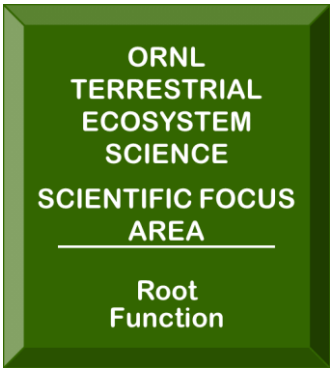
February 18, 2022

Belowground respiration, root traits, and soil characteristics of an East Tennessee deciduous forest, 2019-2020



Summary

This dataset contains empirical physiological, morphological, and chemical data of root systems, and elemental, nutrient content for soils collected on forty individuals of eight temperate tree species, between June 2019 and July 2020 at The University of Tennessee Forest Research Center and Arboretum in Oak Ridge, Tennessee. The project used a novel methodology to empirically derive estimates of the autotrophic and heterotrophic components of soil respiration *in-situ*. The project consists of two measurement approaches. The first set of measurements uses a standard approach for measuring specific root respiration on excised root systems. The second used "*in-situ* root trays"

This dataset includes 10 data files in comma separated (*.csv) ASCII format. Data include measurements of leaf and root functional traits for excised root systems and for living root systems housed within *in-situ* root trays, data on soil carbon and nitrogen pools, *in-situ* measurements of soil moisture and temperature, data on soil respiration rates for *in-situ* root trays (both as soil mass-based fluxes, and soil-area based fluxes), and data on the geographic coordinates and tree sizes of study trees. Forty study trees of eight temperate tree species were studied (five individuals per species). Two *in-situ* root trays were installed per species, each housing one entire root system comprising <3 root orders, and still being attached to the tree via transportive root.

All respiration measurements were conducted with the Li-6800 portable photosynthesis system (Li-COR, Lincoln, NE, USA). Root respiration measurements of excised root tissues were made using the Li-6800 and the Walz 3010-GWK1 gas exchange chamber (Heinz Walz GmbH, Effeltrich, Germany). Root scan images were analyzed using WinRHIZO. These images are companion files to this dataset and are contained in two compressed (*.zip) folders.

Related Publication:

The measurements and results of this study have been described in the following publication:

Hogan J.A., J.L. Labbé, A.A. Carell, J. Franklin, K.P. Hoyt, O.J. Valverde-Barrantes, C. Baraloto & J.M. Warren (*in review*). Functional variability in specific root respiration translates to slight differences in belowground CO_2 efflux in a temperate deciduous forest. *Geoderma*.

Figure 1. Root gas exchange measurements of *in-situ* root trays were conducted using the Li-6800 portable photosynthesis system. *Left:* the entire root system of a Blackgum (*Nyssa sylvatica*, Nyssaceae – root tray #29) at root tray installation (2019-06-24 – 2019-06-27). *Middle*: Li-6800 attached to the custom respiration chamber, with a root tray inside it, which was used to repeatedly measure root system and soil respiration for the root tray system over time. Measurements were taken every two-weeks to one-month for approximately one year. *Right:* Scanned root system after root tray removal.

Data Citation:

Cite this data set as follows:

Hogan, J.A., J.L. Labbé, A.A. Carell, J. Franklin, K.P. Hoyt, O.J. Valverde-Barrantes, C. Baraloto & J.M. Warren. 2022. Belowground respiration, root traits, and soil characteristics of an East Tennessee deciduous forest, 2019-2020. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/ornlsfa.025/1838660.

Acknowledgments:

Data were collected by J. Aaron Hogan (DOE SCGSR fellow from April 2019 to May 2020, and ORISE ASTRO fellow from June to September 2020 at Oak Ridge National Laboratory, Environmental Sciences Division) with the supervision of Dr. Jeffrey Warren. Direct correspondence to: jamesaaronhogan@gmail.com

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Data and Documentation Access:

Get Data

For public access to data from the US Department of Energy Terrestrial Ecosystem Science Scientific Focus Ares (TES-SFA) please visit: https://tes-sfa.ornl.gov/node/80

Description and Links to Additional Information

All gas-exchange measurements were performed with the Li-Cor Li-6800 (Li-Cor Inc., Lincoln, NE USA). *In-situ* measurements were done using a custom chamber, where as excised measurements used a Walz chamber attachment (Heinz Walz GmbH, Eiffeltrich, Germany).

- Li-Cor documentation on custom chamber builds for the Li-6800: https://www.licor.com/env/support/LI-6800/topics/chamber-custom-note.html
- Li-Cor Li-6800 owner's manual: https://www.licor.com/env/support/LI-6800/manuals.html
- Walz 3010-WGK1 Chamber: https://www.walz.com/products/gas_exchange/3010-gwk1/introduction.html

Related Data Sets:

Sequencing data for soil bacteria and fungi have been archived in an sequence read archive in GenBank -- BioProject SRA # PRJNA786934 http://ncbi.nlm.nih.gov/bioproject/PRJNA786934

Table of Contents:

1. Data Set Overview:

Eighty *in-situ* root trays (a novel method for measuring root and soil respiration) were installed on 40 study trees of eight temperate tree species (Tulip Poplar, Redbud, Sourwood, Blackgum, American Beech, Red Maple, Loblolly Pine and Sweetgum - see Section 5: Data Acquisition Materials and Methods for Latin binomial species names) and measured rates of soil respiration for approximately one year. A low-strength biocide (ZeroTol, peracetic acid, hydrogen peroxide) was used as an experimental treatment on the soils on half of the root trays in a paired design. Specific root respiration rates of excised root systems were measured at three different times during the spring/summer of 2020. Soils from the *in-situ* root trays were analyzed for carbon and nitrogen pools using the chloroform extraction technique and sequencing for fungi (ITS) and bacteria (16S).

This dataset contains respiration measurements of roots and soil housed within the *in-situ* root trays, respiration measurements of root tissues, leaf and root functional traits, and soil analysis data for 80 experimental replicates of eight temperate tree species and 6 (3 pairs) of soil-only controls. The experimental treatments are ZeroTol:R+Z+ (ZeroTol treated *in-situ* root trays) Control:R+ Z- (Control, non-ZeroTol *in-situ* root trays), Soil_ZeroTol:R-Z+ (ZeroTol treated soil-only control trays, not containing roots), Soil_Control:R-Z+ (Control, non-ZeroTol soil-only control trays, not containing roots).

2. Data Characteristics:

This dataset includes 10 data files in comma separated (*.csv) ASCII format. See Data File Descriptions for information on the contents of individual files. Additionally, there are two sets of companion files containing images analyzed using WinRHIZO, see Companion Files for additional details.

Temporal Coverage:
The experiment began with the installation of the *in-situ* root trays on June 24 to June 26, 2019. The experiment ran for 11 months and the root trays were collected from the field on May 28, 2020. Measurements of soil CO_2 efflux from the root trays were done on the following dates:

Measurement	Dates
1	2019-07-04 – 2019-07-06
2	2019-07-18,21,22
3	2019-08-01 – 2019-08-03
4	2019-08-16 – 2019-08-17
5	2019-08-30, 2019-09-01,02
6	2019-09-13 – 2019-09-15
7	2019-09-27 – 2019-09-29
8	2019-10-12 – 2019-10-14
9	2019-10-25 – 2019-10-28
10	2019-11-29 – 2019-12-02
11	2019-12-29 – 2020-01-01
12	2020-02-02 – 2019-02-03
13	2020-03-07 – 2019-03-09
14	2020-03-27 – 2019-03-29
15	2020-04-09 – 2019-04-11
16	2020-04-27 – 2019-04-28
17	2020-05-09 – 2019-05-11
18	2020-05-26 – 2019-05-28

Measurements of specific root respiration rates from excised root systems were done on the following dates:

Measurement	Dates
1	2020-03-25
2	2020-05-20
3	2020-07-24

Temporal Resolution:
Measurements were taken every two weeks on average, except during the winter months (November – February), when measurements were then taken once a month.

Spatial Coverage:
This field experiment was conducted at University of Tennessee Forest Resources AgReserach and Education Center & Arboretum (UTArb): 901 South Illinois Avenue Oak Ridge, TN 37830 (36.002222 N, -84.214167 W).

Data File Descriptions:

File Name	Description
UTArb_LeafTrait Data.csv	Leaf functional trait data for the 8 study species. One leaf from each of the five study trees per species was collected on 2019-08-29. Leaves were measured, scanned, dried, weighed, then homogenized and tissues were analysed elementally. Dataset contains data for 16 leaf functional traits.

File Name	Description
UTArb_ExcisedRootTraitData.csv	Entire root system functional trait data for excised root systems, where specific root respiration rates were measured. Dataset contains 15 root functional traits, including rates of root tissue respiration (in per unit area and per unit mass basis).
UTArb_RootTray_RootSystemTraitData.csv	Entire root system functional trait data for root systems housed within the *in-situ* root trays. Dataset contains measurements of 12 root functional traits.
UTArb_SoilCarbon_and_NitrogenPools.csv	Data from soil chloroform extractions for soils from the *in-situ* root trays. Data are from 86 trays, including 6 control trays lacking tree roots. Measurements of fumigated and unfumigated carbon and nitrogen pool concentrations are given.
UTArb_SoilMoisture_and_TempData.csv	Soil moisture and temperature data were collected concurrently with soil CO_2 efflux measurement for each *in-situ* root tray (n = 1548).
UTArb_Tissue RespirationMaster.csv	Entire root systems (containing 4 orders of physiologically active root tissue) were field-collected, washed, and measured using gas exchange equipment for root respiration rates. Root systems were collected in the spring/summer of 2020 at roughly two-month intervals, five root systems per species (8 species total), and measured. Collection and measurement were completed on the same day (within several hours of collection at the latest). Harvest dates were: 1) 2020-03-25 (Measurement 1), 2) 2020-05-20 (Measurement 2), 3) 2020-07-24 (Measurement 3)
UTArb_TrayCoordinates.csv	Global position system (GPS) coordinates in decimal degree format for each of the 40 trees in the study, and for where soil-only controls were placed in the forest.
UTArb_TrayRespiration_MasterDataset_bySoilMass.csv	Soil and root system CO_2 efflux data over time for the *in-situ* root respiration trays (including soil-only controls). Fluxes are calculated by soil mass, therefore S – the surface area constant, used in the denominator for determining CO_2 efflux rates – is the total dry mass of soil in each root tray (as measured at the end of the experiment).
UTArb_TrayRespiration_MasterDataset.csv	Soil and root system CO_2 efflux data over time for the *in-situ* root respiration trays (including soil-only controls). Fluxes are calculated by tray area, therefore S – the surface area constant, used in the denominator for determining CO_2 efflux rates – is the total surface area of the root tray (486.92 cm^2).
UTArb_TreeSizeData.csv	Tree diameters at breast height (dbh) and heights as measured at root tray installation (dbh1, 2019-06-24 to 2019-06-27), and following root tray retrieval (dbh2, 2020-05-30).
UTArb_ExcisedRootSystemScans.zip	Contains images used for WinRHIZO analysis for excised root system trait data (UTArb_RootTraitData.csv). Contains 81 *.jpg images (80 root system scans and 1 ruler scan for calibration).

File Name	Description
UTArb_TrayRootSystem Scans.zip	Contains images used for WinRHIZO analysis for In-situ root tray root system trait data (UTArb_RootTrays_RootSystemTraitData.csv). Contains 121 *.jpg images (120 root system scans and 1 rule scan for calibration).

Root functional trait data are given in separate data files from root respiration measurements.

- Gas exchange measurements were always taken with the Li-6800 portable synthesis system (Li-COR Inc., Lincoln, NE, USA) and are grouped by measurement technique (i.e., there is one dataset for the gas exchange measurements of the *in-situ* root trays and a separate dataset for the specific root respiration rates of excised root systems).
- Leaf and root functional traits (i.e., specific leaf area and tissue and carbon and nitrogen concentrations) are given by species for leaves and by root systems for root datasets where either rates of *in-situ* root tray CO_2 efflux or specific root respiration rates were measured.

Please refer to Section 5, Data Acquisition Materials and Methods, for details of the measurement methods for data reported in each data file.

These data are considered at Quality Level 1. Level 1 indicates an internally consistent data product that has been subject to quality checks and data management procedures.

Data Dictionaries:

Leaf trait data: UTArb_LeafTraitData.csv

Column Number	Column Name	Units/format	Description
1	Species	text	Eight-level factor for tree species from which leaves were collected.
2	Leaf	text	Five level-factor (A-E) for leaf replicate.
3	LeafThickness	mm	Leaf thickness in mm – measured with a vernier caliper precise to the thousandth of a mm.
4	FreshMass	g	Leaf fresh mass in grams – massed immediately after collection from the tree.
5	DryMass	g	Leaf dry mass in grams – massed after drying to constant weight – several days at 70°C.
6	SLA	$m^2\ kg^{-1}$	Specific leaf area – the ratio of leaf area to its dry mass – in square meters per kilogram.

7	SLWC	%	Specific leaf water content in percent, calculated as: [(FreshMass – DryMass) / Dry Mass] * 100.
8	LeafArea	cm^2	Leaf surface area in square centimeters – measurement derived from scanned leaves using ImageJ.
9	Perimeter	cm	Leaf perimeter in cm - measurement derived from scanned leaves using ImageJ.
10	HorizontalWidth	cm	Leaf horizontal width in cm - measured using imageJ.
11	VerticalHeight	cm	Leaf length in cm - measured perpendicular to width using imageJ
12	Circularity	unitless	Leaf circularity, influenced by leaf serration/lobing - ranges from 0 (infinitely narrow) to 1 (perfect circle).
13	AspectRatio	unitless	The ratio of the leaf length to width – ranges from 0 (circular) to no upper bound (infinitely narrow).
14	Roundness	unitless	Leaf roundness, influence by leaf length and width - ranges from 0 (infinitely narrow) to 1 (perfect circle).
15	LeafN	%	Leaf tissue percent Nitrogen – measured using GC-MS elemental analysis.
16	LeafC	%	Leaf tissue percent Carbon – measured using GC-MS elemental analysis.
17	LeafP	%	Leaf tissue percent Phosphorus – measured using acid digestion and Lachat QuikChem system.

Excised root system trait data: UTArb_ExcisedRootTraitData.csv

Column Number	Column Name	Units/format	Description
1	Sample	integer	Continuous variable (1-120) for each individual excised root system.
2	Date	YYYY-MM-DD	Date on which the root system was dug up and measured in YYYY-MM-DD format
3	Collection	text	Collection number. Three-level factor for measurement replicate.

4	Species	text	Eight-level factor for tree species from which root systems were collected.
5	Replicate	integer	Five-level factor (1-5) for the root system replicate.
6	Image	text	Filename (jpeg) of the scanned root system which WinRHIZO analyzed. Scanned images of root systems were used to estimate traits using WinRHIZO. Images be found in the UTArb_ExcisedRootSystemScans.zip companion file.
7	Length	cm	Total linear length (cm) of root system.
8	SRL	cm g^{-1}	Specific root length. The ratio of the total linear length of the root system to the root system dry mass.
9	SurfArea	cm^2	Project root system surface area in square centimeters.
10	SRA	cm^2 g^{-1}	Specific root area, or the ratio of the projected root surface area of the root system to its dry mass in square centimeters per gram.
11	AvgDiam	mm	Root system average diameter in mm.
12	LenPerVol	cm cm^{-3}	Root length to volume ratio in centimetres per cubic centimeter.
13	Vol	cm^{-3}	Root volume in cubic centimeters. Estimated from root diameter and length using the equation for the volume of a cylinder (See documentation for WinRHIZO).
14	SRTA	tips cm^{-1}	Specific root tip abundance. The ratio of number of root tips (terminal ends of the root system to its total linear length.
15	Tips	integer	Number of root tips (terminal ends).
16	RTD	g cm^{-3}	Root tissue density, or the ratio of the root system dry mass to its total volume in grams per cubic centimeter.
17	RootDryMass	g	Root system dry mass in grams.
18	R_r_umols_m2	µmols m^2	Root respiration rate (area-based) in micromoles per square meter.
19	R_r_umols_kg	µmols kg^{-1}	Specific root respiration rate (mass-based) in micromoles per kilogram.
20	RootN	%	Root tissue percent nitrogen (from elemental analysis of homogenized tissue).

21	RootC	%	Root tissue percent carbon (from elemental analysis of homogenized tissue).

***In-situ* root tray root system trait data: UTArb_RootTrays_RootSystemTraitData.csv**

Note: Missing data are indicated by NA.

Column Number	Column Name	Units/format	Description
1	Tray	text	Unique identifier for root tray. Identifiers that include "Z" denote ZeroTol treatment.
2	Species	text	Eight-level factor for tree species from which root systems were collected.
3	Treatment	text	Four-level factor for the treatment of the root tray. R+ corresponds to trays with root systems, whereas R- denotes soil-only control trays (i.e., those lacking roots). Z+ is for trays treated with the ZeroTol solution, where Z- shows trays sprayed with water (control treatment).
4	Image	text	Filename (jpeg) of the scanned root system which WinRHIZO analyzed. Scanned images of root systems were used to estimate traits using WinRHIZO. Images be found in the UTArb_TrayRootSystemScans.zip companion file.
5	Length	cm	Total linear length (cm) of root system
6	SRL	$cm\ g^{-1}$	Specific root length. The ratio of the total linear length of the root system to the root system dry mass.
7	SurfArea	cm^2	Project root system surface area in square centimeters.
8	SRA	$cm^2\ g^{-1}$	Specific root area, or the ratio of the projected root surface area of the root system to its dry mass in square centimeters per gram.
9	AvgDiam	mm	Root system average diameter in mm.
10	LenPerVol	$cm\ cm^{-3}$	Root length to volume ratio in centimetres per cubic centimeter.
11	Vol	cm^{-3}	Root volume in cubic centimeters. Estimated from root diameter and length using the equation for the volume of a cylinder (See documentation for WinRHIZO).
12	SRTA	$tips\ cm^{-1}$	Specific root tip abundance. The ratio of number of root tips (terminal ends of the root system to its total linear length
13	Tips	integer	Number of root tips (terminal ends)

14	RTD	g cm^{-3}	Root tissue density, or the ratio of the root system dry mass to its total volume in grams per cubic centimeter
15	RootDryMass	g cm^{-3}	Root tissue density, or the ratio of the root system dry mass to its total volume in grams per cubic centimeter.
16	TotalSoilMass	g	Root system dry mass in grams.

Soil Chloroform extraction data: UTArb_RootTray_SoilCarbon&NitrogenPools.csv

Column Number	Column Name	Units/format	Description
1	SampleID	integer	Unique identifier for root tray. Identities that include "Z" denote ZeroTol treatment. Identities that include "C" denote soil-only control trays.
2	Species	text	Eight-level factor for tree species for which the root tray was installed.
3	Treatment	text	Four-level factor for the treatment of the root tray. R+ corresponds to trays with root systems, whereas R- denotes soil-only control trays (i.e., those lacking roots). Z+ is for trays treated with the ZeroTol solution, where Z- shows trays sprayed with water (control treatment).
4	UnFumigated_SoilWt	g	The mass of fresh soil in the sample before chloroform extraction (i.e. fumigation)
5	Fumigated_SoilWt	g	The mass of fresh soil in the sample after chloroform extraction (i.e. fumigation)
6	GWG	% g/g dry soil	Soil Gravimetric water content (note that soils were dried and sieved prior to chloroform extraction)
7	UnFumigated_DrySoilWt	g	The mass of dry soil in the sample before chloroform extraction (i.e. fumigation)
8	Fumigated_DrySoilWt	g	The mass of dry soil in the sample after chloroform extraction (i.e. fumigation)
9	K2SO4_Extraction Vol	L	The amount of potassium sulfate used during the extraction
10	TOC_UnFumigated	ppm-- mg L-1	Soil total organic carbon in the unfumigated sample
11	TOC_Fumigated	(ppm-- mg/L)	Soil total organic carbon in the fumigated sample
12	DOC_UnFumigated	mgC/g soil	Soil dissolved organic carbon in the unfumigated soil sample

Column Number	Column Name	Units/format	Description
13	DOC_Fumigated	mgC/g soil	Soil dissolved organic carbon in the fumigated sample
14	MBC_milli	mgC/g soil	Soil microbial carbon (in mg C) – calculated as the difference between the fumigated and unfumigated sample for each replicated
15	DOC	ugC/g soil	Soil dissolved organic carbon – calculated as the difference between the fumigated and unfumigated sample for each replicated
16	MBC_micro	ugC/g soil	Soil microbial carbon (in µg C) – calculated as the difference between the fumigated and unfumigated sample for each replicated
17	TN_UnFumigated	ppm-- mg/L	Soil total nitrogen in the unfumigated sample
18	TN_Fumigated	ppm-- mg/L	Soil total nitrogen in the fumigated sample
19	DN_Fumigated	mgN/g soil	Soil dissolved nitrogen in the unfumigated sample
20	DN_UnFumigated	mgN/g soil	Soil dissolved nitrogen in the fumigated sample
21	MBN_milli	mgN/g soil	Soil microbial nitrogen (in mg N) – calculated as the difference between the fumigated and unfumigated sample for each replicated
22	DN	ugN/g soil	Soil dissolved nitrogen – calculated as the difference between the fumigated and unfumigated sample for each replicated
23	MBN_micro	ugN/g soil	Soil microbial nitrogen (in µg N) – calculated as the difference between the fumigated and unfumigated sample for each replicated

***In-situ* root tray soil moisture and temperature data:**
UTArb_SoilMoisture_and_TempData.csv

Column Number	Column Name	Units/format	Description
1	Date	YYYY-MM-DD	Date on which the *in-situ* root tray was measured in YYYY-MM-DD format
2	Measurement	text	Unique identifier for measurement replicate (range 1-18)
3	Treatment	text	Four-level factor for the treatment of the root tray. R+ corresponds to trays with root systems, whereas R- denotes soil-only control trays (i.e., those lacking roots). Z+ is for trays treated with the ZeroTol

Column Number	Column Name	Units/format	Description
			solution, where Z- shows trays sprayed with water (control treatment).
4	Tray	text	Unique identifier for root tray. Identities that include "Z" denote ZeroTol treatment.
5	Species	text	Eight-level factor for tree species for which the root tray was installed
6	MycorrTypc	text	Two level factor for mycorrhizal type, AM = arbuscular mycorrhizal, ECM = ectomycorrhizal
7	SoilMoisture	%	Volumetric soil water content (i.e., moisture) in the surface soil. Measurement was taken using a ΔT Devices, SM-150 soil moisture probe. Three measurements were taken on each of the three sides (i.e. cardinal directions) of the root tray opposite the entry point of the root system. Measurements were averaged.
8	SoilTemp	°C	Soil temperature, taken using a soil thermometer (Fischer Scientific, USA). Three measurements were taken on each of the three sides (i.e. cardinal directions) of the root tray opposite the entry point of root system. Measurements were averaged.

Root system specific respiration data: UTArb_TissueRespirationMaster.csv

Column Number	Column Name	Units/format	Description
1	Replicate	integer	Root system collection number (range 1-3).
2	Date	yyyy-mm-dd	The date on which the root system respiration measurement was taken
2	obs	integer	Li-6800 observation number, consecutive since the opening of the data file
3	time_elapsed	s	The time (in seconds) elapsed since the Li-6800 was turned on and new data logging file was opened
4	time	hh:mm:ss	The time of day (military time)
5	Species	text	Eight-level factor for tree species for which the root tray was installed
6	Root	integer	Root system replicate for the collected tree individual (range 1-5 per tree)
7	R_r	$\mu mol\ m^{-2}\ s^{-1}$	The respiration rate of the root system (area based). Flux is calculated using S-

Column Number	Column Name	Units/format	Description
			the surface area constant (or area of the respiration chamber)
8	R_r_g	µmol kg^{-1} s^{-1}	The specific root system respiration (mass-based). Flux is calculated using the root system dry mass (in g)
9	Ca	µmol mol^{-1}	Concentration of extracellular CO_2 (i.e., ambient CO_2 concentration of the chamber)
10	Pca	Pa	Pressure of extracellular CO_2 (i.e., in the sample chamber)
11	RHCham	%	Relative humidity of the air in the infra-red gas analyzer (IRGA). calculated as the difference between sample and reference IRGAs
12	S	cm^2	Surface area constant – the area of the WALZ respiration chamber (232 cm^2, dimensions: 16cm × 14.5cm)
13	Root_DryMass	g	The dry mass of the root system (in grams) for which the respiration rate was measured
14	CO2_s	µmol mol^{-1}	Sample IRGA CO_2 concentration
15	CO2_r	µmol mol^{-1}	Reference IRGA CO_2 concentration
16	H2O_s	mmol mol^{-1}	Sample IRGA water concentration
17	H2O_r	mmol mol^{-1}	Reference IRGA water concentration
18	Flow	µmol s^{-1}	Flow rate of air to chamber
19	Pa	kPa	Atmospheric pressure of air in chamber
20	dPCham	kPa	Chamber overpressure
21	Tair	°C	Temperature of air flowing through chamber
22	Fan_speed	rpm	Instrument mixing fan rotation rate
23	Match_time	hh:mm:ss	Time of day for which the last match (match used for calculating fluxes, i.e., ΔIRGA concentrations) was done
24	Match_count	integer	Match number of last match
25	Match_co2_adj	µmol mol^{-1}	The adjustment of the CO_2 concentration measurement between IRGAs at the last match
26	Match_h2o_adj	mmol mol^{-1}	The adjustment of the water concentration measurement between IRGAs at the last match
27	dCO2_Flag	binary	Stability criteria binary variable. Is the CO_2 concentration stable given criteria? 1 indicates yes, 0 indicates no.
28	dH2O_Flag	binary	Stability criteria binary variable. Is the water concentration stable given criteria? 1 indicates yes, 0 indicates no.

Column Number	Column Name	Units/format	Description
29	Stability_State	interger	Integer (of 2) for stability criteria met at time of data point logging

Root system geographic coordinate data: UTArb_TrayCoordinates.csv

Column Number	Column Name	Units/format	Description
1	Tree	integer	Tree identifier (corresponds to paired *in-situ* tray replicate)
2	Species	text	Eight-level factor for the species of tree
3	lat	decimal degrees	Degrees latitude
4	lon	decimal degrees	Degrees longitude

***In-situ* root tray respiration data: soil- and root-mass based fluxes: UTArb_TrayRespiration_MasterDataset_bySoilMass.csv**

Column Number	Column Name	Units/format	Description
1	Measurement	integer	Unique identifier for measurement replicate (range 1-18)
2	Date	yyyy-mm-dd	Date on which the *in-situ* root tray was measured
3	obs	integer	Li-6800 observation number, consecutive since the opening of the data file
4	time_elapsed	s	The time (in seconds) elapsed since the Li-6800 was turned on and new data logging file was opened
5	time	hh:mm:ss	The time of day (military time)
6	Treatment	text	Four-level factor for the treatment of the root tray. R+ corresponds to trays with root systems, whereas R- denotes soil-only control trays (i.e., those lacking roots). Z+ is for trays treated with the ZeroTol solution, where Z- shows trays sprayed with water (control treatment).
7	Tray	text	Unique identifier for root tray. Intensifies that include "Z" denote ZeroTol treatment.
8	E	mol g^{-1} s^{-1}	Soil mass-based evaporation rate (corrected for leaks)
9	R	µmol kg^{-1} s^{-1}	Soil mass-based respiration rate (corrected for leaks), note values in datafile are negative, indicating CO_2 release rather than uptake (see Li-6800 manual for calculation)

Column Number	Column Name	Units/format	Description
10	RHcham	%	Relative humidity of the air in the IRGA. calculated as the difference between sample and reference IRGAs
11	VPcham	kPa	Vapor pressure in the chamber
12	SVPcham	kPa	Saturation vapor pressure in the chamber
13	Fan	µmol s^{-1}	Fan flow rate
14	S	g	Dry soil mass within the tray in g (measured at the end of the experiment). The dry soil mass is used as the surface area constant (S) for the calculation of mass-based CO_2 and water flux – see Li-6800 manual.
15	CO2_s	µmol mol^{-1}	Sample IRGA CO_2 concentration
16	CO2_r	µmol mol^{-1}	Reference IRGA CO_2 concentration
17	H2O_s	mmol mol^{-1}	Sample IRGA water concentration
18	H2O_r	mmol mol^{-1}	Reference IRGA water concentration
19	Flow	µmol s^{-1}	Flow rate of air to chamber
20	Pa	kPa	Atmospheric pressure of air in chamber
21	dPCham	kPa	Chamber overpressure
22	Tair	°C	Temperature of air flowing through chamber
23	Fan_speed	rpm	Instrument mixing fan rotation rate
24	Qamb_out	µmol m^{-2} s^{-1}	Light measurement (PAR: photosynthetically active radiation) from the external quantum sensor
25	Match_time	hh:mm:ss	Time of day for which the last match (match used for calculating fluxes, i.e., ΔIRGA concentrations) was done
26	Match_count	integer	Match number of last match
27	Match_co2_adj	µmol mol^{-1}	The adjustment of the CO_2 concentration measurement between IRGAs at the last match
28	Match_h2o_adj	mmol mol^{-1}	The adjustment of the water concentration measurement between IRGAs at the last match
29	A_Flag	binary	Stability criteria binary variable. Is the CO_2 concentration stable given criteria? 1 indicates yes, 0 indicates no
30	RHcham_Flag	binary	Stability criteria binary variable. Is the water concentration stable given criteria? 1 indicates yes, 0 indicates no
31	Stability_State	interger	Integer (out of 2) for stability criteria met at time of data point logging

***In-situ* root tray respiration data: area-based fluxes:**
UTArb_TrayRespiration_MasterDataset.csv

Column Number	Column Name	Units/format	Description
1	Measurement	integer	Unique identifier for measurement replicate (range 1-18)
2	Date	YYYY-MM-DD	Date on which the *in-situ* root tray was measured in YYYY-MM-DD format
3	obs	integer	Li-6800 observation number, consecutive since the opening of the data file
4	time_elapsed	s	The time (in seconds) elapsed since the Li-6800 was turned on and new data logging file was opened
5	time	hh:mm:ss	The time of day (military time)
6	Treatment	text	Four-level factor for the treatment of the root tray. R+ corresponds to trays with root systems, whereas R- denotes soil-only control trays (i.e., those lacking roots). Z+ is for trays treated with the ZeroTol solution, where Z- shows trays sprayed with water (control treatment).
7	Tray	text	Unique identifier for root tray. Identifies that include "Z" denote ZeroTol treatment.
8	E	mol m^{-2} s^{-1}	Evaporation rate (corrected for leaks)
9	R	µmol m^{-2} s^{-1}	Respiration rate (corrected for leaks), note values in datafile are negative, indicating CO_2 release rather than uptake (see Li-6800 manual for calculation)
10	RHcham	%	Relative humidity of the air in the IRGA. calculated as the difference between sample and reference IRGAs
11	VPcham	kPa	Vapor pressure in the chamber
12	SVPcham	kPa	Saturation vapor pressure in the chamber
13	Fan	µmol s^{-1}	Fan flow rate
14	S	cm^2	Surface area constant – the average area of the *in-situ* root tray, 486.92 cm^2. The average tray dimensions were 8.8675 in × 8.8675 in, or 22.067 cm × 22.067 cm.
15	CO2_s	µmol mol^{-1}	Sample IRGA CO_2 concentration
16	CO2_r	µmol mol^{-1}	Reference IRGA CO_2 concentration
17	H2O_s	mmol mol^{-1}	Sample IRGA water concentration
18	H2O_r	mmol mol^{-1}	Reference IRGA water concentration
19	Flow	µmol s^{-1}	Flow rate of air to chamber
20	Pa	kPa	Atmospheric pressure of air in chamber
21	dPCham	kPa	Chamber overpressure

Column Number	Column Name	Units/format	Description
22	Tair	°C	Temperature of air flowing through chamber
23	Fan_speed	rpm	Instrument mixing fan rotation rate
24	Qamb_out	µmol m^{-2} s^{-1}	Light measurement (PAR: photosynthetically active radiation) from the external quantum sensor
25	Match_time	hh:mm:ss	Time of day for which the last match (match used for calculating fluxes, i.e., ΔIRGA concentrations) was done
26	Match_count	integer	Match number of last match
27	Match_co2_adj	µmol mol^{-1}	The adjustment of the CO_2 concentration measurement between IRGAs at the last match
28	Match_h2o_adj	mmol mol^{-1}	The adjustment of the water concentration measurement between IRGAs at the last match
29	A_Flag	binary	Stability criteria binary variable. Is the CO_2 concentration stable given criteria?
30	RHcham_Flag	binary	Stability criteria binary variable. Is the water concentration stable given criteria
31	Stability_State	fraction	Integer (out of 2) for stability criteria met at time of data point logging

Tree size data: UTArb_TreeSizeData.csv

Column Number	Column Name	Units/format	Description
1	Tree	NA	the unique tree id (ranges from 1-40)
2	Species	NA	One of eight tree species in the study
2	DBH_June2019	cm	The diameter at breast height when root trays were installed. Measurements taken from 2019-06-24 – 2019-06-26.
3	DBH_May2020	cm	The diameter at breast height after root trays had been collected. Measurements taken 2020-05-30.
4	Height	m	Tree height at maximum. Measurements made using telescoping tree height rod on 2020-05-30.

Companion files:

This dataset contains two sets of companion files containing root scan images in *.jpg format that were used for WinRHIZO analysis. These images are stored in two compressed (*.zip) folders:

- UTArb_ExcisedRootSystemScans.zip: Contains images used for WinRHIZO analysis for excised root system trait data (UTArb_RootTraitData.csv). Contains 81 *.jpg images (80 root system scans and 1 ruler scan for calibration).
- UTArb_TrayRootSystemScans.zip: Contains images used for WinRHIZO analysis for *In-situ* root tray root system trait data (UTArb_RootTrays_RootSystemTraitData.csv). Contains 121 *.jpg images (120 root system scans and 1 rule scan for calibration).

3. Applications and Derivation:

Data can be used to partition the contribution of root respiration to total soil CO_2 efflux. Data on specific root respiration rates from excised roots systems can be used to evaluate the functional relationship between respiration rates and morphological and chemical traits of root systems. Data on the soil moisture and temperature conditions for the environmental condition of *in-situ* root trays over time can be used to model their effects on soil autotrophic (i.e., root-associated) and heterotrophic respiration rates. Data are included on soil fertility, microbial nitrogen – a proxy for microbial biomass, and the composition of bacteria (16S gene) and fungi (ITS2 gene), which can be used to evaluate the effect of the experimental treatments on the soils of the *in-situ* root trays. Lastly, data on root and leaf functional traits can be used to understand the ecological difference among species, in terms of plant life-history and economical strategy, which helps with the interpretation and greater understanding of the observed interspecific differences in root and root-associated soil respiration rates.

4. Quality Assessment

These data are considered at Quality Level 1. Level 1 indicates an internally consistent data product that has been subjected to quality checks and data management procedures. Established calibration procedures were followed.

5. Data Acquisition Materials and Methods:

Study Site:

The study took place at the University of Tennessee Forest Resources Education Center and Arboretum in Oak Ridge, Tennessee (35.9935°N, 84.2201°W). The study area was located on the Chestnut Ridge research area to the northwest of the Arboretum grounds. Forty study trees of eight species were selected to target a range of temperate tree life-history and root functional strategies.

Study Species Description:

Eight temperate tree species were studied, which are listed in the table below:

Table 1: Taxonomic information for eight temperate tree species at the University of Tennessee Arboretum research area at Oak Ridge, Tennessee selected for *in-situ* root tray installation.

Species Latin binomial with taxonomic authority	Family	Common name
Pinus taeda L.	Pinaceae	Loblolly Pine
Liriodendron tulipifera L.	Magnoliaceae	Tulip Poplar
Liquidambar styraciflua L.	Altingiaceae	Sweetgum
Cercis canadensis L.	Fabaceae	Eastern Redbud
Fagus grandifolia L.	Fagaceae	American Beech
Acer rubrum L.	Sapindaceae	Red Maple
Nyssa sylvatica Marshall	Nyssaceae	Blackgum
Oxydendrum arboretum L. (DC.)	Ericaceae	Sourwood

The Tennessee Tree ID guide (Williams, 2005) was used to ID all species except Eastern Redbud (*Cercis canadensis*), which was identified using firsthand knowledge. Jennifer Franklin, professor in the Department of Forestry, Wildlife, and Fisheries at the University of Tennessee, helped confirm all species identifications at the tree and root system-level.

Materials and Methods:

In-situ root tray design: a new method for repeated environmental measurements of root system and soil respiration

A new methodology was developed for this experiment (Figure 2). The method permitted the repeated measurement of root and soil CO_2 efflux (i.e., belowground respiration) at the functional unit of the entire fine root system (ERS, i.e., a complete root system of <2mm diameter, containing at least 3 root orders from the finest first order roots to ~4th order transportive root, Fig. 1). The method used *in-situ* root trays to house the ERS for the duration of the study. Trays measured, on average, 8.8675” × 8.8675” (22.067 cm × 22.067 cm) and were constructed from 4-gallon square buckets (part number S-13650W, ULINE, Pleasant Prairie, WI). Buckets were sliced into 2”-high square strips using a table saw; buckets tapered slightly from bottom (8 1/2” square) to top (9 7/8” square), which permitted nesting of strips inside one another. Two strips were nested within each other with a 2 mm fine aluminum wire mesh stretched over the bottom of the tray and secured between the two square strips with contractor staples. A notch was cut in the side of the tray to accommodate the placement of the root system within the tray (Figure 2).

Trays were designed to fit within a custom respiration chamber, which was constructed for use with the Li-6800 portable gas exchange system (Li-COR Inc. Lincoln, NE), following the provider’s theory and recommendation (https://www.licor.com/env/support/LI-6800/topics/chamber-custom-note.html, Figure 2). The chamber measured 12” × 12” × 4 1/4”

with an open front design and was constructed out of 1/2" acrylic sheeting by solvent bonding all pieces together. An 8mm 12V computer fan was installed to the top inside of the chamber to mix the chamber air volume. Holes were drilled in the back of the chamber to accommodate the Li-6800 custom chamber adaptor, which allowed it to interface with the gas exchange system; the chamber was secured to the gas exchange system with rubber washers and machine bolts, which minimized leaking. The front of the chamber was constructed out of the same 1/2" acrylic sheeting with a 2" by 1/2" notch to accommodate the root systems. The front of the chamber was fitted with 1" heavy-duty rubber weather stripping to seal its connection to the root box, and toggle clamps were installed to allow the chamber to be opened and closed as needed (Figure 2).

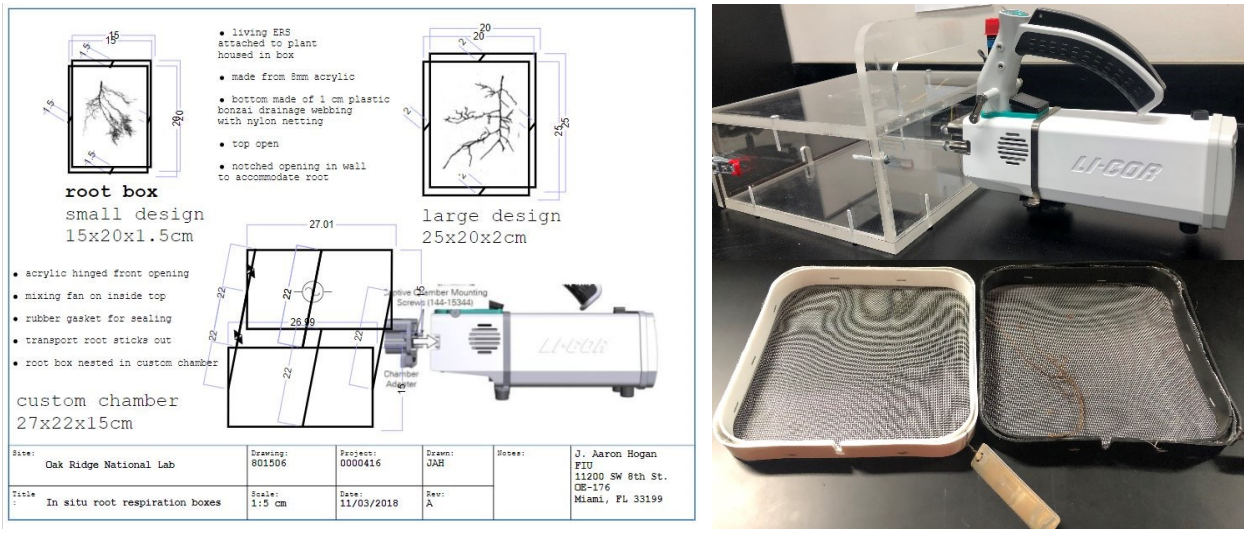

Figure 2. *Left:* Design of custom respiration chamber for attachment to Li-Cor Li-6800 portable gas exchange systems (IRGAs with control console) at it initial inception (November 2018). *Right:* Finished construction of custom respiration chamber (attached to Li-6800 measurement head) and *in-situ* root trays (January 2019).

In-situ root tray installation

Five healthy trees of each the eight study species (Table 1) were selected. The *P. taeda* and *L. styraciflua* trees were found in a planted Loblolly stand at the west end of the research area, the *L. tulipifera* and the *C. canadensis* trees were in an area at the east of the research area that had been clear cut about 15 years prior, and the remaining four study species were located in a mature Eastern deciduous oak-hickory stand between planted stand and the previously clear-cut area.

At each of the forty study trees, the *in-situ* root trays were installed on excavated ERSs. The terminal portion of the ERS, complete to the finest first-order root tips was housed within the tray, while 4th order transportive root, which was still attached to the tree, protruded from the tray at the notch. Installation took place from June 24 – June 26, 2019. ERSs were gently excavated by hand, imaged, and placed into the *in-situ* root trays using soil that was loosened during excavation. Root trays were recessed slightly into the ground, the 4th order transportive root that exited the tray and was still attached to the tree was buried, and leaf litter was replaced over the trays. Trays were promptly watered after installation. The *in-situ* root trays were

installed in a paired design on two separate ERSs per tree, with one white tray (for ZeroTol treatment) and one black tray (for the Control treatment).

Treatments & experiment duration

A treatment was applied to half of the *in-situ* trays (one per individual tree). The treatment was intended to sterilize the soil by reducing the microbial and fungal abundance in an effort to supress rates of soil heterotrophic (i.e., non-root) respiration. A low-level, broad-spectrum fungicide/ bactericide/ algaecide called ZeroTol 2.0 (Biosafe Systems, Hartford, CT) was used. Approximately 350 mL of a 1% concentrated ZeroTol solution was applied biweekly using backpack sprayers (Figure 3) for the duration of the experiment beginning on June 8, 2019 and ending on May 28, 2020.

Figure 3. *Left:* Treatments: A broad-spectrum fungicide, bactericide and algaecide (left, ZeroTol 2.0) was applied throughout the experiment to half of the *in-situ* root trays. The other half of the root trays were treated with deionized water. *Middle:* Treatments were applied using backpack sprayers (middle). Miranda Clark and Aaron Hogan treat trays at the start of the experiment (summer 2019). Treatments had a slight effect at sterilizing soils, wherein bacterial biomass (microbial Nitrogen content) was reduced and community composition of bacteria and fungi was altered slightly (see main article), however observationally, fungi opportunistically colonized ZeroTol treated trays in certain instances, especially after rainfall. *Right:* A white-fuzz fungus grows on the soil surface of recently Zerotol-treated *in-situ* root trays.

Tray removal and processing of soils

On May 28, 2020, all *in-situ* root trays were removed from the forest. Trays were allowed to air dry for 10 days in a cool dry location. Root systems were carefully removed, scanned, and weighed for the quantification of root functional traits. Soils were then sieved using a #2 10mm mesh sieve, which was sterilized with 70% ethanol in between the sieving of separate samples. The total dry weight of the soil contained in each tray was recorded. Three soil samples from the sieved soils from each tray were collected: two approximately 10-gram soil samples were collected for soil chloroform extraction, and about 80-grams of soil was collected and frozen for the sequencing for soil bacterial (16s) and fungal (ITS) communities (see below).

Measurements:

Three types of measurements are contained in this dataset. Two separate types of respiration measurements were taken: 1) belowground (i.e., soil and root-system) respiration rate measurements for the *in-situ* root trays, and 2) specific root respiration rate measurement of excised root systems. Functional trait data was also collected for all root systems studied (i.e., both those housed within the *in-situ* root trays and the excised root systems) and for leaves of study trees.

In-situ measurements of tray respiration:

Gas exchange measurements for each *in-situ* root tray (taken biweekly to monthly for the duration of the experiment, see dataset characteristics above) were done using the Li-Cor Li-6800 (Lincoln, NE USA) portable photosynthesis system outfitted with a custom chamber (Figure 2). The pump was set to high, resulting in a flow rate of ~1500 µmols s^{-1}, with a slight chamber overpressure (0.1kPa). The relative humidity of the air flowing into the chamber was set to between 70-80% humidity depending on the environmental conditions (i.e., soil moisture and air temperature). A silica gel desiccant pack was place in the custom chamber just under the air inflow and outflow to limit the effect of moisture difference on the measurements. The CO_2 concentration of the incoming air was set to 400 µmol mol^{-1}. The air temperature of air inflow was not controlled. The mixing fan on the Li-6800 was set to 12500 rpm and the Auxiliar Power was set to 10 V, which resulted in a moderate spinning rate for the mixing fan located within the chamber.

Gas exchange measurement stability was monitored on two parameters: 1) the relative humidity of the chamber (RHChab), and 2) the respiration rate (R). For RHChamb, stability was defined as having a slope < 0.5% and a standard deviation of less than 1% over a 60 second window. For R, stability was defined as having a slope and standard deviation of <1 µmol m^{-2} s^{-1}. For measurement in the field, *in-situ* root trays were placed in the custom chamber and sealed around the protruding root using Oatey putty. The system was allowed to equilibrate, which took between 10 and 30 minutes per measurement, depending on the humidity of the air and the soil moisture conditions. Then, the data point was promptly logged. IRGAs were matched regularly.

Figure 4 (previous page). *Left:* Root system tissue respiration measurement set-up. Root systems were excised from identified trees (not those which were outfitted with the *in-situ* root trays, but comparable trees nearby within the same stands), washed, and respiration rates were measured at 25°C using the WALZ 3010-GWK1 chamber (Heinz Walz GmbH, Bamberg, Germany). *Right:* an entire root system is placed within the WALZ chamber.

Root tissue respiration measurements of excised root systems:

Entire root systems (containing three or more root orders) were gently excavated and excised on three occasions during the spring/summer of 2020. Root systems were excavated from similar trees next to the study trees outfitted with the *in-situ* root respiration trays, therefore the excised root systems should be comparable to those housed within the *in-situ* root trays. Root systems were thoroughly washed (removing all dirt, however in cases ectomycorrhizal fungi were left attached to the root, if removing them would have led to damage to the root system), and measured for respiration rates using the respiration system in Figure 2, where a WALZ chamber was attached the Li-6800. The WALZ respiration chamber was set to 25°C and the flow rate of the system was set to 600 µmol mol^{-1}. The fan mixing speed of the Li-6800 was set to 10000 rpm. The temperature and the relative humidity of the inflowing air were not controlled.

Measurements took about 5-10 minutes to reach stability, wherein they were promptly logged. The stability criteria used were: 1) the difference between the sample and reference CO_2 concentrations (ΔCO_2) slope of <0.25 µmol m^{-2} s^{-1} and standard deviation of <0.1 µmol m^{-2} s^{-1} over a 20 second interval, and the difference in the difference between the sample and reference air humidity (ΔH_2O) slope of <0.5 mol m^{-2} s^{-1} and standard deviation of <0.1 mol m^{-2} s^{-1}.

Quantification of leaf and root functional traits

Leaf and root functional traits were measured for each of the eight study species. Five leaves of each of the eight study species (one per study individual outfitted with *in-situ* root trays) were collected on August 29, 2019. Leaf thickness was promptly measured using a micrometer precise to the thousandth of a millimeter (Mitutoyo America). Leaves were then scanned at high resolution (1200 dpi) and images were analyzed in ImageJ (Schneider et al. 2012) for leaf area and other anatomical leaf attributed (leaf width and length etc.). Leaves were dried at 60°C for several days (to constant mass) and weighed for dry mass. Leaf tissue carbon (C), nitrogen (N) and phosphorus (P) were measured on dried leaf material. Dried leaf tissues were homogenized in sterile plastic screwcap vials (15mL centrifuge tubes) using sterile stainless

steel beads on an Sample Prep Mini-G 1600 (SPEX Inc. Metuchen, NJ, USA). Several 1000-rpm 30-second shaking cycles with 30-seconds rest bead-beat samples to fine powder. The homogenized powder was microweighed to ~2mg of sample in sterile tin capsules. The elemental carbon and concentration of samples was determined using a Model 4010 Elemental Combustion System (Costech Analytical Technologies, Valencia, CA, USA) at Oak Ridge National Lab. For tissue phosphorus concentration, a Kjeldahl digestion was used to create solutions of each sample. then samples were run on a QuikChem 8500 analyzer (Lachat Instruments, Loveland, CO, USA) using Lachat Quikchem Method 13-115-01-1-B at Oak Ridge National Lab.

Root functional traits were measured for both the root systems housed within the *in-situ* root trays (at the end of the experiment, after tray collection) and for the excised root systems. For all root systems, they were washed thoroughly then stored in the refrigerator until they could be scanned, usually within a few days of collection. Root systems were scanned in acrylic trays while submerged in water, at high resolution (1200 dpi). Root topological parameters (total root length, average diameter, root tip abundance etc.) were measured with WinRHIZO (2016 version (Regeant Instrument, Quebec, Canada). Root systems were dried in paper bags at 60°C for several days (to constant mass) and weighed for dry mass. Root tissue carbon and nitrogen were measured for the excised root systems (n=15 per species). Root tissue homogenization followed the same procedure as for leaves, except analyses were done at the Blue Carbon (Seagrass) lab at Florida International University using a Carlos Erba NA-1500 Elemental Analyzer (Fissons Instruments Inc., Danvers, MA, USA).

Measurements of soil carbon and nitrogen pools

Soil total (inorganic plus organic), total organic carbon and nitrogen were measured in the laboratory using a Shimadzu TOC-L CSH/CSN analyzer (located at Oak Ridge National Lab), and the microbial carbon and nitrogen pools were determined using the soil chloroform fumigation method (Brookes et al. 1985).

For the measurement of total nitrogen (TN), 24 ml samples are placed on an auto sampler. When a sample is introduced into the combustion tube (furnace temperature 720 C), the TN in the sample decomposes to become nitrogen monoxide. Nitrogen gas does not become nitrogen monoxide at this time. The carrier gas, which contains the nitrogen monoxide, is cooled and dehumidified by an electronic dehumidifier. Then it enters a chemiluminescence gas analyzer, where the nitrogen monoxide is detected. The detection signal from the chemiluminescence gas analyzer generates a peak and the TN concentration in the sample can then be measured.

For the measurement of total carbon (TC), the same sample is measured. In the TOC-L instrument, the carrier gas is controlled using a pressure regulator and mass flow controller. The carrier gas flows at rate of 150 mL min-1 to the combustion tube, which has been filled with an oxidation catalyst and heated to 680 °C. The TC of a sample is burned in the combustion tube to for carbon dioxide. The carrier gas, containing the carbon dioxide and other combustion products, flows from the combustion tube to an electronic dehumidifier, where it is cooled and dehydrated. Then it passes through a halogen scrubber before it reaches the cell of a non-

dispersive infrared NDIR gas analyzer, where the carbon dioxide is detected. The analog detection signal of the NDIR forms a peak, and the area of this peak is measured by an internal data processor. The area of the peak is proportional to the TC concentration of the sample. Therefore, when a TC standard solution has been analyzed to create a calibration curve, the equation expressing the relationship between TC concentration and peak area, the TC concentration in the sample can be calculated.

Dissolved organic carbon (DOC), dissolved nitrogen (DN), microbial biomass carbon (MBC), and microbial biomass nitrogen (MBN) were determined using the K_2SO_4 extraction and chloroform fumigation-extraction method. Briefly, 7 g soil was combined with 35 mL of 0.5M K_2SO_4 and placed on a shaker for 1 hour then filtered through Whatman #1 filters. A second set of soils were placed in a desiccator with 20 ml chloroform under a vacuum of 11 atms and fumigated for 48 hours prior to extraction with K_2SO_4 (as described above). Filtrate C and N contents were determined with the combustion catalytic oxidation method on the Schmadzu TOC-L analyzer. Unfumigated samples represent DOC and DN, while MBC and MBN are calculated as the difference between fumigated and unfumigated samples.

Sequencing of soil bacterial and fungal communities

Soil DNA extraction was performed with the DNEASY Powersoil HTP 96 Kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions with DNA eluted in 100 µL water. Extractions were quantified with the Qubit dsDNA BR Assay kit and diluted to a concentration of 10 ng μL^{-1}. A two-step PCR (polymerase chain reaction) amplification approach was used with barcode tagged templates and primers targeting the V4 region of the 16S rRNA gene for archaea & bacteria and the ITS2 region for fungi using pooled primer sets to increase coverage of archaeal, bacterial, and fungal taxa (see Table 2). PCR amplification of target regions was performed using the following thermal cycler conditions: 95°C for 3 minutes, 25 cycles of 95°C for 30 seconds, 78°C for 30 seconds, 55°C for 30 seconds and 72°C for 30 seconds, and a final extension of 72°C for 5 minutes. The second step of PCR indexed reads using the conditions: 95°C for 3 minutes, eight cycles of 95°C for 30 seconds, 55°C for 30 seconds and 72°C for 30 seconds, and a final extension of 72°C for 5 minutes.

After PCR amplification and indexing, all samples were pooled equimolar and purified with Agencourt AMPure XP beads (0.7:1 bead to DNA ratio; Beckman Coulter Inc., Pasadena, CA, USA). Paired-end sequencing (2 x 251) was completed on pooled prepared libraries on an Illumina MiSeq instrument (San Diego, CA) at Oak Ridge National Laboratory using V2 chemistry, which included a ≥15% PhiX sequencing control library. DNA sequencing data for soil bacteria and fungi are not included in this dataset. They have been archived in an sequence read archive in GenBank -- BioProject SRA # PRJNA786934: https://www.ncbi.nlm.nih.gov/bioproject/PRJNA786934h

Table 2. List of primer sequences used for polymerase chain reaction (PCR) amplification of fungal and bacterial DNA sequences.

Primer name	Sequence	Direction	Target	Reference
ITS3NGS1	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCATCGATGAAGAACGCAG	Forward	Fungi	White et al. (1990)
ITS3NGS2	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCAACGATGAAGAACGCAG	Forward	Chytridiomycota	Tedersoo et al. (2014)
ITS3NGS3	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCACCGATGAAGAACGCAG	Forward	Sebacinales	Tedersoo et al. (2014)
ITS3NGS4	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCATCGATGAAGAACGTAG	Forward	Glomeromycota	Tedersoo et al. (2014)
ITS3NGS5	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCATCGATGAAGAACGTGG	Forward	Sordariales	Tedersoo et al. (2014)
ITS3NGS10	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGCATCGATGAAGAACGCTG	Forward	Stramenopila	Tedersoo et al. (2014)
ITS4NGR	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGTCCTSCGCTTATTGATATGC	Reverse	Fungi	White et al. (1990)
ARCH-ITS4	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGTCCTCGCCTTATTGATATGC	Reverse	Archaearhizomycetes	Cregger et al. (2018)
515F	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGTGCCAGCMGCCGCGGTAA	Forward	Bacteria/Archaea	Lane et al. (1985)
515F_f1C	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGTGCCAGCMGCWGCGGTAA	Forward	Cloroflexi	Shakya et al. (2013)
515F_f1TM7	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGTGCCAGCMGCCGCGGTCA	Forward	TM7	Shakya et al. (2013)
515F_f4Arc	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGTGKCAGCMGCCGCGGTAA	Forward	Archaea	Shakya et al. (2013)
806R	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGGGACTACHVGGGTWTCTAAT	Reverse	Bacteria/Archaea	Lane et al. (1985)

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7. Data access

Get Data

For public access to data from the US Department of Energy Terrestrial Ecosystem Science Scientific Focus Ares (TES-SFA) please visit: https://tes-sfa.ornl.gov/node/80