quantum sensor).

1348 Three permanent sampling locations were selected in each vegetation age. A metal base frame (60cm x
1349 60cm) was permanently installed using small, narrow sandbags to provide a seal between the frame and
1350 the soil surface and fixed with metal pins. Measurement of CO₂ fluxes commenced immediately prior to
1351 the Perspex chamber being placed on the frame so as to capture the point at which the chamber was
1352 sealed and NEE occurred entirely within the chamber. The LICOR measurement program ran for 180
1353 seconds however, the results obtained while the chamber was being fitted were later discarded so that
1354 only data obtained from the sealed chamber (approximately 150 seconds) were utilized for calculation
1355 of NEE rates. After the NEE measurements, the chamber was vented and measurements of the ER rate
1356 were obtained by covering the chamber with a fitted blackout-cloth, in which the outer layer was white
1357 and the inner lining was black, to minimize any heating effect within the darkened chamber.

1358 In most cases, NEE decreased from the first to the third minute of measurement, indicating an effect of
1359 the chamber by the decreasing CO₂ concentration as photosynthesis progressed. Therefore a linear
1360 regression did not provide a good fit for all measurements. To overcome this problem, the HMR
1361 procedure was used (Pedersen et al. 2010). This procedure was developed for soil-atmosphere trace-gas
1362 flux estimation with static chambers and tests the fit of both log-linear and linear regression models to
1363 the NEE or ER data at each measurement. If linear regression provided the best fit, the flux value was
1364 determined by the slope of the regression line. If non-linear regression gave the best fit, the flux was
1365 determined by the slope at t = 0 sec. The HMR procedure is implemented in an R-package (Pedersen
1366 2011) and this implementation was used in our study.

1367

1368 **References**

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1373 gas flux estimation with static chambers, *Eur. J. Soil Sci.*, 61, 888-902, 2010.
1374 .
1375
1376

1377 **Appendix B Details of the Soil Moisture Model**

1378 The soil moisture model used in this study is a zero-dimensional finite difference model using a daily
1379 time resolution of rainfall data and air temperature data as model inputs. It was constructed and
1380 calibrated on approximately one year of observed soil moisture, rainfall and temperature data for 12
1381 individual soil moisture sensors. The model comprises the following equations:

1382

$$1383 \quad Drain_t = \max(0; Smoist_{t-1} - depth \cdot fc) \cdot df \quad (B1)$$

$$1384 \quad AvSmoist_t = \max(0; Smoist_{t-1} - depth \cdot wp) \quad (B2)$$

$$1385 \quad ET_t = \min(Temp_t \cdot tf; AvSmoist_t) \cdot ef \quad (B3)$$

$$1386 \quad EfRain_t = Rain_t \cdot \left(\frac{Smoist_t}{(depth \cdot poros)} \right)^{rf} \quad (B4)$$

$$1387 \quad Smoist_t = \min(depth \cdot poros; Smoist_{t-1} + EfRain_t) - Drain_t - ET_t \quad (B5)$$

1388

1389 In the equations, t refers to a day. Equation (B1) calculates drainage ($Drain_t$, in mm day⁻¹) as a linear
1390 reservoir with soil moisture ($Smoist_{t-1}$, in mm) above a threshold ($depth \cdot fc$) as the driving force.
1391 $Drain_t$ refers to the drainage of soil moisture from the soil layer under consideration (i.e. the top of the
1392 mineral soil down to $depth$ mm); $Smoist_{t-1}$ refers to the soil moisture in the soil layer under
1393 consideration, and $depth$, fc (field capacity, as a fraction of the soil volume) and df (drainage
1394 fraction) are model parameters. The $depth$ parameter is set to 100 mm, while the values for fc and df
1395 were identified by model calibration.

1396

1397 Equation (B2) calculates the soil moisture available for evapotranspiration ($AvSmoist_t$, in mm) and the
1398 parameter wp (as a fraction of the soil volume) represents the wilting point below which only a
1399 negligible rate of evapotranspiration occurred. The value for wp was found by model calibration.

1400

1401 Evapotranspiration (ET_t) is calculated in Eq. (B3). Evapotranspiration is a modeled linear reservoir with
1402 either the air temperature or the available soil moisture as the driving force, depending of which factor
1403 is limiting. The parameter tf is set to 1 mm (degree C)⁻¹, and the value for the parameter ef was
1404 identified by model calibration.

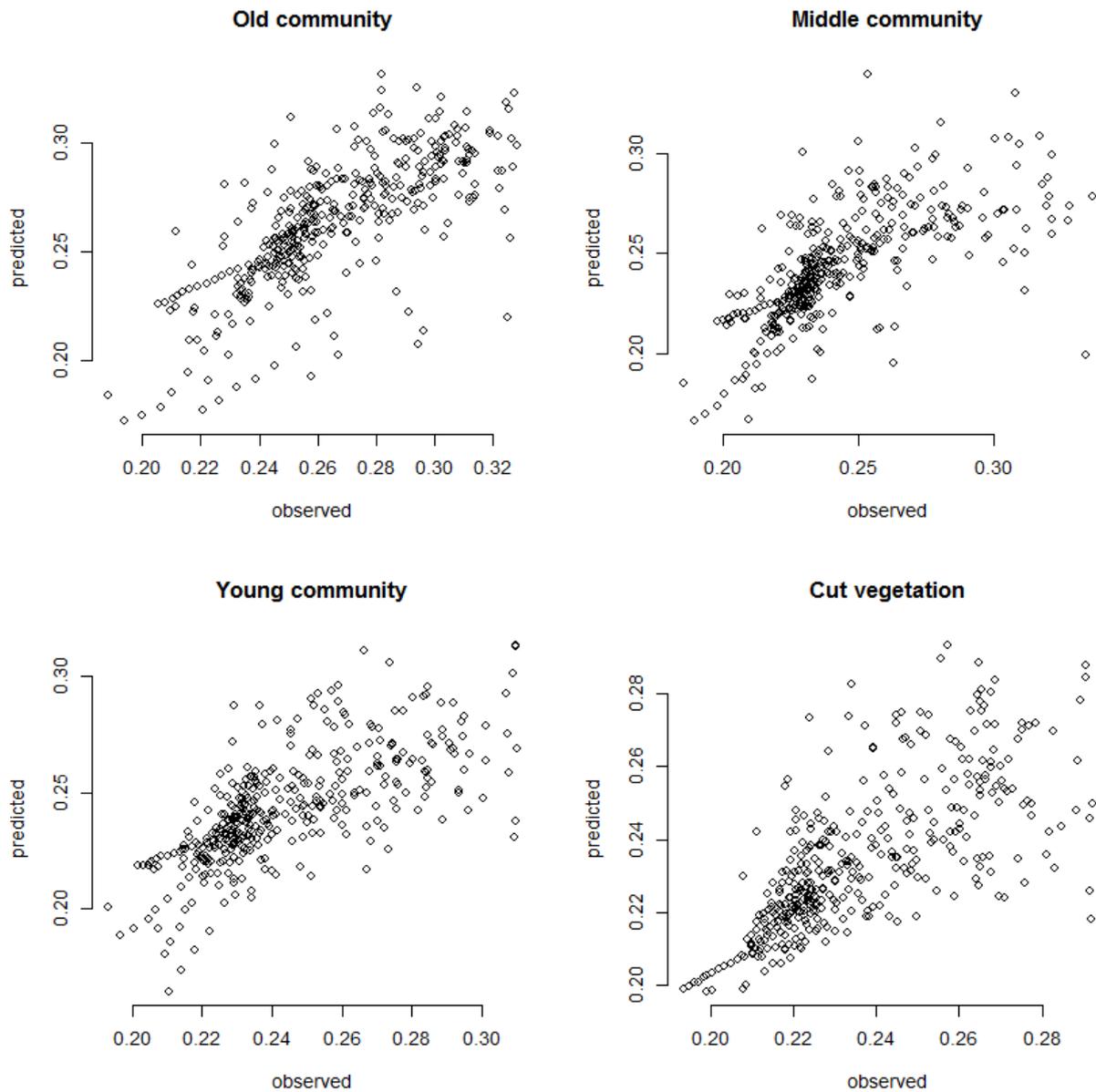
1405 The effective rain, i.e. the rainfall which enters the soil layer under consideration ($EfRain_t$, in mm), is
1406 calculated in Eq. (B4). $EfRain_t$ is proportional to a soil saturation factor which contains two
1407 parameters: soil porosity ($poros$) and a rainfall factor (rf). The porosity is calculated by taking the

1408 maximum observed soil moisture content over the measurement period, while the rainfall factor is
1409 calculated by model calibration.

1410
1411 In Eq. (B5), an update of the soil moisture is calculated by a balance equation, whereby it is assumed
1412 that any rainfall which cannot be stored in the soil layer under consideration is lost as surface runoff.

1413
1414 The water balance model thus contains eight parameters, three of which have fixed values ($depth =$
1415 $100mm$, $poros = \max_{all t}(Smoist_t/depth)$, and $tf = 1 \text{ mm}(\text{degree C})^{-1}$), and five of which were
1416 found via calibration (df , ef , fc , rf and wp). Calibration was undertaken by minimizing the root mean
1417 squared error between observed and predicted soil moisture, using the optimization routine by Byrd et
1418 al. (1995), as implemented in the standard R function ‘optim’.

1419
1420 The fit of the soil moisture model for the different treatments is shown in the diagnostic plots of Figure
1421 B1. The plots illustrate that there is still quite some room for improvement in the soil moisture model.
1422 For each of the cases, the explained variance in the observed versus predicted plot is approximately 0.7.



1423
 1424 **Figure B.1** Observed versus predicted soil moisture for the four different vegetation communities in
 1425 this study.

1426
 1427 **References**

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 1429 Optimization. *Siam Journal on Scientific Computing*, **16**, 1190-1208.

1430
 1431

1432 **Appendix C Dealing with Additional Validation Methods and Error Metrics**

1433

1434 A large number of different methods exist for model calibration, model validation and for the
1435 assessment of model fit. Calibration Type I, Validation Type I and Validation Type II are described and
1436 applied in the main paper. Additional methods were selected for further consideration in the model
1437 selection procedure and these are summarized in Table C1.

1438

1439 Calibration Type II uses all the available data (calibration and validation data) from the same period to
1440 calibrate the model and the error is shown as $RMSE_{C2}$. In this case, there is no validation using Type I
1441 or Type II model validation. The Validation Type III is also commonly known as cross-validation,
1442 where the dataset is partitioned and one subset is used to calibrate the model and then the remaining
1443 subset calibrates the model. Multiple rounds of cross-validation were performed using different
1444 partitions and the validation results averaged ($RMSE_{V3}$).

1445

1446 Additional error metrics that were generated and available for model comparison included the mean
1447 error, absolute mean error, mean squared error, root mean square error (RMSE), percentage bias, Nash-
1448 Sutcliffe Efficiency (NSE), Refined Index of Agreement (RIA), Akaike Information Criteria (AIC),
1449 Persistence Index and Volumetric Efficiency.

1450

1451 To compare the different calibration / validation methods, the RMSE, NSE and RIA for selected R_S and
1452 R_H models are provided in Table C2 and Table C3. The NSE is a normalized statistic that determines
1453 the relative magnitude of the residual variance compared to the measured data variance (Nash 1970).
1454 This can range from $-\infty$ to 1.0, where a value of 1.0 corresponds to a perfect match of modeled data to
1455 the observed data and a value < 0 occurs when the observed mean is a better predictor than the model.
1456 The RIA is another statistical index of model performance, which is dimensionless and ranges from
1457 -1.0 to 1.0 (Willmott 2012). The Calibration Type I $RMSE_{C1}$ values are graphed in the main article,
1458 while the associated NSE_{C1} values are plotted in Figure C1.

1459

1460 The results also showed that where Calibration Type II was undertaken, the RMSE values for both R_S
1461 and R_H models were marginally higher than the Calibration Type I output, although the general ranking
1462 of the models did not change, with the exception of the a number of models including a P variable in
1463 which the parameters were no longer significant.

1464
1465 The error metrics for both the R_S and R_H models indicated that NSE_{C1} and RIA_{C1} were closest to 1.0 for
1466 the models with the lowest $RMSE_{C1}$ values and showed the same general ranking as for the RMSE
1467 values. The exception to this trend was again in a number of models with the P variable, where the
1468 NSE_{C2} was lower than the NSE_{C1} and $RMSE_{C2}$ was higher than the $RMSE_{C1}$, thus confirming that these
1469 models should be excluded from further consideration.

1470
1471 In the R_S models, the NSE_{C1} and RIA_{C1} values were generally > 0.77 where the $RMSE_{C1}$ values were
1472 < 0.5 , (Table C2), with the NSE_{C1} values being closest to 1.0 in the Selsted and GLMM models (Figure
1473 C1). The R_S models in which parameters were significant for all community ages were the GLMM T_{soil} ,
1474 GLMM T_{air} , GLMM T_{air+P} , Selsted T_{soil} and Selsted T_{air} . Given that the Calibration Type II results
1475 showed the inclusion of the P variable increased the $RMSE_{C2}$ and reduced the NSE_{C2} values, this model
1476 was excluded from further consideration.

1477
1478 The final selection of either a Selsted model or GLMM model is discussed within the main article and
1479 the additional calibration, validation methods and error metrics provided to illustrate that the correlation
1480 between the metrics as well as the different calibration-validation methods is very high..

1481

1482 **References**

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1487

1488 **Table C1** Description of the data used for model calibration and validation.

Modelling Stage	Total Soil Respiration Models (R_S)	Heterotrophic Respiration Models (R_H)
Calibration (Type I)	<i>Data:</i> Untrenched plots <i>Dates:</i> September 2011 – August 2012	<i>Data:</i> Trenched plots <i>Dates:</i> September 2011 – August 2012
Calibration (Type II)	<i>Data:</i> Untrenched plots and Untrenched Validation plots <i>Dates:</i> September 2011 – August 2012	<i>Data:</i> Trenched plots and Trenched Validation plots <i>Dates:</i> September 2011 – August 2012
Validation (Type I)	<i>Data:</i> Untrenched Validation plots <i>Dates:</i> September 2011 – August 2012	<i>Data:</i> Trenched Validation plots <i>Dates:</i> September 2011 – August 2012
Validation (Type II)	<i>Data:</i> Untrenched Validation plots <i>Dates:</i> November 2010 – August 2011	—
Validation (Type III)	<i>Data:</i> Partial data from Untrenched plots <i>Dates:</i> November 2010 – August 2011	<i>Data:</i> Partial data from Trenched plots <i>Dates:</i> September 2011 – August 2012

1489

1490

1491

1492 **Table C2.** The measures of model fit for a selection of R_S models using the Calibration data, Validation Type I
 1493 data, Validation Type II data, Validation Type III data. See Table C1 for Type definitions and Table 3 in the main
 1494 article for parameter explanations. Calibration Type I models which included non-significant parameters are not
 1495 shown in the table. Grey italicized text indicates that not all variables in this Calibration Type II model were
 1496 significant.

Model	Variables	Age	Calibration Type I			Calibration Type II			Validation Type I			Validation Type II		
			RMSE _{C1}	NSE _{C1}	RIA _{C1}	RMSE _{C2}	NSE _{C2}	RIA _{C2}	RMSE _{V1}	NSE _{V1}	RIA _{V1}	RMSE _{V2}	NSE _{V2}	RIA _{V2}
GLMM	T _{soil} +M	Old	0.32	0.82	0.81	0.47	0.64	0.74	0.62	0.38	0.64	0.58	0.35	0.74
GLMM	T _{soil} +P	Old	0.32	0.82	0.81	0.46	0.64	0.74	0.51	0.57	0.7	0.48	0.55	0.74
GLMM2	T _{soil} +M+P	Middle	0.34	0.85	0.83	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{soil} +P	Middle	0.34	0.85	0.83	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{air} +P	Old	0.35	0.78	0.79	0.67	0.25	0.62	0.57	0.47	0.66	0.52	0.49	0.74
GLMM	T _{soil} +M	Middle	0.36	0.83	0.83	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{soil} +P	Middle	0.36	0.84	0.82	As per Calibration Type I			-	-	-	-	-	-
GLMM2	T _{air} +M+P	Old	0.36	0.77	0.79	<i>0.73</i>	<i>0.12</i>	<i>0.61</i>	0.69	0.23	0.59	0.45	0.62	0.74
GLMM	T _{soil}	Middle	0.37	0.82	0.82	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{soil}	Middle	0.37	0.82	0.82	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{soil}	Old	0.37	0.76	0.79	0.46	0.65	0.74	0.51	0.58	0.71	0.57	0.38	0.74
LMM	T _{soil} +M	Old	0.37	0.76	0.76	0.47	0.64	0.71	0.59	0.43	0.65	0.48	0.55	0.74
Selsted	T _{soil}	Old	0.37	0.76	0.78	0.46	0.65	0.73	0.51	0.58	0.71	0.56	0.4	0.74
GLMM	T _{air} +P	Middle	0.39	0.81	0.8	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{air} +P	Middle	0.39	0.81	0.8	As per Calibration Type I			-	-	-	-	-	-
GLMM2	T _{air} +M+P	Middle	0.4	0.79	0.8	As per Calibration Type I			-	-	-	-	-	-
LMM	T _{soil}	Middle	0.41	0.78	0.8	As per Calibration Type I			-	-	-	-	-	-
GLMM	T B	Young	0.42	0.83	0.8	As per Calibration Type I			-	-	-	-	-	-
LMM2	T P	Middle	0.43	0.76	0.78	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{soil}	Young	0.49	0.77	0.78	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{soil}	Young	0.49	0.77	0.78	As per Calibration Type I			-	-	-	-	-	-
LMM2	T _{air} +M	Old	0.5	0.55	0.64	0.57	0.46	0.61	0.64	0.33	0.58	0.59	0.34	0.74
GLMM	T _{air} +M	Old	0.52	0.52	0.66	0.59	0.41	0.61	0.69	0.24	0.58	0.55	0.41	0.74
Selsted	T _{air} +P	Young	0.54	0.72	0.74	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{air}	Old	0.55	0.47	0.6	0.59	0.42	0.57	0.61	0.39	0.54	0.53	0.46	0.74
GLMM	T _{air} +P	Young	0.55	0.71	0.74	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{air}	Old	0.56	0.45	0.64	0.6	0.41	0.6	0.61	0.39	0.58	0.55	0.43	0.74
GLMM2	T _{air} +P	Young	0.56	0.7	0.75	As per Calibration Type I			-	-	-	-	-	-
LMM2	T _{air} +P	Young	0.56	0.7	0.74	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{air} +M	Middle	0.58	0.56	0.68	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{air}	Middle	0.59	0.55	0.63	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{air}	Middle	0.61	0.52	0.66	As per Calibration Type I			-	-	-	-	-	-
Selsted	T _{air}	Young	0.66	0.58	0.65	As per Calibration Type I			-	-	-	-	-	-
GLMM	T _{air}	Young	0.68	0.56	0.68	As per Calibration Type I			-	-	-	-	-	-
LMM	T _{soil} +M+P	Old	1.21	-1.58	0.35	1.07	-0.92	0.39	1.38	-2.1	0.31	1.63	-4.11	0.31
GLMM	T _{air} +M+P	Old	1.91	-5.41	0.32	<i>1.15</i>	<i>-1.22</i>	<i>0.46</i>	2.19	-6.78	0.27	2.73	-13.29	0.27
GLMM	T _{soil} +M+P	Old	2.32	-8.49	0.27	<i>1.68</i>	<i>-3.67</i>	<i>0.35</i>	2.86	-12.24	0.21	3.54	-22.98	0.21

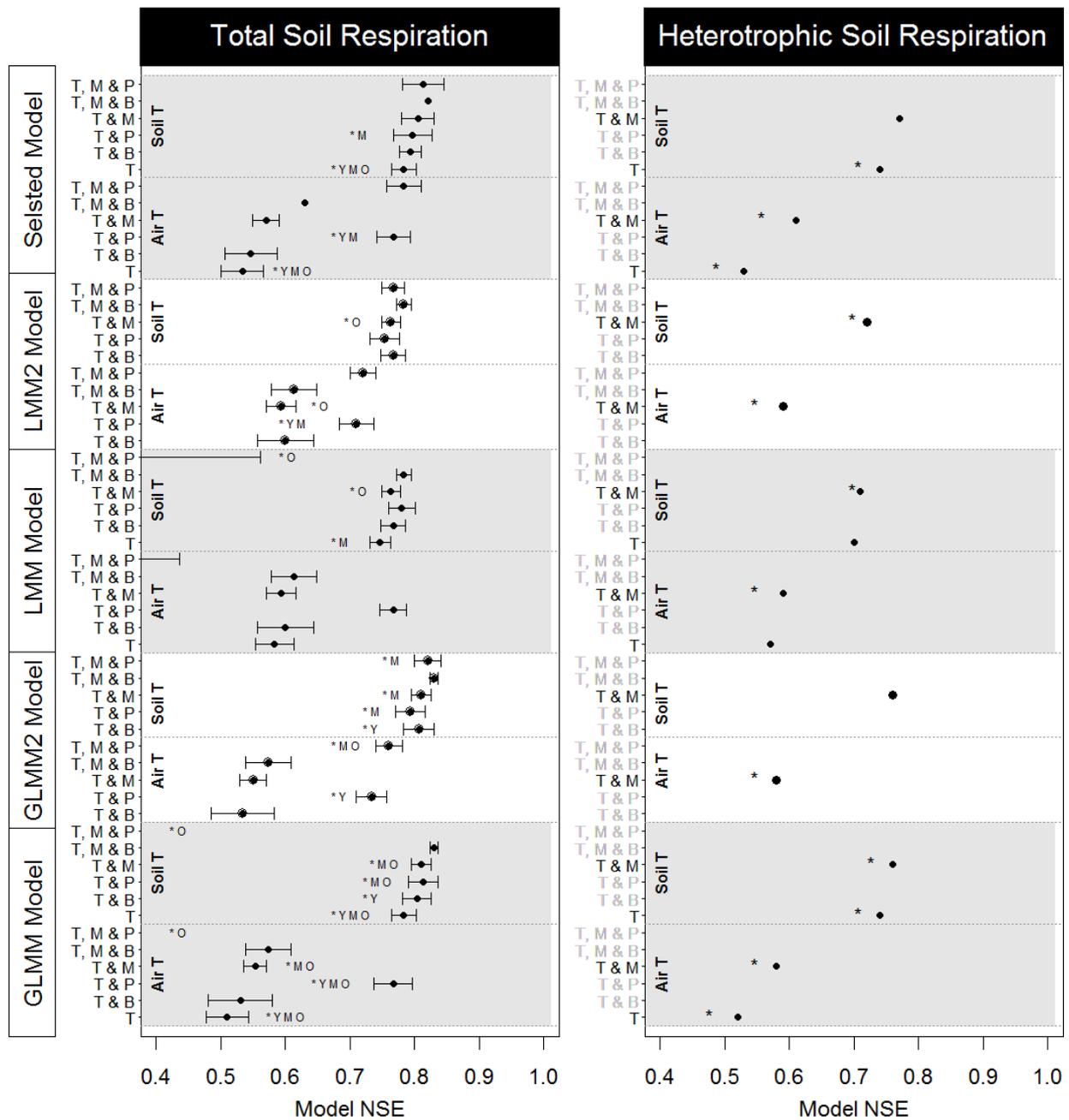
1497
 1498 **Table C3** The measures of model fit for a selection of R_H models using the Calibration data, Validation Type I
 1499 data, and Validation Type III data. See Table C1 for Type definitions and Table 3 in main article for parameter
 1500 explanations.

Model	Variables	Age	Calibration Type I			Calibration Type II			Validation Type I			Validation Type II		
			RMSE _{C1}	NSE _{C1}	RIA _{C1}	RMSE _{C2}	NSE _{C2}	RIA _{C2}	RMSE _{V1}	NSE _{V1}	RIA _{V1}	RMSE _{V2}	NSE _{V2}	RIA _{V2}
GLMM	T _{soil} +M	Trenched	0.31	0.76	0.77	0.34	0.71	0.74	0.37	0.64	0.72	-	-	-
GLMM	T _{soil}	Trenched	0.32	0.74	0.76	0.35	0.68	0.73	0.39	0.61	0.71	-	-	-
Selsted	T _{soil}	Trenched	0.32	0.74	0.76	0.35	0.68	0.73	0.39	0.61	0.71	-	-	-
LMM2	T _{soil} +M	Trenched	0.34	0.72	0.74	0.36	0.67	0.71	0.38	0.61	0.69	-	-	-
LMM	T _{soil} +M	Trenched	0.34	0.71	0.74	0.36	0.67	0.71	0.39	0.61	0.69	-	-	-
Selsted	T _{air} +M	Trenched	0.39	0.61	0.67	0.42	0.55	0.64	0.45	0.48	0.61	-	-	-
LMM2	T _{air} +M	Trenched	0.4	0.59	0.67	0.42	0.55	0.65	0.44	0.5	0.62	-	-	-
LMM	T _{air} +M	Trenched	0.4	0.59	0.67	0.42	0.55	0.65	0.44	0.5	0.62	-	-	-
GLMM2	T _{air} +M	Trenched	0.41	0.58	0.68	0.43	0.53	0.65	0.46	0.45	0.62	-	-	-
GLMM	T _{air} +M	Trenched	0.41	0.58	0.68	0.43	0.53	0.65	0.46	0.45	0.62	-	-	-
GLMM	T _{air}	Trenched	0.44	0.52	0.65	0.46	0.47	0.62	0.49	0.39	0.59	-	-	-
Selsted	T _{air}	Trenched	0.43	0.53	0.63	0.45	0.48	0.6	0.47	0.42	0.56	-	-	-

1501

1502

1503



1504
 1505 **Figure C1** Comparison of NSE_C values for models of (a) total soil respiration data (Untrenched plots)
 1506 and (b) heterotrophic soil respiration data (Trenched plots). The models tested are listed on the left side
 1507 of the figure. The explanatory variables within each model are listed on the y axis and are abbreviated
 1508 as: T = temperature (soil or air °C as indicated), M = soil moisture, B = relative biomass, P = relative
 1509 photosynthesis. The “*” indicates that all model parameters were significant for one of either “Y”
 1510 (Young), “M” (Middle) or “O” (Old) vegetation community models. The SEM bars on the total soil
 1511 respiration means were calculated from the NSE_C ’s of the three community ages. SEM bars could not

1512 be calculated for the heterotrophic models. Four mean values are outside the plotted range: GLMM

1513 $T_{\text{air}+M+P}$ (-1.966), GLMM $T_{\text{soil}+M+P}$ (-2.803), LMM $T_{\text{air}+M+P}$ (-0.136) and LMM $T_{\text{soil}+M+P}$ (-0.153).

1514

1515

1516 **Appendix D - Diagnostic Plots for selected Models**

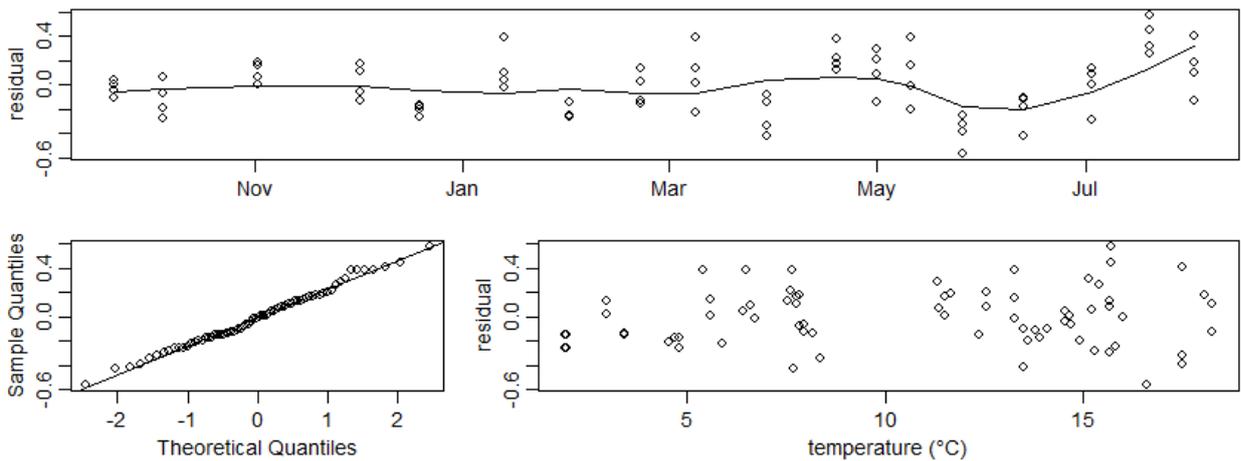
1517

1518 In this appendix, diagnostic plots are shown for the R_S and R_H models listed in Tables 4 and 5, using
1519 Calibration data (see Table 2). Per model, the residuals are plotted over time as well as over
1520 Temperature and furthermore the quantile of the residuals is plotted against that of the theoretical
1521 distribution. While the error-properties of the residuals of nearly all of these models are good, a clear
1522 temporal autocorrelation of the residuals is visible for the R_S models (with a slight over-prediction in
1523 February and an under-prediction of the observed respiration in May). This structure is important, since
1524 it points at some underlying variable or process which was not observed in this study.

1525

1526 **Models for total respiration, using soil temperature (T_{soil})**

1527 Old community



1528

1529

1530

1531

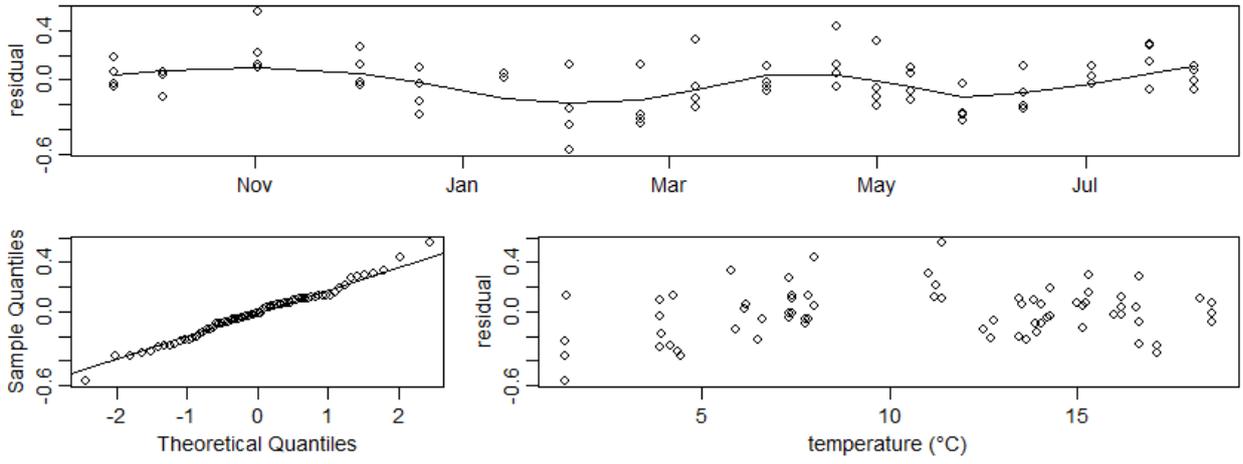
1532

1533

1534

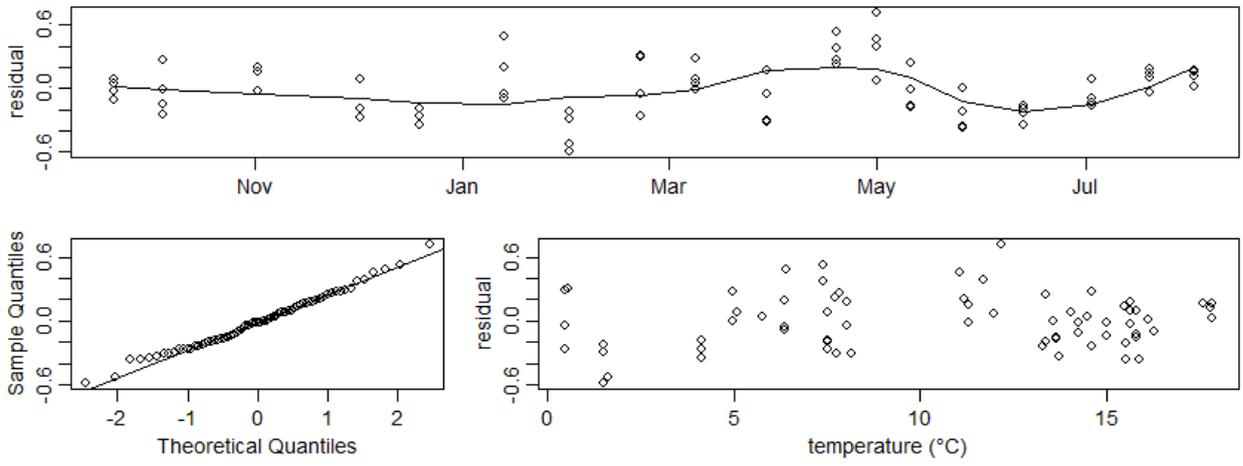
1535

1536 Middle community



1537

1538 Young community

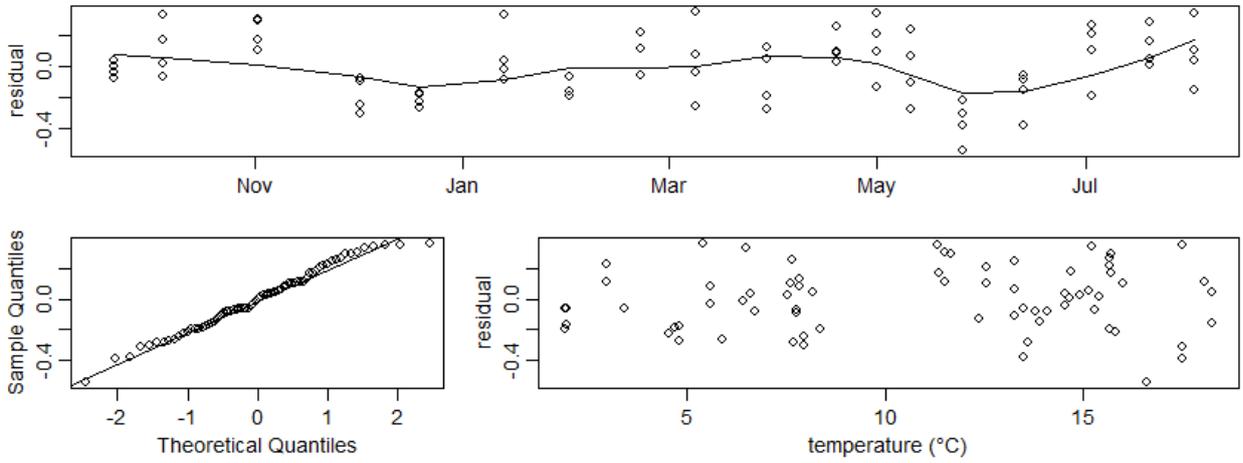


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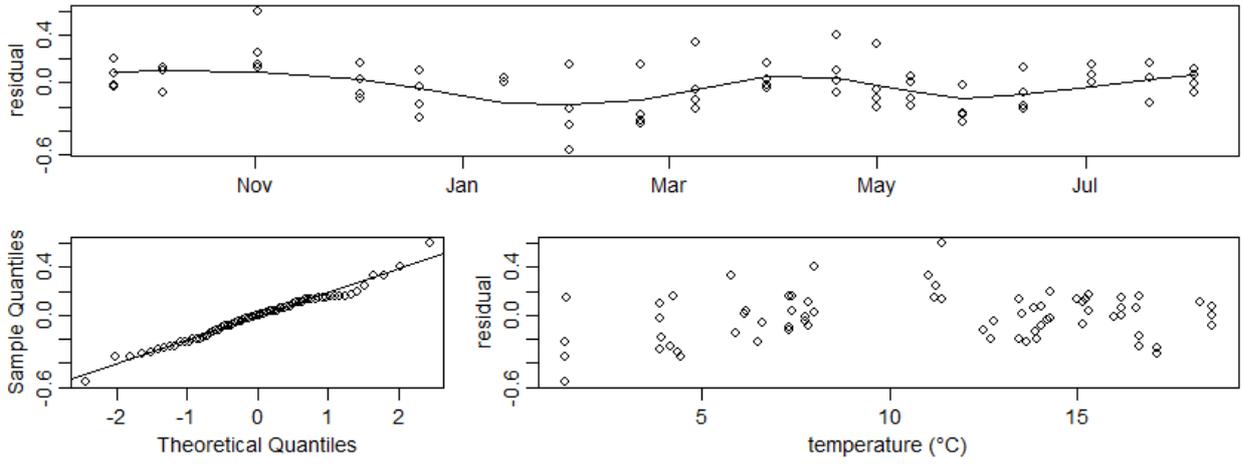
1540

1541 **Models for total respiration, using soil temperature and relative moisture ($T_{soil}+M$)**

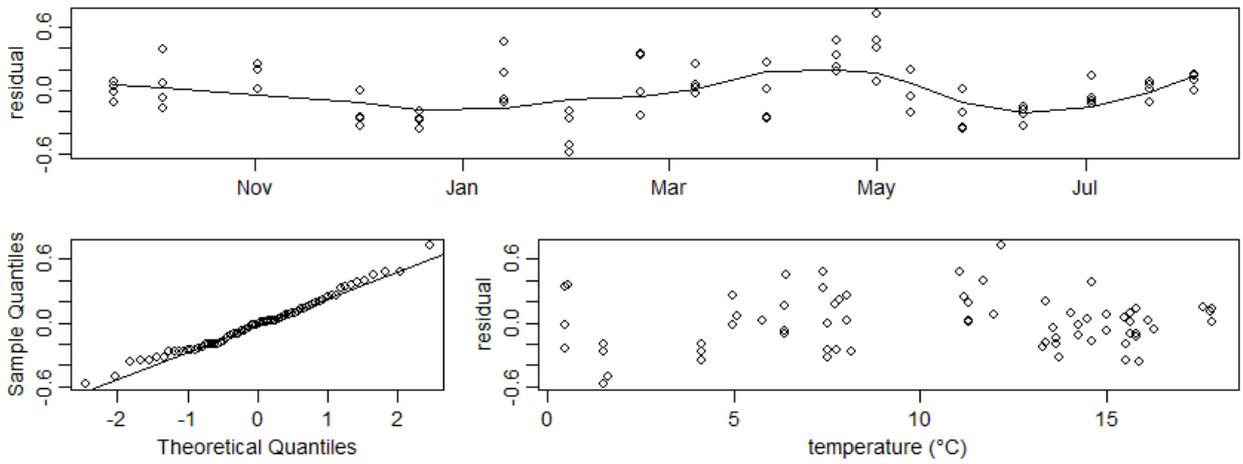
1542 Old community



1543
1544 Middle community



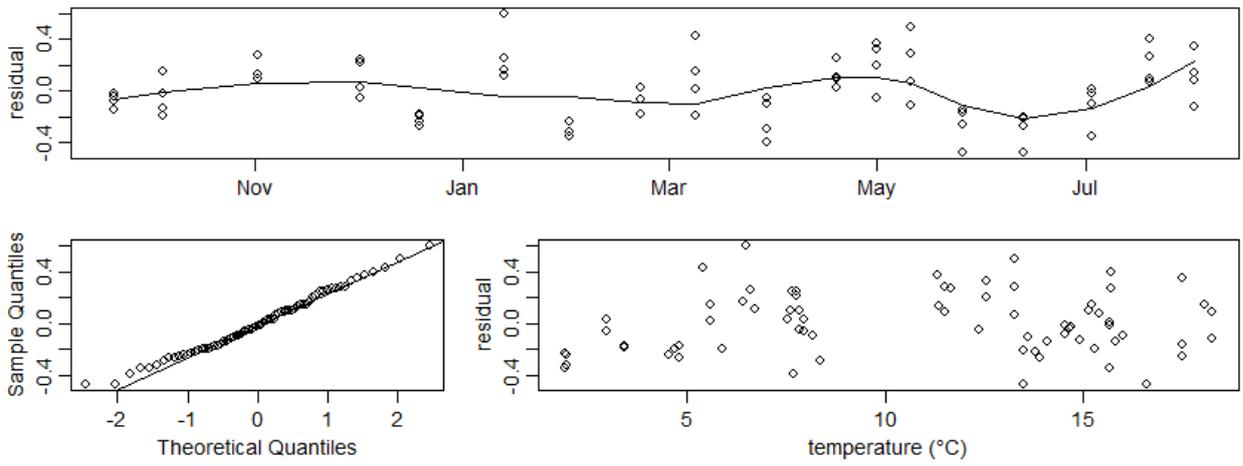
1545
1546 Young community



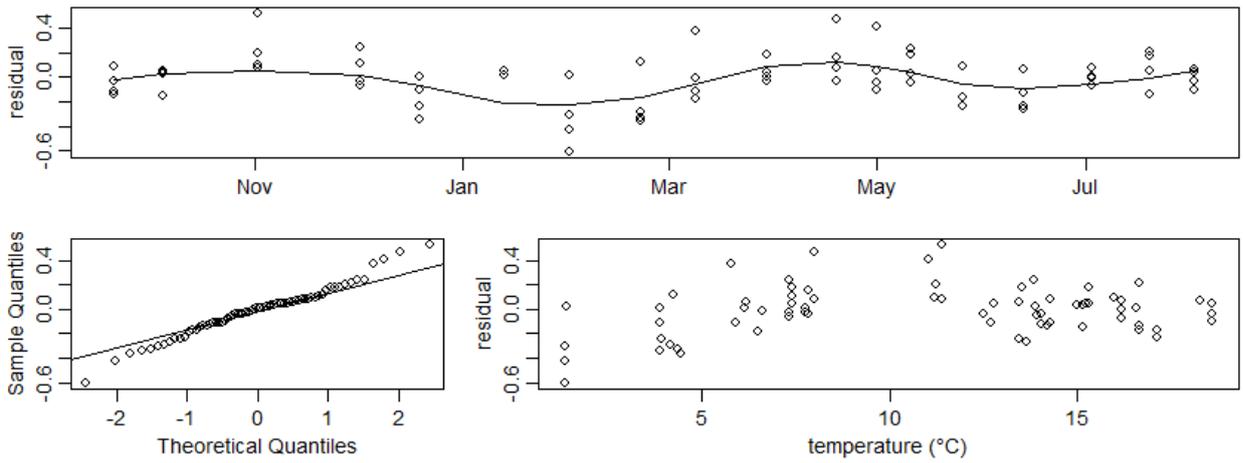
1547
1548

1549 **Models for total respiration, using soil temp. and relative photosynthetic activity (T_{soil+P})**

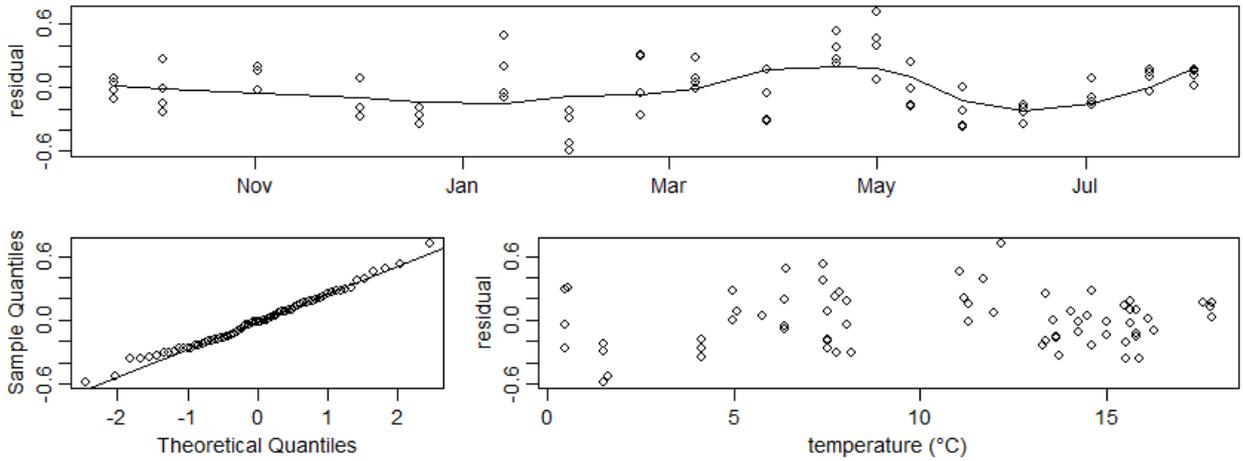
1550 Old community



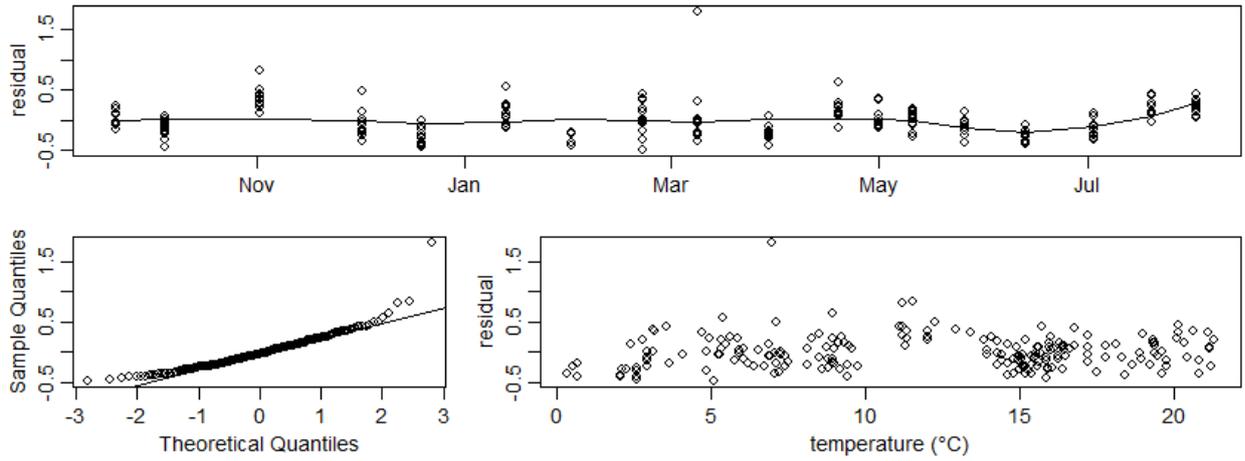
1551
1552 **Middle community**



1553
1554 **Young community**



1555
1556 **Models for heterotrophic respiration, using soil temperature (T_{soil})**

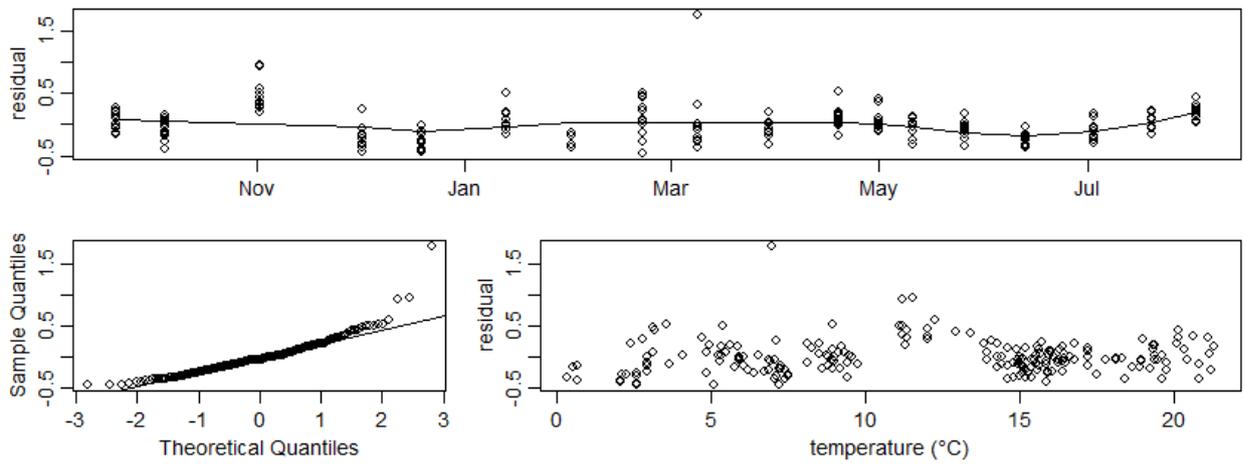


1557

1558

1559

1560 **Models for heterotrophic respiration, using soil temperature ($T_{\text{soil}+M}$)**



1561

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