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Supplementary Materials for Mycorrhizal Association as a Primary Control of the CO₂ Fertilization Effect

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Materials and Methods

Data collection

We collected published and unpublished data on total, aboveground and belowground biomass (g m^{-2}) from eCO₂ experiments. We consulted the list of CO₂ experiments from INTERFACE (<https://www.bio.purdue.edu/INTERFACE/experiments.php>), the Global List of FACE Experiments from the Oak Ridge National Laboratory (http://facedata.ornl.gov/global_face.html), the ClimMani database on manipulation experiments (www.climmani.org), and the database described by Dieleman *et al.* (35), and freely available (https://www.researchgate.net/publication/276839560_Database_of_Global_Change_Manipulation_Experiments). We used Google Scholar to locate the most recent publications for each of the previously listed experiments, and collected data on total, aboveground and belowground plant biomass for ambient and elevated CO₂ treatments. When the data were presented in figures we extracted mean values and standard error using GraphClick. Additionally, we collected available data about the vegetation, sample size, soil fertility, land use history, MAP, MAT and the age of the vegetation at the start of the experiment. Some experiments were not included in the meta-analysis if they met any of the following exclusion criteria: i) species did not form associations with AM or ECM; ii) papers did not report biomass data; iii) standard error or standard deviation was not provided; iv) information about the fertility of the site was not reported (e.g. soil type, pH, or qualitative assessments of N-availability); v) duration of the experiment was less than 3 months.

Experimental units

Where possible, data were collected at the species level, and different species within experiments were considered independent when grown in monoculture; when available data were pooled across several species, these were only included in the analysis if all the species were associated with the same type of mycorrhizal fungi. Experiments in which the most abundant species were C₄ or N₂-fixing species were excluded from the main analysis to avoid confounding effects. Different N-fertilization treatments within experiments were considered independent. These selection criteria allowed us to assess N-status and mycorrhizal association in the individual experimental units. Overview of the experiments included in the dataset is in table S1, and the data included in the meta-analysis in figs. S1-3.

Nitrogen classification

N-classification followed a similar approach as refs. 17, 36, 37, but did not consider limitations of nutrients other than N. Experiments were classified as “low” or “high” in terms of N-availability based on the amount of N-fertilizer applied (if so), as well as the original N-availability at the sites, as a function of available data such as soil type, nitrogen and carbon content, pH, land use history, and the assessment of N-availability (reported in the literature or provided by the site principal investigators -PIs-). For example, sandy soils have an inherently low nitrogen retention capacity, and are typically N poor if not fertilized or exposed to high N deposition. The C:N ratio of soil is indicative of the decomposability of soil organic matter. Especially high C:N ratios (>25) suggest low availability of N and potential N immobilization by microorganisms (38).

For some experiments, the available soil data were scarce. We then requested direct expert assessment by the PIs. More information regarding the classification of each experiment and the underlying reasons is given in Table S2. We classified all sites that had indications of N limitation to “low N”; sites that were unlikely N-limited (e.g., where N fertilization had no effects on plant growth) were designated as “high N”. We created an alternative N-classification with an additional “medium” class that grouped all those experiments with intermediate N-availability (e.g. moderately fertile soils with no N-fertilization, or N-poor soils with modest N-fertilization, but in the range in which N-availability limits growth). This alternative classification was used as a sensitivity analysis to test that the observed effects were not driven by sites with intermediate N availability classified as “low N” in the main classification.

Mycorrhizal status classification

We used the check-list in ref. 39, with additional classifications derived from the literature, to classify plant species as ECM, or AM. Species that form associations with both ECM and AM fungi (e.g. *Populus* spp.) were classified as ECM because these species can potentially benefit from increased N-availability due to the presence of ECM-fungi, as hypothesized. Overall, CO₂ responses from species associated with AM and ECM were similar to strictly ECM species, and their exclusion did not alter the results of the meta-analysis.

N-fixing species.

When the data were presented at the plot level, with specification of the species present in each plot, all plots containing N-fixing species were not included in the main analysis because they might be particularly responsive to eCO₂ (40). We analysed the role of N-fixing species in a separate meta-analysis that included AM-species in N-limited ecosystems only, using the same methods as in the main meta-analysis, and including the responses from both N-fixing and their accompanying non N-fixing species. There were three N-limited-AM-dominated experiments that included N-fixing experiments for total biomass and seven for aboveground biomass. Therefore, the analysis of N-fixing species was performed using aboveground biomass only. The list of experiments with N-fixing species included in the analysis is in Table S3.

ΔCO₂

Ambient CO₂ treatments had concentrations ranging from 280 to 400 μmol mol⁻¹, whereas elevated CO₂ treatments had concentrations ranging from 420 to 780 μmol mol⁻¹, with an average of ~650 μmol mol⁻¹. ΔCO₂ was calculated as the natural log of the difference in CO₂ concentrations between elevated and ambient treatments: $\Delta\text{CO}_2 = \ln(\text{eC}_a/\text{aC}_a)$. Results from meta-analysis shown here were normalised for ΔCO₂ from 400 (current) to 650 (average [eCO₂]) μmol mol⁻¹, after including ΔCO₂ 400-650 as a variable in a mixed-effects meta-regression.

MAT, MAP and age of the vegetation

MAT and MAP data were collected from the original source or from WorldClim Global Climate Data (41). When the experimental units were irrigated we did not use MAP data in the analysis, but instead we assigned the maximum value of MAP in the dataset (1750

mm y^{-1}) to all irrigated experimental units. When the age of the vegetation at the start of the experiment was not specified in the study, we assigned a value of 1 for seedlings, annuals, frequently grazed vegetation, or experiments under controlled burning, and the maximum value in the dataset (50 years) when the site was classified as “intact” or similar.

Calculation of effect sizes

We used the response ratio (RR , mean response in elevated to ambient CO_2 plots) to measure effect sizes (42). We calculated the natural logarithm of the response ratio ($\log RR$) and its variance for each experimental unit to obtain a single response metric (42) in a weighted, mixed-effects model using the R package *metafor* (43). Measurements across different time-points (e.g. over several years or harvests) were considered non-independent, and we computed a combined effect across time-points so that only one effect size was analysed per experimental unit. The combined variance that takes account of the correlation among the different time-point measurements was calculated following the method described in Borenstein *et al.* (44):

$$\text{var}\left(\frac{1}{m} \sum_{i=1}^m Y_i\right) = \left(\frac{1}{m}\right)^2 \left(\sum_{i=1}^m V_i + \sum_{i \neq j} (r_{ij} \sqrt{V_i} \sqrt{V_j}) \right) \quad (1)$$

where V_i is the variance of effect size Y_i for several time-points $i=1,\dots,m$ and r_{ij} as the correlation between Y_i and Y_j , with $r=0$ equivalent to treating two outcomes as independent, underestimating the error (and overestimating the precision). We used a conservative approach with $r=1$ (assuming non independence). The outcome was not sensitive to the assumption of $r=1$, with $r=0$ (independence) and $r=0.5$ rendering slightly different SE terms (and *P-value*) that did not alter the conclusions (Table S4).

Weighting functions

Effect size measurements from individual studies in meta-analysis are commonly weighted by the inverse of the variance (45) (W_V). For this particular analysis, not only well replicated, but also long-term studies provide more reliable estimates of ecosystem CO_2 responses (46). Thus, we weighted the individual effects by both replication and experimental duration by using the function in refs 11, 47:

$$W_{NY} = (n_a * n_e) / (n_a + n_e) + (yr * yr) / (yr + yr) \quad (2)$$

with n_a and n_e as the number of replicates under ambient and elevated CO_2 , and yr as the length of the study in years. Both weighting functions were used, but W_V assigned about half of the total weight to two experiments with very low variance creating a sub-optimal imbalance, and the results using W_V are only shown for comparison purposes in Fig. S6). Results shown in the main report and figures correspond to the meta-analysis using W_{NY} as weights. In all cases, the conclusions were consistent across various weighting functions.

Calculation of the overall true effect

We used the R package *metafor* (43) to calculate overall effect sizes and 95% confidence intervals (CI). The mixed-effects meta-regression model was fitted using maximum likelihood for the amount of residual heterogeneity. The Knapp and Hartung method (48) was included as an adjustment to the standard errors of the estimated coefficients to control the Type I error rate (49). This method leads to an F-test for sets of model predictors (test of moderators) to test their significance to influence the average effect of CO₂. For individual model coefficients, the method leads to t-tests. We inferred CO₂ effects if the calculated 95 % CI did not overlap with zero. The log response ratio was back-transformed and expressed as percentage CO₂ effect ($[\log RR - 1] \times 100$) to ease interpretation in figures and text.

Model selection

We analysed the plausibility of models containing all potential combinations of the studied predictors in a mixed-effects meta-regression model using maximum likelihood estimation. For this purpose, we used the R packages *gmulti* (50) and *metafor* (43). Model selection was based on AICc. The relative importance value for a particular predictor was equal to the sum of the Akaike weights (probability that a model is the most plausible model) for the models in which the predictor appears. Hence, a predictor that is included in models with large Akaike weights will receive a high importance value. These values can be regarded as the overall support for each variable across all models. A cut-off of 0.8 was set to differentiate between important and non-essential predictors.

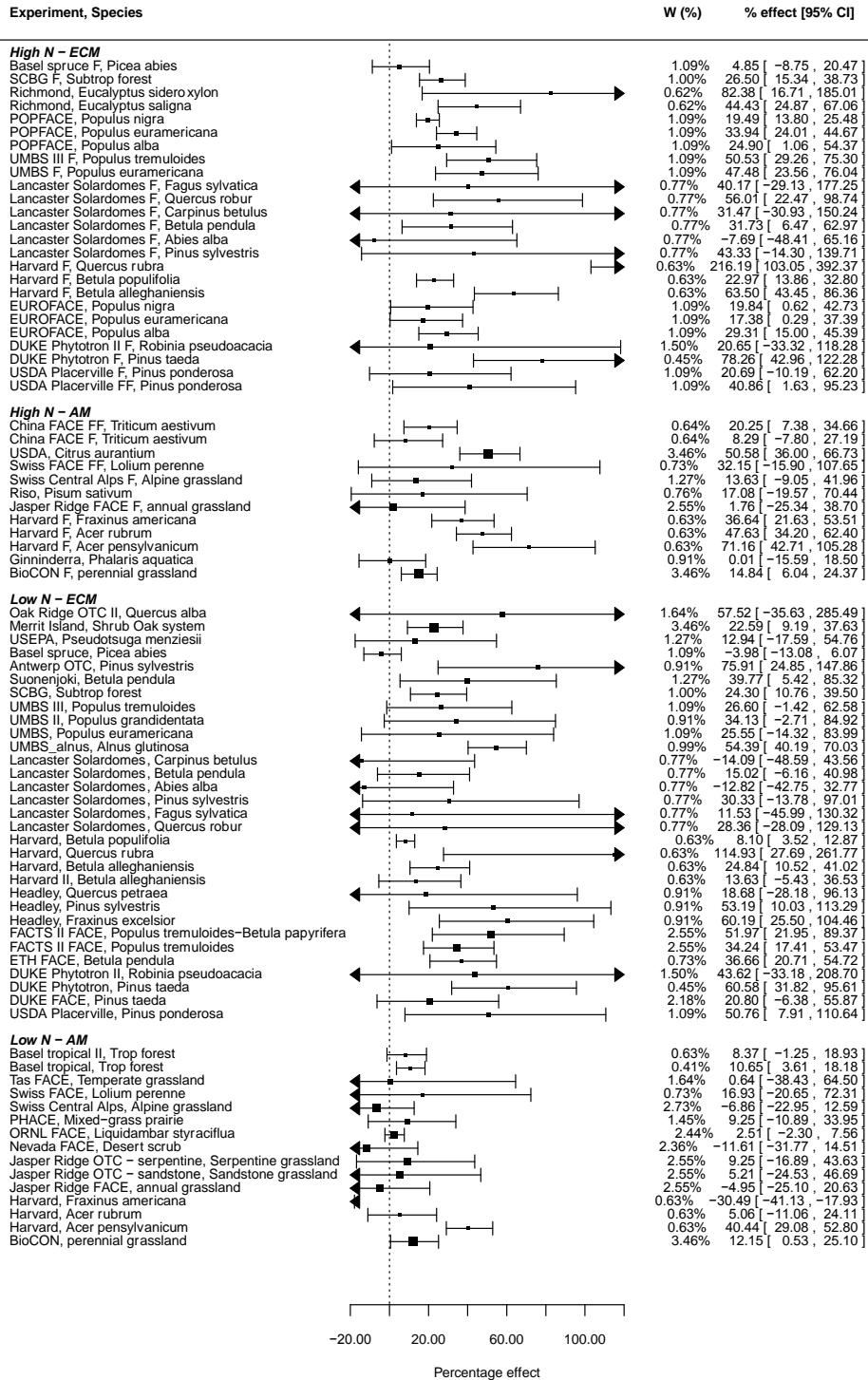


Fig. S1.

Total biomass data included in meta-analysis in Fig. 2A. W (%) are the weights used in the meta-analysis, based on the number of replicates and the length (years) of the studies.

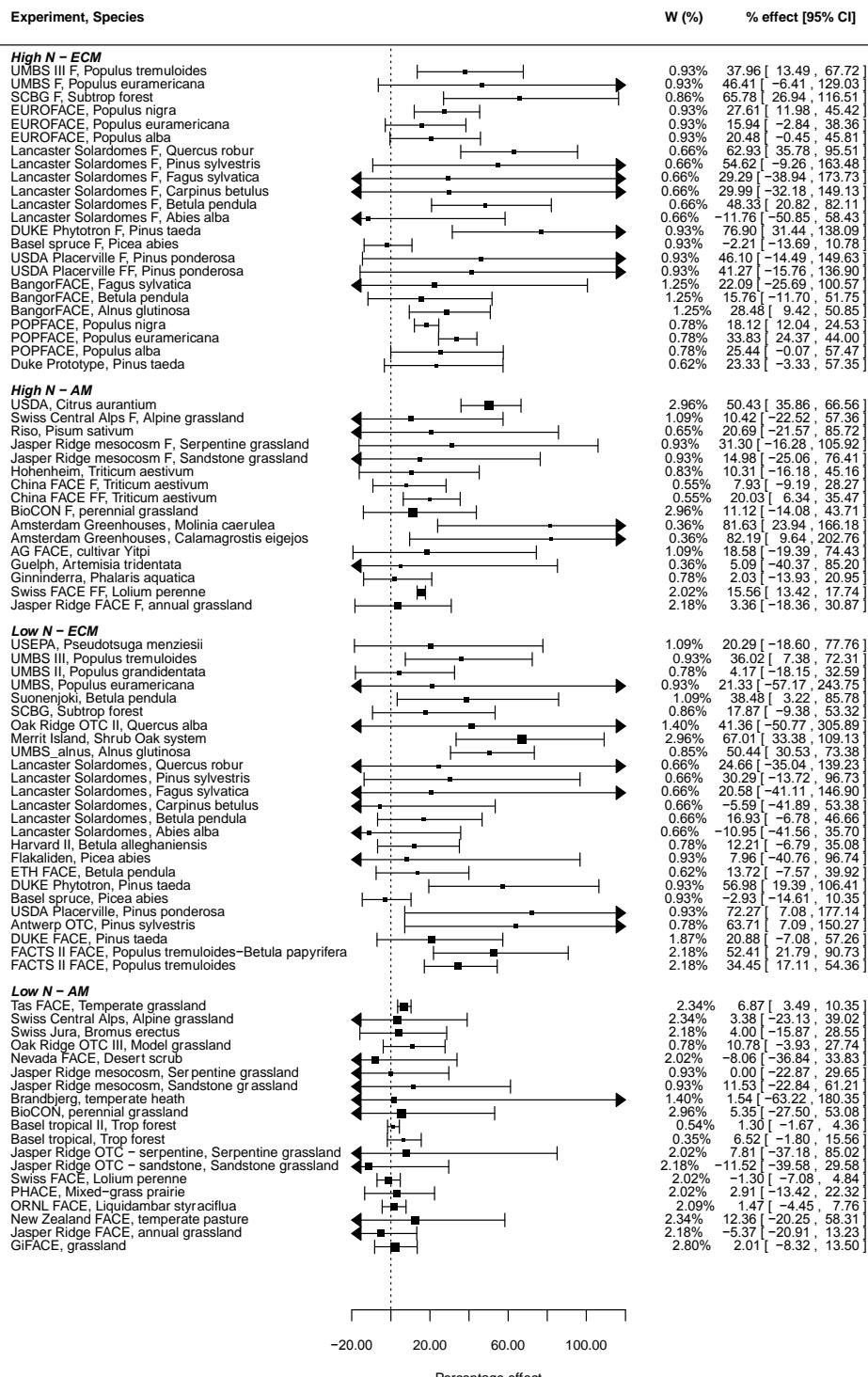


Fig. S2.

Aboveground biomass data included in meta-analysis in Fig. 2B. W (%) are the weights used in the meta-analysis, based on the number of replicates and the length (years) of the studies.

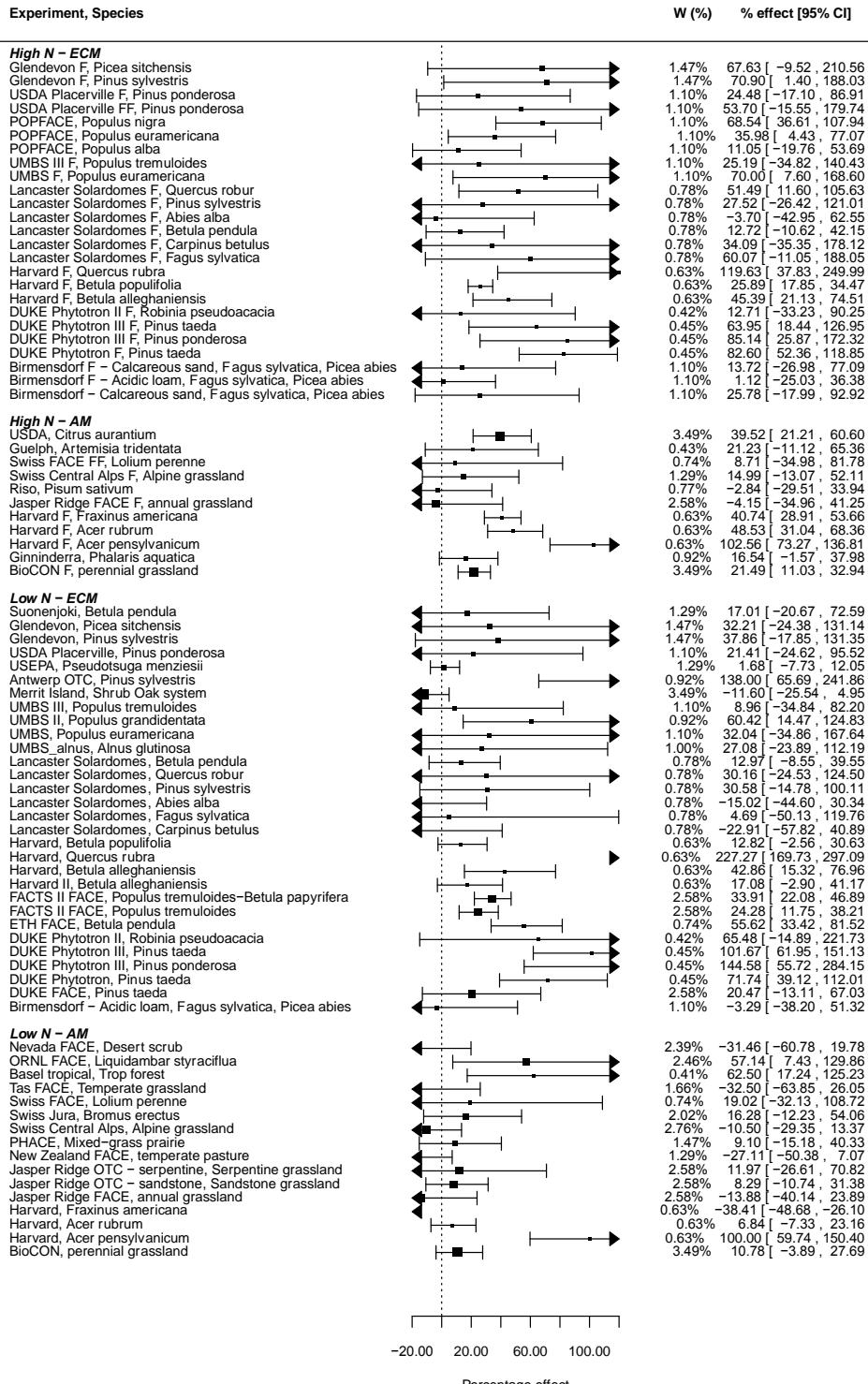


Fig. S3.

Belowground biomass data included in meta-analysis in Fig. 2C. W (%) are the weights used in the meta-analysis, based on the number of replicates and the length (years) of the studies.

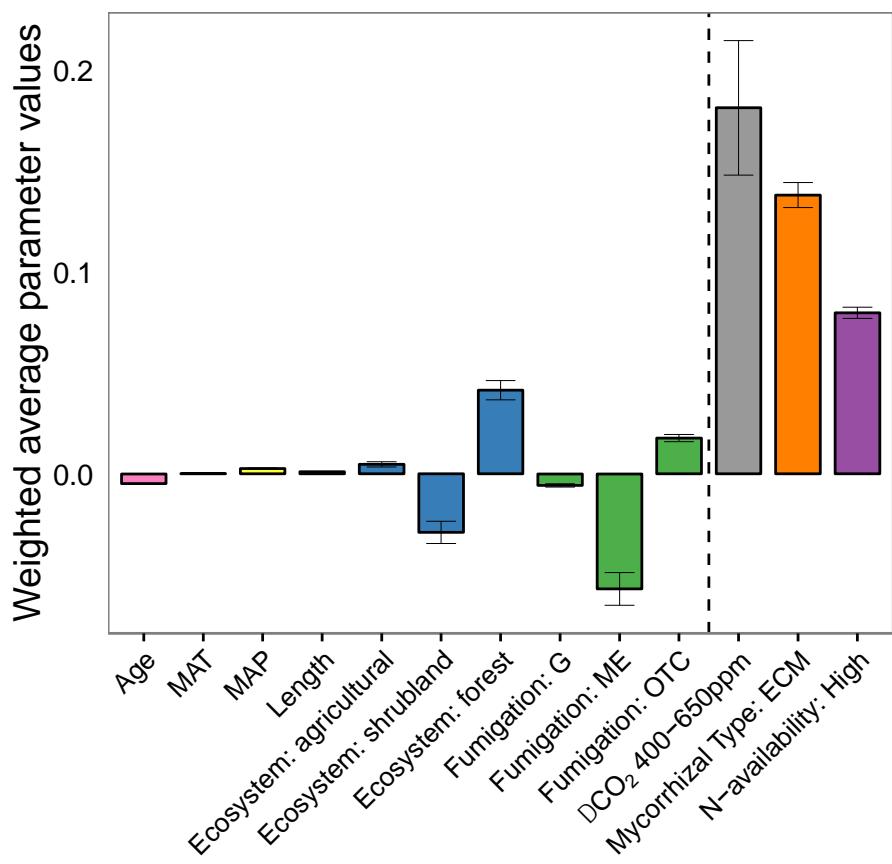


Fig. S4.

Weighted average parameter values of model coefficients. Weights equal to the model probabilities. Error bars are weighted SE. Model parameters in increasing relative importance, with predictors on the right side of the dashed line as the terms included in the AICc-selected best model and sum of Akaike weights > 0.8 . G = Greenhouse/Growth chamber, ME = Model ecosystem, OTC = Open Top Chamber, ΔCO_2 = $[\text{CO}_2]$ increment from 400 to 650 ppm. Reference parameters for qualitative factors are Fumigation: FACE, Ecosystem: grassland, Mycorrhizal type: AM, N-availability: Low.

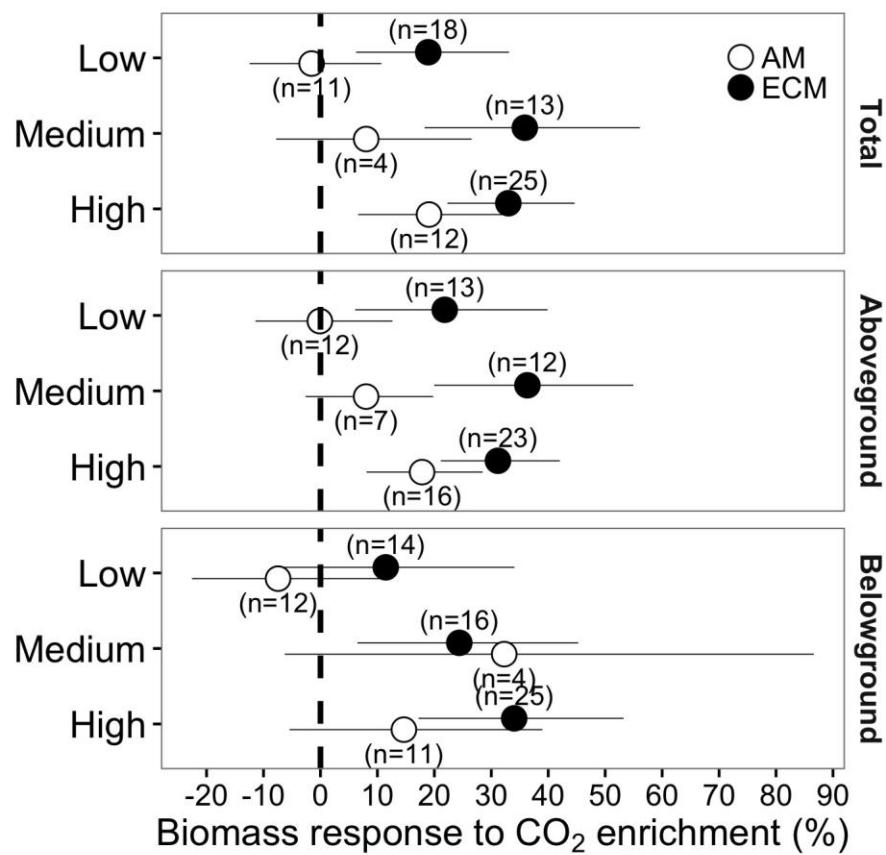


Fig. S5.

Overall effects of CO₂ on total, aboveground, and belowground biomass for two types of mycorrhizal plants species (AM: arbuscular mycorrhizae and ECM: ectomycorrhizae) in strongly N limited experiments (low N), moderately N limited experiments (medium N) or experiments that are unlikely N limited (high N). Overall means and 95% confidence intervals are given; we interpret CO₂ effects when the zero line is not crossed.

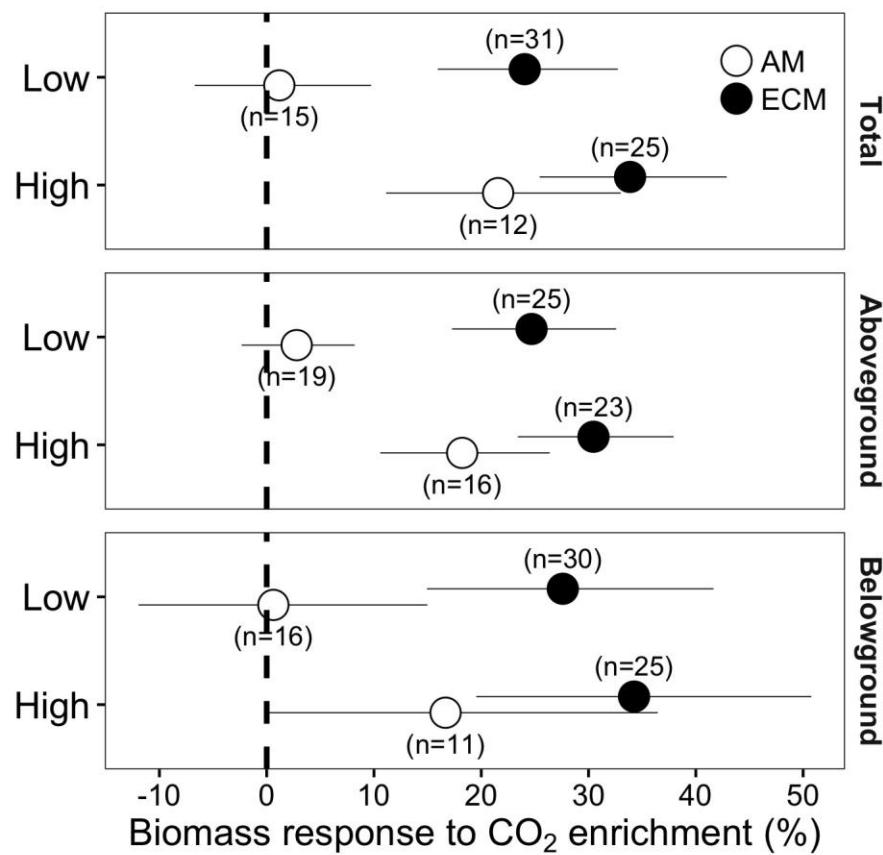


Fig. S6.

Overall effects of CO_2 on total, aboveground, and belowground biomass for two types of mycorrhizal plants species (AM: arbuscular mycorrhizae and ECM: ectomycorrhizae) in N limited experiments (low N) or experiments that are unlikely N limited (high N).

Experiments in this meta-analysis are weighted by the inverse of the variance, whereas weights in main meta-analysis in Fig. 2 are based on sample size and length (years) of the experiments. Overall means and 95% confidence intervals are given; we interpret CO_2 effects when the zero line is not crossed.

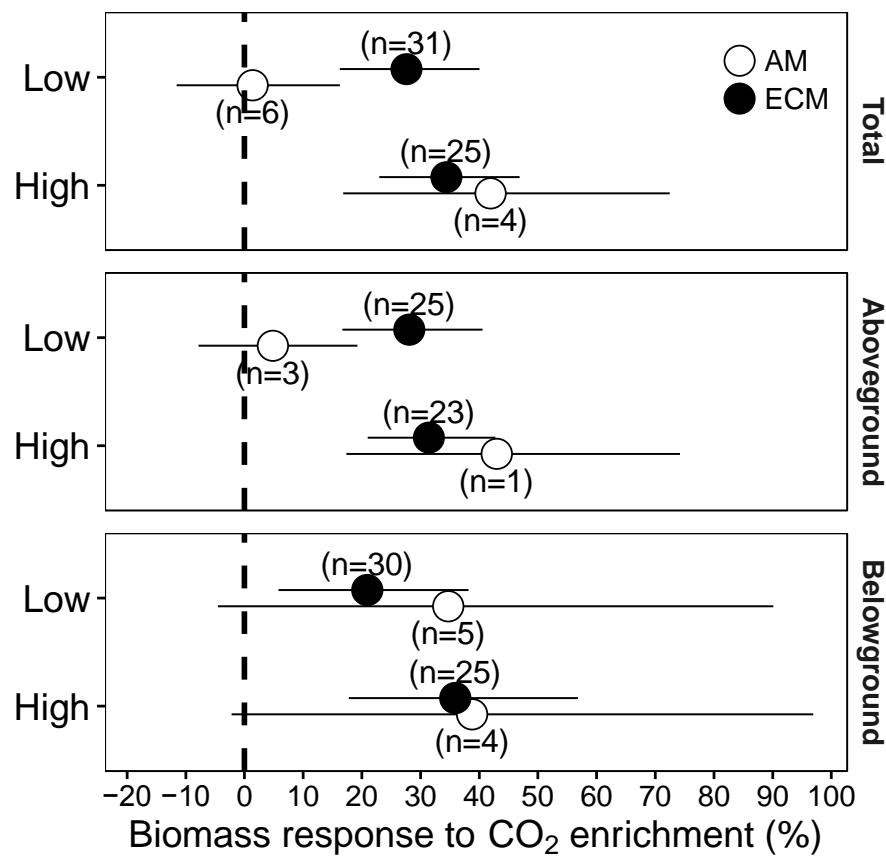


Fig. S7.

Meta-analysis output for the subset of experiments with tree species, showing the effects of CO₂ on total, aboveground, and belowground biomass for two types of mycorrhizal plants species (AM: arbuscular mycorrhizae and ECM: ectomycorrhizae) in N limited experiments (low N) or experiments that are unlikely N limited (high N). Overall means and 95% confidence intervals are given; we interpret CO₂ effects when the zero line is not crossed.

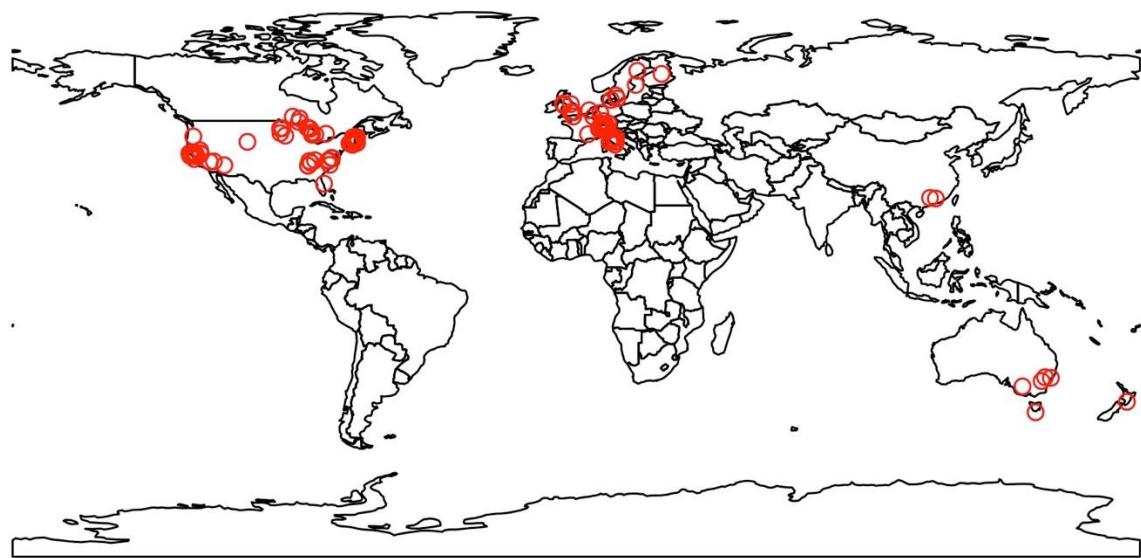


Fig. S8.

Location of elevated CO₂ experiment with total biomass data included in the dataset (Fig. S1). Experiments from the same site are spaced to avoid overlapping.

Table S1.

Overview of CO₂ enrichment experiments included in our analysis. Abbreviations: Myc: mycorrhizal type (AM: arbuscular mycorrhizae, ECM: ectomycorrhizae); N-class: main soil N availability classification (L: low, H: high); N-class2: alternative N-availability classification (L: low, M: medium, H: high); TB = Total Biomass, AB = Aboveground Biomass, BB = Belowground Biomass, FACE = Free Air Carbon Dioxide Enrichment, G = Greenhouse/Growth chamber, ME = Model ecosystem, OTC = Open Top Chamber.

Site	Species	Country	Myc	N-class	N-class2	Facility	References		
							TB	AB	BB
AG FACE	cultivar Yitpi	Australia	AM	H	H	FACE		51	
Amsterdam Greenhouses	Calamagrostis eigejos	The Netherlands	AM	H	H	G		52	
		The Netherlands	AM	H	H	G		52	
Antwerp OTC	Pinus sylvestris	Belgium	ECM	L	L	OTC	53	53	53
BangorFACE	Alnus glutinosa	UK	ECM	H	H	FACE		54	
BangorFACE	Betula pendula	UK	ECM	H	H	FACE		54	
BangorFACE	Fagus sylvatica	UK	ECM	H	H	FACE		54	
Basel spruce	Picea abies	Switzerland	ECM	L	L	ME	55	55	
Basel spruce F	Picea abies	Switzerland	ECM	H	H	ME	55	55	
Basel tropical	Trop forest	Switzerland	AM	L	M	ME	56	56	56
Basel tropical II	Trop forest	Switzerland	AM	L	M	ME	57	57	
BioCON	perennial grassland	USA	AM	L	L	FACE	5	5	5
BioCON F	perennial grassland	USA	AM	H	H	FACE	5	5	5
Birmensdorf - Acidic loam	Fagus sylvatica, Picea abies	Switzerland	ECM	L	M	OTC		58	
Birmensdorf - Calcareous sand	Fagus sylvatica, Picea abies	Switzerland	ECM	H	H	OTC		58	
Birmensdorf F - Acidic loam	Fagus sylvatica, Picea abies	Switzerland	ECM	H	H	OTC		58	
Birmensdorf F - Calcareous sand	Fagus sylvatica, Picea abies	Switzerland	ECM	H	H	OTC		58	
Brandbjerg	temperate heath	Denmark	AM	L	L	FACE		59	

China FACE F	Triticum aestivum	China	AM	H	H	FACE	60	60	
China FACE FF	Triticum aestivum	China	AM	H	H	FACE	60	60	
DUKE FACE	Pinus taeda	USA	ECM	L	M	FACE	25	25	25
DUKE Phytotron	Pinus taeda	USA	ECM	L	M	G	61	61	61
DUKE Phytotron F	Pinus taeda	USA	ECM	H	H	G	61	61	61
DUKE Phytotron II	Pinus ponderosa	USA	ECM	L	M	G			62
DUKE Phytotron II	Pinus taeda	USA	ECM	L	M	G			62
DUKE Phytotron II F	Pinus ponderosa	USA	ECM	H	H	G			62
DUKE Phytotron II F	Pinus taeda	USA	ECM	H	H	G			62
Duke Prototype	Pinus taeda	USA	ECM	H	H	FACE		8	
ETH FACE	Betula pendula	Switzerland	ECM	L	M	FACE	63	63	63
EUROFACE	Populus alba	Italy	ECM	H	H	FACE	64	64	
EUROFACE	Populus euramericana	Italy	ECM	H	H	FACE	64	64	
EUROFACE	Populus nigra	Italy	ECM	H	H	FACE	64	64	
FACTS II FACE	Populus tremuloides	USA	ECM	L	M	FACE	9	9	9
FACTS II FACE	Populus tremuloides-Betula papyrifera	USA	ECM	L	M	FACE	9	9	9
Flakaliden	Picea abies	Sweden	ECM	L	L	OTC		65	
GiFACE	grassland	Germany	AM	L	M	FACE		66	
Ginninderra	Phalaris aquatica	Australia	AM	H	H	G	67	67	67
Glendevon	Pinus sylvestris	UK	ECM	L	M	OTC			68
Glendevon	Picea sitchensis	UK	ECM	L	M	OTC			68
Glendevon F	Pinus sylvestris	UK	ECM	H	H	OTC			68
Glendevon F	Picea sitchensis	UK	ECM	H	H	OTC			68
Guelph	Artemisia tridentata	Canada	AM	H	H	G		69	69
Harvard	Acer pensylvanicum	USA	AM	L	L	G	70		70
Harvard	Acer rubrum	USA	AM	L	L	G	70		70

Harvard	<i>Betula alleghaniensis</i>	USA	ECM	L	L	G	70	70
Harvard	<i>Fraxinus americana</i>	USA	AM	L	L	G	70	70
Harvard	<i>Quercus rubra</i>	USA	ECM	L	L	G	70	70
Harvard	<i>Betula populifolia</i>	USA	ECM	L	L	G	70	70
Harvard F	<i>Acer pensylvanicum</i>	USA	AM	H	H	G	70	70
Harvard F	<i>Acer rubrum</i>	USA	AM	H	H	G	70	70
Harvard F	<i>Betula alleghaniensis</i>	USA	ECM	H	H	G	70	70
Harvard F	<i>Betula populifolia</i>	USA	ECM	H	H	G	70	70
Harvard F	<i>Fraxinus americana</i>	USA	AM	H	H	G	70	70
Harvard F	<i>Quercus rubra</i>	USA	ECM	H	H	G	70	70
Harvard II	<i>Betula alleghaniensis</i>	USA	ECM	L	L	G	71	71
Headley	<i>Fraxinus excelsior</i>	UK	ECM	L	L	OTC	72	
Headley	<i>Pinus sylvestris</i>	UK	ECM	L	L	OTC	72	
Headley	<i>Quercus petraea</i>	UK	ECM	L	L	OTC	72	
Hohenheim	<i>Triticum aestivum</i>	Germany	AM	H	H	FACE		73
Jasper Ridge FACE	annual grassland	USA	AM	L	L	FACE	74	74
Jasper Ridge FACE F	annual grassland	USA	AM	H	H	FACE	74	74
Jasper Ridge OTC	Sandstone grassland	USA	AM	L	L	OTC	75, 76	75-77
Jasper Ridge OTC	Serpentine grassland	USA	AM	L	L	OTC	75	75, 77
Jasper Ridge mesocosm	Sandstone grassland	USA	AM	L	L	G		78
Jasper Ridge mesocosm	Serpentine grassland	USA	AM	L	L	G		78
Jasper Ridge mesocosm F	Sandstone grassland	USA	AM	H	H	G		78
Jasper Ridge mesocosm F	Serpentine grassland	USA	AM	H	H	G		78
Lancaster Solardomes	<i>Quercus robur</i>	UK	ECM	L	L	G	79	79
Lancaster Solardomes	<i>Fagus sylvatica</i>	UK	ECM	L	L	G	79	79
Lancaster Solardomes	<i>Pinus sylvestris</i>	UK	ECM	L	L	G	79	79
Lancaster Solardomes	<i>Abies alba</i>	UK	ECM	L	L	G	79	79

Lancaster Solardomes	<i>Betula pendula</i>	UK	ECM	L	L	G	79	79	79
Lancaster Solardomes	<i>Carpinus betulus</i>	UK	ECM	L	L	G	79	79	79
Lancaster Solardomes F	<i>Pinus sylvestris</i>	UK	ECM	H	H	G	79	79	79
Lancaster Solardomes F	<i>Abies alba</i>	UK	ECM	H	H	G	79	79	79
Lancaster Solardomes F	<i>Betula pendula</i>	UK	ECM	H	H	G	79	79	79
Lancaster Solardomes F	<i>Carpinus betulus</i>	UK	ECM	H	H	G	79	79	79
Lancaster Solardomes F	<i>Quercus robur</i>	UK	ECM	H	H	G	79	79	79
Lancaster Solardomes F	<i>Fagus sylvatica</i>	UK	ECM	H	H	G	79	79	79
Merrit Island	Shrub Oak system	USA	ECM	L	L	OTC	80	80	80
Nevada FACE	Desert scrub	USA	AM	L	L	FACE	81	81	81
New Zealand FACE	temperate pasture	New Zealand	AM	L	M	FACE		82	83
Oak Ridge OTC	<i>Liriodendron tulipifera</i>	USA	AM	-	-	OTC	84	84	84
Oak Ridge OTC	<i>Acer saccharum, Acer rubrum</i>	USA	AM	-	-	OTC			85
Oak Ridge OTC II	<i>Quercus alba</i>	USA	ECM	L	M	OTC	86	86	
Oak Ridge OTC III	Model grassland	USA	AM	L	M	OTC		87	
ORNL FACE	<i>Liquidambar styraciflua</i>	USA	AM	L	M	FACE	6	6	88
PHACE	Mixed-grass prairie	USA	AM	L	L	FACE	89	89	89, 90
POPFACE	<i>Populus alba</i>	Italy	ECM	H	H	FACE	91	91	91
POPFACE	<i>Populus euramericana</i>	Italy	ECM	H	H	FACE	91	91	91
POPFACE	<i>Populus nigra</i>	Italy	ECM	H	H	FACE	91	91	91
Richmond	<i>Eucalyptus saligna</i>	Australia	ECM	H	H	G	92		
Richmond	<i>Eucalyptus sideroxylon</i>	Australia	ECM	H	H	G	92		
Riso	<i>Pisum sativum</i>	Denmark	AM	H	H	G	93	93	93
SCBG	Subtrop forest	China	ECM	L	M	OTC	94	94	
SCBG F	Subtrop forest	China	ECM	H	H	OTC	94	94	
Suonenjoki	<i>Betula pendula</i>	Finland	ECM	L	M	OTC	95	95	96

Swiss Central Alps	Alpine grassland	Switzerland	AM	L	L	OTC	97	97	97
Swiss Central Alps F	Alpine grassland	Switzerland	AM	H	H	OTC	97	97	97
Swiss FACE	Lolium perenne	Switzerland	AM	L	M	FACE	98	99	98
Swiss FACE FF	Lolium perenne	Switzerland	AM	H	H	FACE	98	99	98
Swiss Jura	Bromus erectus	Switzerland	AM	L	L	G		100	101
Tas FACE	Temperate grassland	Australia	AM	L	L	FACE	102	14	102
UMBS	<i>Populus euramericana</i>	USA	ECM	L	L	OTC	103	103	103
UMBS F	<i>Populus euramericana</i>	USA	ECM	H	H	OTC	103	103	103
UMBS II	<i>Populus grandidentata</i>	USA	ECM	L	L	OTC	104	104	104
UMBS III	<i>Populus tremuloides</i>	USA	ECM	L	M	OTC	105	105	105
UMBS III F	<i>Populus tremuloides</i>	USA	ECM	H	H	OTC	105	105	105
UMBS_alnus	<i>Alnus glutinosa</i>	USA	ECM	L	M	OTC	106	106	106
USDA	<i>Citrus aurantium</i>	USA	AM	H	H	OTC	107	107	108
USDA Placerville	<i>Pinus ponderosa</i>	USA	ECM	L	M	OTC	109	109	109
USDA Placerville F	<i>Pinus ponderosa</i>	USA	ECM	H	H	OTC	109	109	109
USDA Placerville FF	<i>Pinus ponderosa</i>	USA	ECM	H	H	OTC	109	109	109
USEPA	<i>Pseudotsuga menziesii</i>	USA	ECM	L	M	G	110	110	110

Table S2.

Justification for the soil nitrogen (N) availability classification. N-class: main soil N availability classification (L: low, H: high); N-class2: alternative N-availability classification (L: low, M: medium, H: high); N-fert: fertilized site (yes or no) and indication of the amount of N fertilizer in g N m⁻² y⁻¹, unless other units are specified; %N: soil N content (%); %C: soil carbon content (%); pH: when available pH in CaCl₂ was reported, otherwise from water solution; C:N: C:N ratio; Report: N-availability or soil fertility assessment of the site found in the literature or confirmed by the site PI. Lack of information on N-availability in some experiments did not allow to assess them in N-class, but were classified as “medium” in N-class2.

Site	N-class	N-class2	N-fert.	Extra fert.	Soil type	Soil texture	%N	%C	pH	C:N	Ref	Remarks
AG FACE	H	H	0 -13.8	P, S		clay (60%)	0.03 - 0.10	1.24	8.4	12	51	1
Amsterdam Greenhouses	H	H	yes	P, K					5.5		52	2
Antwerp OTC	L	L	no		poor forest soil	sandy	0.12		4.3		53	3
BangorFACE	H	H	no		Dystric Cambisol	Fine loamy brown earth over gravel; 62.2 sand, 28.5 silt, 9.3 clay	2.6	4.6	10.5	111	4	
Basel spruce	L	L	no		podzol				4.5		55	5
Basel spruce F	H	H	9		podzol				4.5		55	6
Basel tropical	L	M	13.3	fertilizer pellets	fresh tropical soil						56	7
Basel tropical II	L	M	11.8	Osmocote and OM	fresh tropical soil						57	8
BioCON	L	L	no		Nymore series, subgroup Typic Upidsamment, subborder Psamments, Order Entisols	93% sand, 3% silt, and 4% clay	0.001				112	9
BioCON F	H	H	4		Nymore series, subgroup Typic Upidsamment, subborder Psamments, Order Entisols	93% sand, 3% silt, and 4% clay	0.001				112	10
Birmensdorf - Acidic loam	L	M	0.5 - 0.7		Haplic Halisol	acidic sandy loamy; 55% sand, 29% silt, 16% clay	12.9 mg kg ⁻¹	4.1			113	11
Birmensdorf - Calcareous sand	H	H	0.5 - 0.7		Fluvisol	calcareous loamy sandy; 84% sand, 10% silt, 6% clay	13.1 mg kg ⁻¹	7.2			113	12
Birmensdorf F - Acidic loam	H	H	5 - 7		Haplic Halisol	acidic sandy loamy; 55% sand, 29% silt, 16% clay	12.9 mg kg ⁻¹	4.1			113	13

Birmensdorf F - Calcareous sand	H	H	5 - 7		Fluvisol	calcareous loamy sandy; 84% sand, 10% silt, 6% clay	13.1 mg kg ⁻¹	7.2	113	14		
Brandbjerg	L	L	no			sandy deposit hill		5	59	15		
China FACE F	H	H	15	P, K	Shajiang Aquic Cambisol	sandy-loamy ; total porosity: 54%; clay 13.6%, silt 28.5%, sand 57.8%	0.145	1.84	7.2	60	16	
China FACE FF	H	H	25	P, K	Shajiang Aquic Cambiosol	sandy-loamy ; total porosity: 54%; clay 13.6%, silt 28.5%, sand 57.8%	0.145	1.84	7.2	60	17	
DUKE FACE	L	M	no		Ultic Hapludalfs	Clay loam; well-developed soil horizons with mixed clay mineralogy.	0.079	5.75	18.9	114	18	
DUKE Phytotron	L	M	1.75 mM	Hoagland solution	mixture of Turface, vermiculite, gravel and soil (4:2:2:1)				61	19		
DUKE Phytotron F	H	H	5.5 mM	Hoagland solution	mixture of Turface, vermiculite, gravel and soil (4:2:2:1)				61	20		
DUKE Phytotron II	L	M	1 mM	Hoagland solution	sterilized sand				62	21		
DUKE Phytotron II F	H	H	5 mM	Hoagland solution	sterilized sand				62	22		
Duke Prototype	H	H	11.2		Ultic Hapludalfs	Clay loam		5.75	18.9	8	23	
ETH FACE	L	M	no		from an agricultural site, used for maize cultivation since 1962	5.6% clay, 17.7% loam, 76.8% sand	0.08	5.05	63	24		
EUROFACE	H	H	21.2- 29	P, K	Xeric Alfisol	heavy clay loam; 37% sand, 44% silt, 19% clay	0.13	1.06- 1.13	4.89- 5.18	9.31	64	25
FACTS II FACE	L	M	no		Alfic Haplorthods	Mixed, frigid, coarse loamy ; 56% sand, 36% silt, 8% clay	0.12		5.5	12.9- 13.5	115	26
Flakaliden	L	L	no		Typic Haplocryods	silty-sandy till; O-layer average depth is 3cm		4.4		65	27	
GiFACE	L	M	4		stagnno-fluvic gleysol	porosity 60 - 65%; loamy- sandy sediments over clay	0.45	4.7	5.9	10.5	116	28
Ginninderra	H	H	10	P, K						67	29	
Glendevon	L	M	no		brown forest soil 40-60 cm deep.	loam of shallow brown earth, locally podzolized	NO ₃ : 0.49; NH ₄ : 0.26	4.7		117	30	
Glendevon F	H	H	7	other nutrients	brown forest soil 40-60 cm deep.	loam of shallow brown earth, locally podzolized	NO ₃ : 0.54; NH ₄ : 0.22	4.7		117	31	

Guelph	H	H	400 ml	Hoagland solution	Turface		69	32				
Harvard	L	L	0.18 g	P, K + micronutrients	pots with a 1:1:1 mixture of sand:perlite:peat	5	70	33				
Harvard F	H	H	1.8 g	P, K + micronutrients	pots with a 1:1:1 mixture of sand:perlite:peat	5	70	34				
Harvard II	L	L	no		Canton low density O2 horizon; stony to sandy loams		71	35				
Headley	L	L	no		humo-ferric podzol	Sandy		72	36			
Hohenheim	H	H	14		slightly stagnic luvisol			73	37			
Jasper Ridge FACE	L	L	no		Typic Haploxeralfs			74	38			
Jasper Ridge FACE F	H	M	7		Typic Haploxeralfs			74	39			
Jasper Ridge OTC - serpentine	L	L	no		Lithic Haploxerolls	Clay loam	0.16	1.8	6.6	11.2	118	40
Jasper Ridge OTC - sandstone	L	L	no		Lithic Xerochrepts	Loamy	0.12	1.2	5.5	10	76	41
Jasper Ridge mesocosm	L	L	no		0.8 m subsoil from serpentine quarry and 0.15 m serpentine topsoil						78	42
Jasper Ridge mesocosm F	H	H	20	P, K	0.8 m subsoil from serpentine quarry and 0.15 m serpentine topsoil						78	43
Lancaster Solardomes	L	L	no		Udertic Paleustoll	silt loam or silty clay loam (clay 26-34%)					79	44
Lancaster Solardomes F	H	H	2.5 g L ⁻¹	P, K, Mg and trace elements	Udertic Paleustoll	silt loam or silty clay loam (clay 26-34%)					79	45
Merrit Island	L	L	no		Pomello (Arenic Haplhumod) and Poala sands (Spodic Quartzipsamment)	moderately well drained sandy soils	2-7	3.9-4.1			119	46
Nevada FACE	L	L	no		Aridosols derived from calcareous alluvium	Loamy and coarse sand; well-drained	0.01-0.08	0.18-1.8	7-8		120	47
New Zealand FACE	L	M	no	P, S, K	Mollie Psammaquent	fine sand; 0.25m black loamy top horizon underlain by grayish-brown horizon	0.37-0.41	4.52-5.02	5.9-6	12.4	82, 121	48
Oak Ridge OTC	-	-	no								84	49

Oak Ridge OTC II	L	M	no		silt loam				86	50	
Oak Ridge OTC III	L	M	no	Typic Fragiudult	well-drained; fine-silty, siliceous, mesic				87	51	
ORNL FACE	L	M	no	Aquic Hapludult	silty clay loam, moderately well drained; 21% sand, 55% silt, 24% clay	0.112	1.08	5.7	114	52	
PHACE	L	L	no	Aridic Argiustoll	fine-loamy, mixed mesic			7.9	89	53	
POPFACE	H	H	no	Xeric Alfisol	loam; 37% sand, 44% silt, 19% clay	0.11-0.14	0.9 - 1.13	4.9-5.18	8.7-9.9	91	54
Richmond	H	H	0.2 g N L ⁻¹	P, K, S, Fe, Mn, B	loamy sand	<1 mg kg ⁻¹	0.7	5.5	92	55	
Riso	H	H	20 mg N kg ⁻¹	from an arable layer	49.9% sand, 31.8% silt, 16% clay	0.14	1.36		122	56	
SCBG	L	M	no	from an evergreen broadleaved forest					94	57	
SCBG F	H	H	10	from an evergreen broadleaved forest					94	58	
Suonenjoki	L	M	2.2 - 4.1	soil composed of sand and clay; no humus layer on top of the mineral soil	0.046			21	123	59	
Swiss Central Alps	L	L	no	alpine stagnic pseudo-gleysols				4	97	60	
Swiss Central Alps F	H	H	4	P, K	alpine stagnic pseudo-gleysols			4	97	61	
Swiss FACE	L	M	10 - 14	eutric Cambisol	clay loam; 28% clay, 33% silt, 36% sand	0.28-0.46	2.9-5.1	6.5-7.6	124	62	
Swiss FACE FF	H	H	40 -56	eutric Cambisol	clay loam; 28% clay, 33% silt, 36% sand	0.28-0.46	2.9-5.1	6.5-7.6	124	63	
Swiss Jura	L	L	no	P	silty clay-loam underlain with calcareous debris.	0.33	7-8	100, 125	64		
Tas FACE	L	L	no	black Vertisol	formed of basaltic clay	0.2	6	126	65		
UMBS	L	L	no	Rubicon sand + Kalkaska series topsoil		0.45-0.46	1		127	66	
UMBS F	H	H	no	Kalkaska series topsoil		1.5-1.52	2.7		127	67	
UMBS II	L	L	4.5	Entic Haplorthod	sandy, mixed, frigid	0.0079-0.01			104	68	
UMBS III	L	L	no	Rubicon sand + Kalkaska series topsoil	93% sand, 2.5% clay	0.021	6.74	14.8	105	69	
UMBS III F	H	H	no	Kalkaska series topsoil	72% sand, 10.1% clay	0.097	6.08	13.3	105	70	

UMBS_almus	L	M	no	Rubicon sand + Kalkaska series topsoil		0.016		106	71	
						- 0.020				
USDA	H	H	ample	ample	Avondale	loam		107	72	
USDA Placerville	L	L	no		Aiken clay loam		0.09	5.1 - 5.5	24- 25	
USDA Placerville F	H	M	yes		Aiken clay loam		0.1	5.1 - 5.5	24- 25	
USDA Placerville FF	H	H	yes		Aiken clay loam		0.11	5.1 - 5.5	24- 25	
USEPA	L	M	no		Typic Hapludand	coarse, loamy, mixed, frigid	0.06- 0.11	6.2- 6.3	110	76

1. Large soil mineral N content ($\sim 300 \text{ kg N ha}^{-1}$) at the site precluded any significant effect of applied N, indicating the site was initially N-rich.
2. The experiment simulates conditions of mesotrophic soils, thereby, inherently fertile.
3. Sandy soils with low pH, classified by the authors as “poor forest soils”.
4. Former agricultural field, fertile soil type and low C:N, therefore high nitrogen availability. Analysis of P-availability indicates that plants in this site are P-limited, but not N-limited (pers. comm).
5. Authors reported the soil is “natural nutrient-poor montane soil”.
6. N-fertilization in the site increased fertility from “nutrient-poor” to “medium-high” N availability, as reported by the authors.
7. Low-fertility litter compost mix was added to the soil to simulate nutrient cycling, but no fertilizer was applied. Nutrients were kept low, and plants showed visual signs of nutrient limitations in CO_2 -fumigated plots as seen by the yellowish appearance of the vegetation. PI described the soils as N-low to moderately fertile (pers. comm).
8. Low-fertility litter compost mix was added to the soil to simulate nutrient cycling, but no fertilizer was applied. Nutrients were kept low, and plants showed visual signs of nutrient limitations in CO_2 -fumigated plots as seen by the yellowish appearance of the vegetation. PI described the soils as N-low to moderately fertile (pers. comm).
9. Authors reported that plants in this low SOM (1.4%), low N ($10 \mu\text{g g}^{-1}$) and high P content ($46.5 \mu\text{g g}^{-1}$) sandy soil were “N-limited”. In addition, N-availability constrained the CO_2 biomass response (128).
10. Same soils as in 9, but N-amended with 4 g N m^{-2} , corresponding to high N deposition rates.
11. Authors reported this acidic soil as “nutrient-poor”, with low SOC content in the subsoil (2.3 g kg^{-1}). N-addition simulated “low levels of N deposition”, and higher levels of N-fertilization in adjacent plots increased growth further, indicating N-limitations in these plots, therefore N-class2=M.

12. This Fluvisol is reported as “nutrient-rich” by the authors, with high SOC content, pH and CEC (127 mmol/kg soil), therefore N-class=H. Although N-addition addition levels simulated “low levels of N deposition”, increasing N-fertilization in adjacent plots did not increase biomass further, therefore N-class2=H.
13. Soils in 11 with higher levels of N-fertilization.
14. Soil in 12 with higher levels of N-fertilization.
15. Sandy soils reported as “nutrient-poor”.
16. Soil type and texture indicate intermediate fertility, but fertilization is high.
17. Soils in 16 with even higher fertilization.
18. Soil type and high C:N ratio indicate low N-availability. The soil is classified as “moderately low fertile” by the authors (8), and forest production showed a substantial response to N fertilization (129), indicating N-limitations. However, plants initially had not yet fully explored soil resources due to high initial spacing among seedlings (expanding systems), which may increase N availability at the individual plant level (130), therefore N-class2=M.
19. Artificial soil with modest N-fertilization. The authors reported that “N is believed to be the primary limiting factor”. Based on the scarce soil data, the soil was classified as L-M despite N-fertilization, because fertilization with higher amount of N in soil 20 increased biomass by 20%.
20. Same soil as 19 with higher N fertilization.
21. Available soil data scarce, but artificial soil (sand) with modest N-fertilization.
22. Same soil as 21 with higher N fertilization.
23. Same soil as 18 with N amendments.
24. CEC is low and the site was not N-fertilized, but it was formerly a maize field, reason we assumed it was fertilized in the past and we assigned N-class2=M.
25. Fertile soils (Alfisol) with good texture (loam) and former agriculture land. The site was classified as “nutrient-rich” by the authors. N-fertilization in the second rotation of the experiment did not enhance plant growth, indicating high N availability.
26. According to the authors N-availability is medium due to previous agricultural use prior to 1972, hence N-class2=M. N-class=L because the soil is sandy, SOM is relatively low (pers. comm) and it is not fertilized.
27. Boreal forest, classified as “strongly nutrient limited” (131). Long term (25 years) fertilization of experimental plots in this forest quadrupled productivity (pers. comm.).
28. Classified as “nitrogen limited” by the authors (pers. comm). The fertilization rate is smaller than what is removed by the harvest, so the site is considered N limited even though it is fertilized (pers. comm), hence N-class=L. However, the soil is moderately fertile based on soil texture and intermediate C:N, therefore N-class2=M.
29. No soil information was available, but N and other nutrients are supplied in abundance.

30. The soil was classified as “intermediate nutrient status” by the authors, hence N-class2=M. Based on the lack of fertilization, N-class=L.
31. Same soil as 30 with intermediate nutrient availability with extra N and other nutrients.
32. Soil was sterilized Turface, low-nutrient calcined clay (AM fungi inoculation), but plants were fertilized frequently with Hoagland’s solution.
33. The soil “simulated poor-nutrient forest soil at Harvard Forest”. They further showed that nitrogen mineralization rates were low in this forest ($34 \text{ kg ha}^{-1} \text{ yr}^{-1}$) (132), and higher N supply in adjacent plots greatly increased plant growth, therefore N-class=L.
34. The nutrient treatment simulated high N deposition and organic matter mineralization rates ($400 \text{ kg N ha}^{-1} \text{ yr}^{-1}$).
35. They used a 1:1:1 mix of coarse sand, peat and field soil (from a nutrient poor forest soil). No fertilized was supplied, therefore N availability was low.
36. Sandy soils classified as “nutrient-poor” and “low soil N content” by the authors.
37. Soil type typically nutrient-rich, and very high N-fertilization.
38. Soil classified as “nutrient-poor” by the authors, and N addition increased plant growth significantly.
39. Same soil as 38 but N-fertilized. N-class2=H because even though Haploxeralfs soils are N-poor, the supply of N is high.
40. Soil reported as “low nutrient availability”, and “low N availability” (pers. comm). Serpentine grasslands at Jasper Ridge consistently respond to N and P additions, with N almost doubling growth (133). CEC=0.7 mmhos cm^{-1} , SOM=7.5%.
41. This sandstone-derived soil had lower CEC (0.1 mmhos cm^{-1}), N content and SOM (5.2%) than soil 40.
42. Same soil as 40.
43. Same soil as 40 and 42, but highly fertilized.
44. Authors reported this soil was characterised by “low organic matter content” and “low nitrogen availability” (pers. comm), as also observed by the increase in growth upon fertilization.
45. Same as 44 but fertilized with N and other nutrients.
46. Sandy soils with nutrient content. Reported “infertile sandy soils”.
47. Calcareous soil with very high C:N ratio. Authors reported “low N concentration”.
48. Sheep create N-rich urine patches with larger CO₂ response, which indicates that the site is N-limited in general (pers. comm). Classified as “N-limited” (134). N-class2=M because C:N ratio is moderate, and sheep excrete and N₂-fixing species may increase N-availability.
49. Not included in the meta-analysis due to the lack of available soil information.
50. “Low in available P and estimated annual N availability of $50 \mu\text{g g}^{-1}$ ”. N-class2=M because it was not possible to assign N availability with certainty based on available information.

51. Soil type and low C:N indicate intermediate N-availability, but given the lack of fertilization we classified this soil as L-M.
52. Plant productivity is N-limited at this site (6), N-class=L. Moderately fertile soil type, low C:N ratio and evidence for nitrogen fixation (135, 136), therefore N-class2=M.
53. The high pH suggests low availability of P and some other nutrients. Reported as “nutrient-poor”, and N-availability limits plant growth.
54. Same soil as in 25, except fertilizer was not used. Nevertheless, these soils were “nutrient-rich” given past agricultural use and soil type. N-fertilization did not enhance plant growth, indicating high N-availability.
55. Even though soil organic matter content was low, we classified these soils as high due to fertilization with N and other nutrients.
56. Soil type is fertile with low C:N ratio, and was also N-fertilized. Reported as “nutrient rich”.
57. N-fertilization enhanced plant growth in the experiment, suggesting N-limitations, therefore we classified the soil as L-M.
58. Same soil as 57 but heavily fertilized with N.
59. N fertilization was kept modest so trees would not become totally deficient of it, but plants were N-limited (pers. comm).
60. Very nutrient-poor soils, in situ, very old, late successional system (pers. comm).
61. Same soil as in 60, amended with NPK.
62. Soil type characterized by high fertility. However, the authors reported that the “reduced availability of N constantly limited the response of harvestable biomass to elevated CO₂ throughout the experiment”. These plots were fertilized with 15 g N m⁻², and yet, fertilization with 45 g N m⁻² in adjacent plots produced more yield (137), suggesting that 15 g N m⁻² fertilization is in the range of N-limitations (138), classifying plants in these plots as moderately N-limited (pers. comm).
63. Same soil as in 62 with high levels of N-fertilization.
64. “Very nutrient poor despite high rates of N deposition” (pers. comm), with P probably at least as limiting as N.
65. Many Vertisols are N-deficient, in line with low SOM, and have low available P (http://www.fao.org/docrep/003/y1899e/y1899e06.htm#P381_59788). Authors reported “low total N and extractable P”.
66. Sandy soils, low in organic matter content and %N. N Mineralization = 45 µg N g⁻¹ day⁻¹. Authors reported “low soil N” and “P not limiting”.
67. N Mineralization = 348 µg N g⁻¹ day⁻¹. Authors reported “high soil N”. Since plants were well watered and P was not limiting, the major difference between soils 67 and 66 was N content, therefore, we classified it as H.
68. Nutrient-poor sandy soil, despite modest N-fertilization.
69. Equivalent to soil 66. N Mineralization = 89 µg N g⁻¹ day⁻¹. Plants received an initial dose of N-fertilizer, and for that reason N-class2=M.
70. Equivalent to soil 67. N Mineralization = 333 µg N g⁻¹ day⁻¹.

71. Similar to soils 66 and 69, and authors reported “nutrient-poor” and “low soil N”. However, *Alnus* spp. is a N₂-fixing species, therefore N-class2=M.
72. Avondale are very fertile soils used for growing cultivated crops and pasture under irrigation. Ample nutrients were added.
73. The low N treatment consisted of unamended soil which had a total N concentration of approximately 900 $\mu\text{g g}^{-1}$, that we assume as low to moderate, therefore N-class2=M. N-fertilization in adjacent plots increased growth, therefore plants were N-limited and N-class=L.
74. “Intermediate soil N fertility treatment” was imposed by supplying soil 73 with sufficient $(\text{NH}_4)_2\text{SO}_4$ to increase total soil N by 100 $\mu\text{g g}^{-1}$ N. Higher levels of N-fertilization in soil 75 did not significantly increase growth, suggesting plants in this soils were not N-limited, therefore N-class2=H.
75. “High soil N fertility treatment” was imposed by supplying soil 73 with sufficient $(\text{NH}_4)_2\text{SO}_4$ to increase total soil N by 200 $\mu\text{g g}^{-1}$ N.
76. Typic Hapludand soils are usually moderately fertile, and pH is good, therefore N-class2=M. Authors reported that the soil was “nutrient-poor”, with “soil N concentration lower than optimum for highly productive Douglas-fir forest in Oregon”, hence N-class=L.

Table S3

Experiments with arbuscular mycorrhizal plant species under low N-availability grown with N-fixing species. es: effect size, var: varianze, W_{NY} : weights used for the meta-analysis, based on the number of replicates and the duration (years) of the experiment. The data used for the analysis is aboveground biomass for all the species sampled (i.e. non N-fixing and N-fixing species).

Site	Species	Data source	es	var	W_{NY}
GiFACE	grassland	66	0.0198	0.0029	9
New Zealand FACE	temperate pasture	82	0.1165	0.0305	7.5
ORNL FACE	Liquidambar styraciflua	6	0.0146	0.0009	6.7
Swiss Central Alps	Alpine grassland	97	0.0331	0.0228	7.5
Tas FACE	Temperate grassland	14	0.0664	0.0002	7.5
Swiss Jura	Calcareous grassland	100	0.1870	0.0158	7
BioCON	perennial grassland	5	0.0892	0.0013	9.5

GiFACE: legumes (mainly *Lathyrus pratensis*) contribute less than 0.5% to the total plant biomass (116); New Zealand FACE: mixture of plant species including legumes, principally *Trifolium repens* L. And *Trifolium subterraneum* L. (139); ORNL FACE: evidence for nitrogen fixation, and an increasing presence of *Elaeagnus umbellata* (an invasive actinorhizal N fixing shrub) (135, 136); Swiss Central Alps: *Trifolium alpinum* L. is the only legume species and comprises less than 2% of the total phanerogam biomass; Tas FACE: N fixing forbs, including *Trifolium subterraneum* and *T. striatum*, form an extremely small fraction (0.01%) of the biomass. The community also contains the N-fixing woody twining species *Bossiaea prostrata*, that forms only a small fraction of the total biomass (1%) (126); Swiss Jura: data pooled across all species.

Table S4

Meta-analysis output with three different correlation factors (r) to aggregate repeated measurements over time. %es=effect size (%), se=standard error, Myc=mycorrhizal status, N=nitrogen availability.

Biomass	Myc	N	$r=1$			$r=0.5$			$r=0$		
			%es	se	P-value	%es	se	P-value	%es	se	P-value
Total	AM	High	19.71	5.92	0.002	19.71	5.72	0.001	19.71	5.55	0.001
		Low	0.35	5.29	0.946	0.35	5.13	0.945	0.35	4.98	0.943
	ECM	High	33.21	4.35	0	33.21	4.34	0	33.21	4.34	0
		Low	27.98	4.64	0	27.98	4.65	0	27.98	4.68	0
Aboveground	AM	High	18.36	4.44	0	18.04	4.49	0	18.36	4.49	0
		Low	2.3	4.36	0.595	3.55	5.59	0.523	2.3	4.45	0.425
	ECM	High	31.09	4.03	0	31.14	3.75	0	31.09	3.5	0
		Low	30.16	4.71	0	29.84	4.73	0	30.16	4.72	0
Belowground	AM	High	16.49	10.29	0.123	16.49	10.11	0.117	16.49	9.95	0.111
		Low	-0.92	8.25	0.907	-0.92	8.11	0.906	-0.92	7.99	0.905
	ECM	High	35.39	7.01	0	35.39	6.74	0	35.39	6.47	0
		Low	20.38	6.36	0.003	20.38	6.3	0.003	20.38	6.27	0.003

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