

Supplementary Information

Fungal community composition predicts forest carbon storage at a continental scale

Mark A. Anthony^{1,2,3*}, Leho Tedersoo⁴, Bruno De Vos⁵, Luc Croisé⁶, Henning Meesenburg⁷, Markus Wagner⁷, Henning Andreea⁸, Frank Jacob⁸, Paweł Lech⁹, Anna Kowalska⁹, Martin Greve¹⁰, Genoveva Popova¹¹, Beat Frey², Arthur Gessler^{1,2}, Marcus Schaub², Marco Ferretti², Peter Waldner², Vicent Calatayud¹², Roberto Canullo¹³, Giancarlo Papitto¹⁴, Aleksander Marinšek¹⁵, Morten Ingerslev¹⁶, Lars Vesterdal¹⁶, Pasi Rautio¹⁷, Helge Meissner¹⁸, Volkmar Timmermann¹⁹, Mike Dettwiler¹, Nadine Eickenscheidt²⁰, Andreas Schmitz^{20, 21}, Nina van Tiel^{1,22}, Thomas W. Crowther¹, Colin Averill¹

¹Department of Environmental Systems Science, ETH Zürich, Zürich, Switzerland

²Swiss Federal Institute for Forests, Snow, and the Landscape Research (WSL), Birmensdorf, Switzerland

³Center for Microbiology and Environmental Systems Science, University of Vienna, Vienna, Austria.

⁴Mycology and Microbiology Center, University of Tartu, Tartu, Estonia

⁵Environment & Climate Unit, Research Institute for Nature and Forest, Geraardsbergen, Belgium

⁶French National Forest Office, Fontainebleau, France

⁷Northwest German Forest Research Institute, Göttingen, Germany

⁸Sachsenforst State Forest, Pirna OT Graupa, Germany

⁹Forest Research Institute, Sękocin Stary, Poland

¹⁰Research Institute for Forest Ecology and Forestry, Trippstadt, Germany

¹¹Executive Environmental Agency at the Ministry of Environment and Water, Sofia, Bulgaria

¹²Mediterranean Center for Environmental Studies, Paterna, Spain

¹³Department of Plant Diversity and Ecosystem Management, University of Camerino, Camerino, Italy

¹⁴Arma dei Carabinieri Forestry Environmental and Agri-food protection Units, Rome, Italy

¹⁵Slovenian Forestry Institute, Ljubljana, Slovenia

¹⁶Department of Geosciences and Natural Resource Management, University of Copenhagen, Frederiksberg C, Denmark

¹⁷Natural Resources Institute Finland, Rovaniemi, Finland

¹⁸Division of Forest and Forest Resources, Norwegian Institute of Bioeconomy Research, Ås, Norway

¹⁹Division of Biotechnology and Plant Health, Norwegian Institute of Bioeconomy Research, Ås, Norway

²⁰State Agency for Nature, Environment and Consumer Protection of North Rhine-Westphalia, Recklinghausen, Germany

²¹Thuenen Institut of Forest Ecosystems, 16225 Eberswalde, Germany

²²Environmetnal Computational Science and Earth Observation Laboratory, EPFL, Lausanne, Switzerland

*Correspondence should be addressed to Mark A. Anthony (manthony5955@gmail.com)

55 **Supplementary Table 1.** Statistical summaries comparing models of tree growth with fungal
 56 versus bacterial species richness (# of operational taxonomic units; OTUs) as predictor
 57 variables. Data shows the results of generalized additive modeling, including model
 58 estimates (i.e., coefficients), standard errors (SE), and *P*-values from full statistical models
 59 including all co-variables. Note that each model contains only one microbiome predictor.
 60

Parametric terms	Estimate	SE	<i>t</i> -value	<i>P</i> -value
(Intercept)	-2.6573108	2.9009066	-0.916	0.36387
Fungal richness	0.5111837	0.2304736	2.218	0.03094
N deposition	0.1259197	0.364195	0.346	0.73092
Mineral Nitrogen Stock	0.4918893	0.1429244	3.442	0.00115
MAT	0.000856	0.0061821	0.138	8.90E-01
MAP	-0.000385	0.0004432	-0.869	3.89E-01
Soil pH	-0.1918281	0.1133522	-1.692	0.09656
Soil clay content	-0.0072983	0.0194758	-0.375	0.70938
Functional tree type	-0.7137729	0.242046	-2.949	0.00477
Stand age	0.0073393	0.0042508	1.727	0.09018
Smooth term	Est. DF	Ref. DF	<i>F</i> -value	<i>P</i> -value
Stem density	1.918	1.993	18.91	1.59E-06
Parametric terms	Estimate	SE	<i>t</i> -value	<i>P</i> -value
(Intercept)	-1.33E+01	4.14E+00	-3.198	0.00256
Bacterial richness	1.04E+00	5.24E-01	1.983	0.05363
N deposition	7.89E-01	3.12E-01	2.531	0.01503
Mineral Nitrogen Stock	3.79E-01	1.30E-01	2.907	0.0057
MAT	-1.15E-02	6.62E-03	-1.744	0.08807
MAP	-4.28E-04	5.36E-04	-0.798	0.42891
Soil pH	7.73E-02	1.13E-01	0.683	0.49821
Soil clay content	1.02E-02	2.02E-02	0.505	0.6158
Functional tree type	-3.77E-01	2.21E-01	-1.706	0.09514
Stand age	8.78E-03	4.03E-03	2.177	0.03491
Smooth term	Est. DF	Ref. DF	<i>F</i> -value	<i>P</i> -value
Stem density	1.966	1.999	15.93	7.68E-06

61
 62
 63
 64
 65
 66
 67
 68
 69

70 **Supplementary Table 2.** Statistical summaries of models of tree growth with fungal
 71 composition (PCoA1 and PCoA2) in the organic horizon. Predictors included PCoA axes for
 72 total fungi. Data shows the results of generalized additive modeling, including model
 73 estimates (i.e., coefficients), standard errors (SE), and *P*-values from full statistical models
 74 including all co-variables. Note that each model contains only one microbiome predictor.
 75

	Estimate	SE	<i>t</i> -value	<i>P</i> -value
(Intercept)	1.2344238	2.722149	0.453	0.6521
Fungal PCoA1	-1.6202941	0.962764	-1.683	0.0985
N deposition	-0.0033244	0.4283468	-0.008	0.9938
Organic N stock	-0.1031805	0.0898635	-1.148	0.2562
MAT	0.0064607	0.0083623	0.773	0.4433
MAP	0.000696	0.0004715	1.476	1.46E-01
Soil pH	-0.320192	0.128201	-2.498	1.58E-02
Soil clay content	-0.0176682	0.0207586	-0.851	0.3987
Functional tree type	-0.8488874	0.2546885	-3.333	0.0016
Stand age	0.011126	0.0051554	2.158	0.0356
Smooth term	Est. DF	Ref. DF	F-value	P-value
Stem density	1.811	1.964	15.33	1.42E-05
	Estimate	SE	<i>t</i> -value	<i>P</i> -value
(Intercept)	-0.985611	2.4867703	-0.396	0.6935
Fungal PCoA2	-2.8524385	1.0723338	-2.66	0.01041
N deposition	0.4128144	0.3980336	1.037	0.30455
Organic N stock	-0.0119976	0.0921148	-0.13	0.89688
MAT	0.0023515	0.0078527	0.299	0.76581
MAP	0.0004503	0.000466	0.966	3.39E-01
Soil pH	-0.3668078	0.1238869	-2.961	4.64E-03
Soil clay content	-0.0244192	0.0203232	-1.202	0.23508
Functional tree type	-0.8019711	0.2451975	-3.271	0.00192
Stand age	0.0122024	0.0048105	2.537	0.01429
Smooth term	Est. DF	Ref. DF	F-value	P-value
Stem density	1.88	1.985	18.09	1.94E-06

76
 77
 78
 79
 80
 81
 82
 83
 84
 85
 86
 87

88 **Supplementary Table 3. The relative abundance of fungal functional group**
89 **annotations.** Italicized groups are those within the group above (e.g., wood saprotroph are
90 a group within the saprotrophs annotated as “wood saprotrophs” in FUNGuild). Values
91 represent the mean relative abundance across all samples.
92

Soil horizon	Group	Relative abundance (%)
Organic	Ectomycorrhizal	23.8
	<i>'Pure' ectomycorrhizal</i>	19.3
	Saprotroph	38.5
	<i>'Pure' saprotroph</i>	15.8
	<i>Woot saprotroph</i>	14.2
	Endophyte	13.5
	Plant pathogen	7.3
	Ericoid mycorrhizal	2.6
Mineral	Ectomycorrhizal	26.4
	<i>'Pure' ectomycorrhizal</i>	22.7
	Saprotroph	33.6
	<i>'Pure' saprotroph</i>	15.5
	<i>Wood saprotroph</i>	8.9
	Endophyte	11.2
	Plant pathogen	3.8
	Ericoid mycorrhizal	3.5

93
94
95
96
97
98
99
100
101
102
103
104
105
106
107
108
109
110
111
112
113
114
115
116
117
118
119
120

121 **Supplementary Table 4.** Statistical summaries comparing models of tree growth with
122 different fungal functional group richness predictor variables. Data shows the model
123 estimates (coefficients), standard errors (SE), and *P*-values from full statistical models
124 including all co-variables. We only show the results for microbiome model output for brevity
125 because other co-variable effects are shown in Table S1. Note that each model contained
126 only one microbiome predictor. Models with ‘pure’ ectomycorrhizal fungal (Ectomyco.) and
127 saprotroph richness are those which were exclusively assigned to one versus multiple
128 functional guilds (e.g., saprotroph-pathogen). Note that pure category models were not made
129 for endophytes nor pathogens because it is expected that these groups will have more than
130 one trophic mode.
131

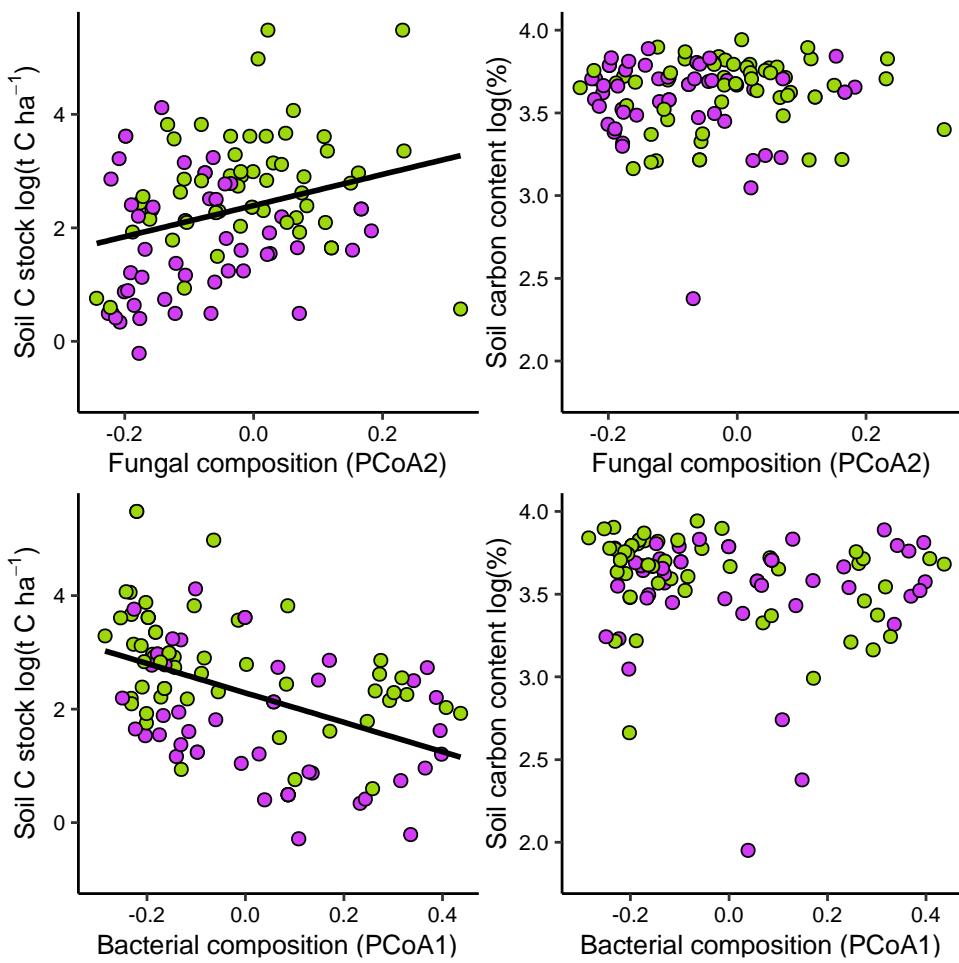
	Estimate	SE	<i>P</i> -value
Ectomyco. richness	0.04590836	0.1492676	0.7596
Endophyte richness	0.67559496	0.2333078	0.0055
Plant pathogen richness	0.41004186	0.195254	0.04059
Wood saprotroph richness	0.37284712	0.2035421	0.0727
Ericoid richness	4.68E-01	0.1973914	0.02168
Saprotroph richness	0.50018564	0.2269	3.19E-02
“Pure” ectomyco. richness	0.02906632	0.1470056	0.8440
“Pure” saprotroph richness	0.55919943	0.2279858	0.0176

132
133
134
135
136
137
138
139
140
141
142
143
144
145
146
147
148
149
150
151
152
153
154
155
156
157
158
159
160
161
162

163 **Supplementary Table 5.** Eigenvalues for principal coordinate analysis axes 1 and 2 for all
164 microbial groups considered in the study.
165

Group	Axis	Eigenvalue
Bacteria	PCoA1	25
Bacteria	PCoA2	11
Fungi	PCoA1	11
Fungi	PCoA2	9
Ectomycorrhizal	PCoA1	9
Ectomycorrhizal	PCoA2	8
Pure' ectomycorrhizal	PCoA1	10
Pure' ectomycorrhizal	PCoA2	9
Saprotoph	PCoA1	13
Saprotoph	PCoA2	11
Pure' saprotroph	PCoA1	21
Pure' saprotroph	PCoA2	9
Endophytes	PCoA1	17
Endophytes	PCoA2	14
Wood saprotroph	PCoA1	20
Wood saprotroph	PCoA2	11
Ericoid mycorrhizal	PCoA1	4
Ericoid mycorrhizal	PCoA2	3
Plant pathogen	PCoA1	13
Plant pathogen	PCoA2	11

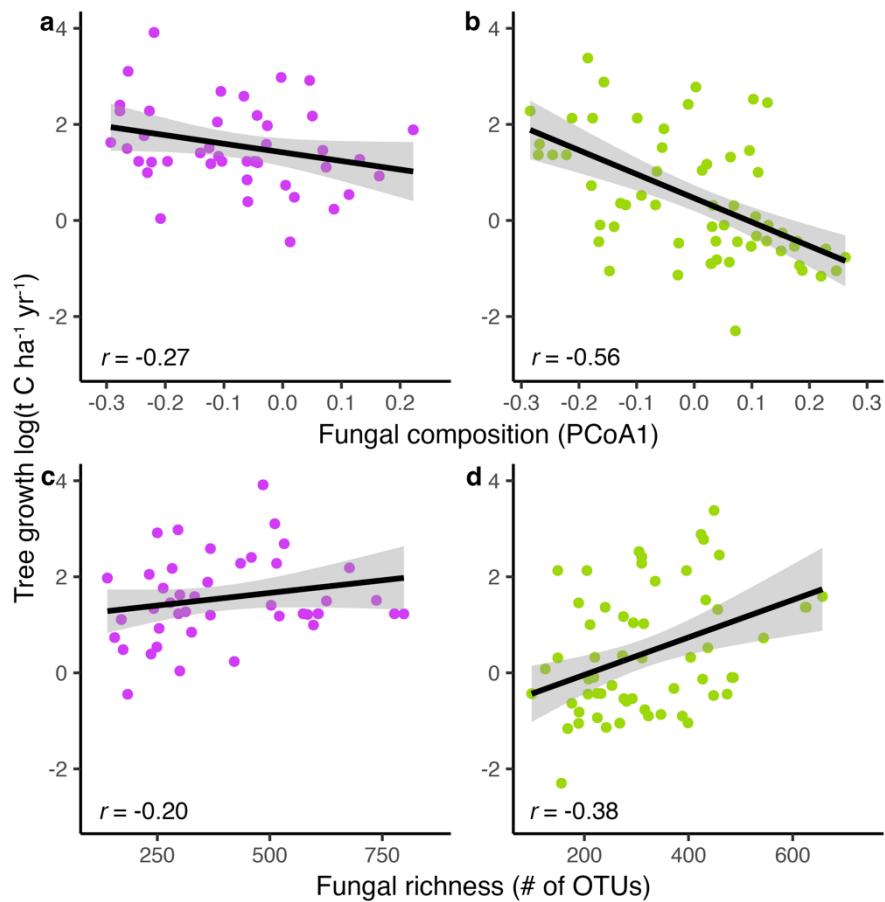
166
167
168
169
170
171
172
173
174
175
176
177
178
179
180
181
182
183
184
185
186
187
188
189
190
191
192



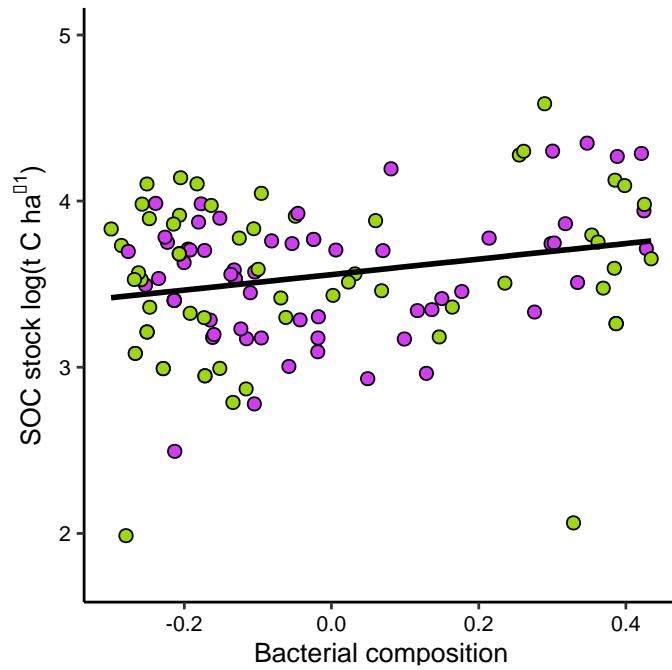
193
194
195

196 **Supplementary Fig. 1.** Scatterplots showing that microbiome composition is only correlated
197 with soil carbon (C) stocks, not soil carbon content. Green and purple points show conifers
198 and broadleaves, respectively. See Figure 4 in the main text for statistical overview of
199 significant correlations.
200
201
202
203
204
205
206
207
208
209
210
211
212
213
214
215
216
217
218
219
220
221

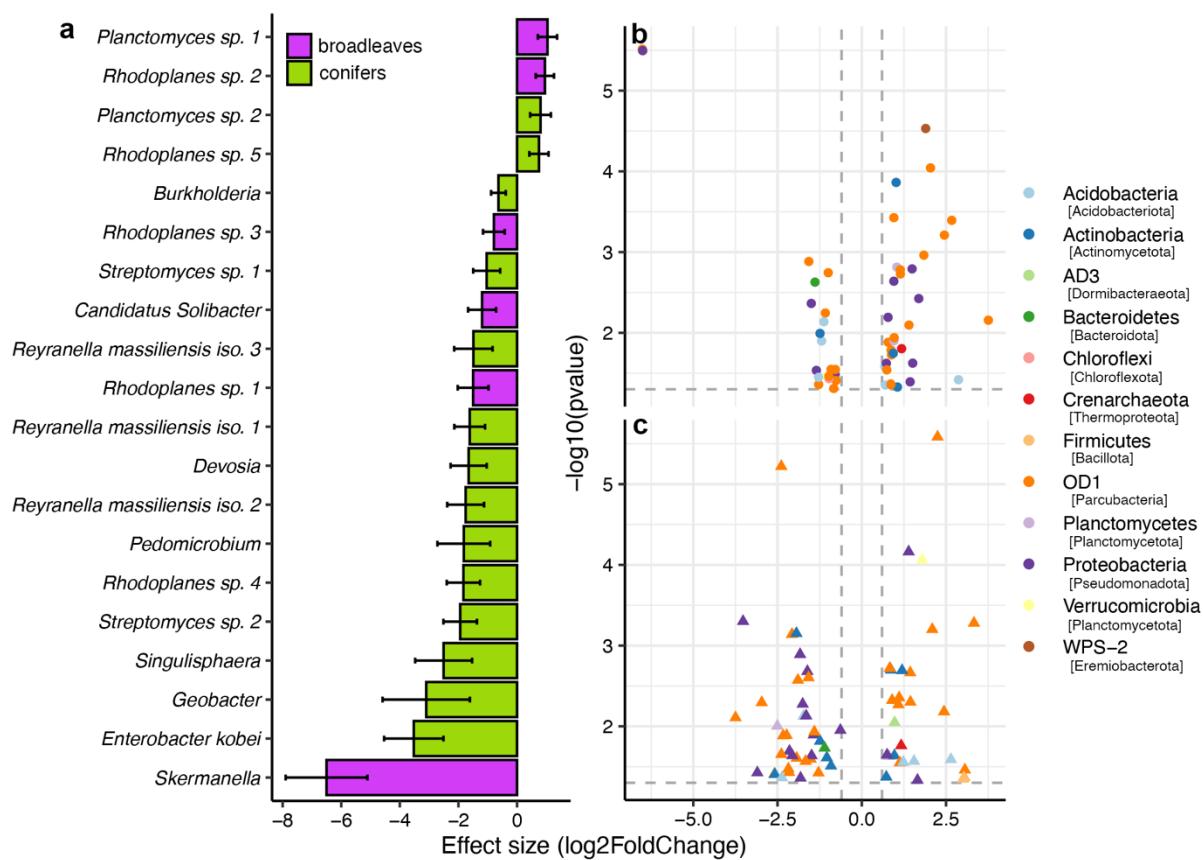
222
223



224
225 **Supplementary Fig. 2.** Correlations between tree growth and fungal composition and
226 richness separated by dominant forest tree type. Green and purple points show conifers and
227 broadleaves, respectively.
228
229
230
231
232
233
234
235
236



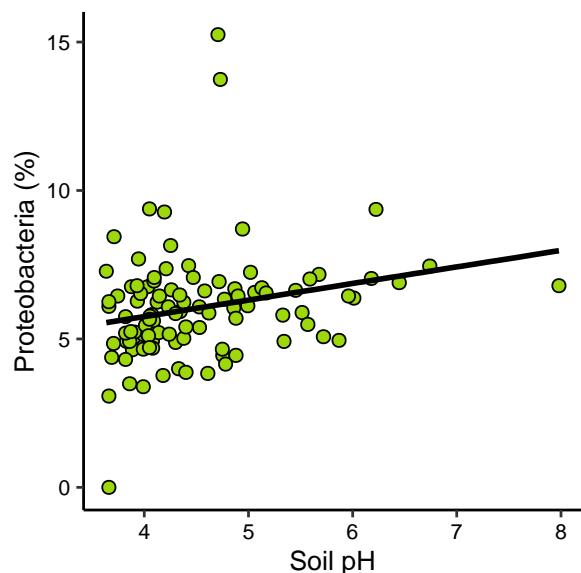
237
238
239 **Supplementary Fig. 3.** The weak correlation between bacterial community composition
240 (PCoA1) and mineral horizon SOC stocks ($r = 0.24, P = 0.006$). Only PCoA1 is shown since
241 PCoA2 was not correlated with mineral horizon SOC stocks. Green and purple points show
242 conifers and broadleaves, respectively.
243
244
245
246
247
248
249
250
251
252
253
254
255
256
257
258
259
260
261
262
263
264
265
266
267
268
269
270
271
272
273
274



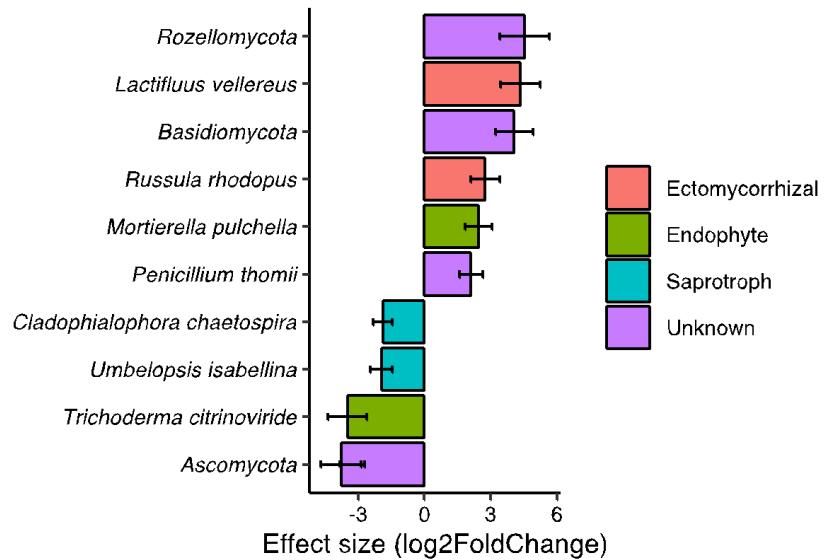
275
276

Supplementary Fig. 4. Bacterial taxa linked to variation in organic horizon soil organic carbon stocks. Significant bacterial indicator species identified from DESeq2 analysis summarized from the species to phylum level. Significant indicator species of organic horizon carbon stocks that could be assigned genus and species level taxonomies using reference databases (a). Volcano plot showing the strength of all bacterial OTUs significantly correlated to soil carbon stocks in broadleaf (b) and conifer (c) forests. Values less and more than 0 indicate negative and positive correlations with organic horizon carbon stocks, respectively. Taxonomic labels in square brackets reflect the updated International Committee on Systematics of Prokaryotes (ICSP) phyla names.

286
287
288
289
290
291
292
293
294
295
296



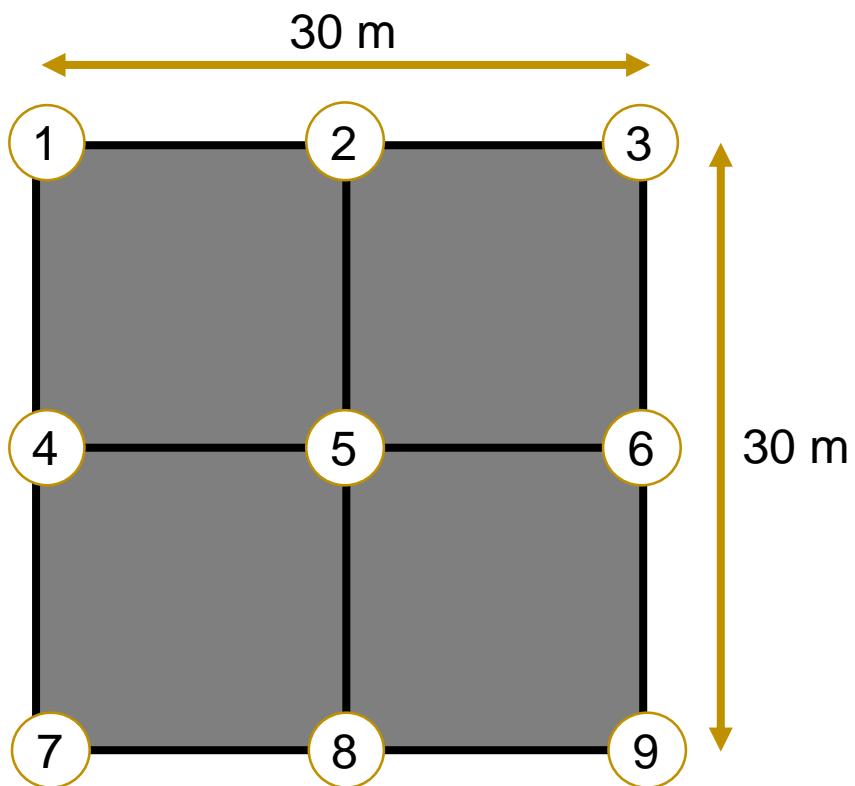
297
298 **Supplementary Fig. 5.** Correlation between Proteobacteria relative abundances and soil pH
299 in the organic horizon of coniferous forests ($r = 0.24$, $P = 0.02$)
300
301
302
303
304
305
306
307
308
309
310
311
312
313
314
315
316
317
318
319
320
321
322
323
324



325
326

Supplementary Fig. 6. Fungal taxa linked to variation in organic horizon soil organic carbon stocks. Significant fungal indicator species were identified from DESeq2 analysis (see methods for a complete description). Taxa are labelled at the finest taxonomic resolution possible. Values less and more than 0 indicate negative and positive correlations with organic horizon carbon stocks, respectively.

332
333
334
335
336
337
338
339
340
341
342
343
344
345
346
347
348
349
350
351
352
353
354
355
356
357
358



359
360
361
362
363

Supplementary Fig. 7. Visualization of the grid-design used for soil sampling. Soil cores were collected at positions 1-9 and pooled into one composite sample per depth increment (organic horizon and mineral soil at a 0-10 cm depth).