### FY2022 PROGRESS REPORT OAK RIDGE NATIONAL LABORATORY'S TERRESTRIAL ECOSYSTEM SCIENCE — SCIENTIFIC FOCUS AREA

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### ABSTRACT

Understanding fundamental responses and feedbacks of terrestrial ecosystems to climatic and atmospheric change is the aim of the Terrestrial Ecosystem Science Scientific Focus Area (TES SFA). Improved predictive knowledge of ecosystem dynamics is the long-term motivation for our research. Overarching science questions are:

- 1) How will atmospheric and climate change affect the structure and functioning of terrestrial ecosystems at spatial scales ranging from local to global and at temporal scales ranging from sub-annual to decades and centuries?
- 2) How do terrestrial ecosystem processes, and the interactions among them, control biogeochemical cycling of carbon and nutrients, the exchanges of water and energy, and the feedback to the atmosphere, now and in the future?

The proposed science includes manipulations, multi-disciplinary observations, database compilation, and fundamental process studies integrated and iterated with modeling activities. The centerpiece of our climate change manipulations is the Spruce and Peatland Responses Under Changing Environments (SPRUCE) experiment that tests multiple levels of warming at ambient and elevated CO<sub>2</sub> on the vegetation response and biogeochemical feedbacks from a *Picea-Sphagnum* ecosystem. Other efforts aim to improve mechanistic representation of processes within terrestrial biosphere models by furthering our understanding of fundamental ecosystem functions and their response to environmental change. The TES SFA integrates experimental and observational studies with model building, parameter estimation, and evaluation to yield reliable model projections. This integrated model-experiment approach fosters an enhanced, interactive, and mutually beneficial engagement between models and experiments to further our predictive understanding of the terrestrial biosphere in the context of Earth system functions.

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### **1.0 PROGRAM OVERVIEW**

Oak Ridge National Laboratory's (ORNL) Terrestrial Ecosystem Science Scientific Focus Area (TES SFA) conducts fundamental research in support of the DOE BER Earth and Environmental Systems Sciences Division (EESSD) as outlined in the most recent Strategic Plan (US DOE 2018). The TES SFA addresses all five EESSD grand challenges by integrating data and models to understand and predict Earth System drivers and biogeochemical cycling, especially in critical ecosystems such as high-latitude regions. The TES SFA strives to expand fundamental knowledge of terrestrial systems and translate that knowledge into mechanistic Earth System Models (ESMs), especially the Energy Exascale Earth System Model (E3SM). The TES SFA also addresses grand challenges identified in the 2017 BERAC Grand Challenges Report (BERAC 2017) by advancing our understanding of the interactions among key players of the Earth System (e.g., microbial communities, vegetation, and humans) with a systems science approach. The TES SFA emphasizes information translation through levels of ecosystem organization connecting complex fine-scale biological processes with large-scale biosphere-climate feedbacks. Data management and access activities are an integral part of TES SFA efforts to share not only scientific progress but also data products and research results with the broader scientific communities.

## Vision: Improved integrative understanding of terrestrial ecosystem processes to advance Earth System predictions through experiment-model-observation synergy

The TES SFA is guided by the vision that sensitivities, uncertainties and recognized weaknesses of ESM predictions inform observations, laboratory and field experiments and the development of ecosystem process modeling. In turn, predictive understanding and findings from the field and laboratory and improved process modeling are incorporated, with the associated uncertainties, into ESMs as explicitly and expeditiously as possible. TES SFA research integrates laboratory and field experiments across a range of scales, observations from landscape greenhouse gas flux, field sites and remote sensing, and multiple process models. This integration is realized through the development and application of empirically-driven process model development, model-data fusion, model-data inter-comparison, model performance benchmarking, and uncertainty characterization and quantification. The integration occurs within the context of predictive Earth System modeling and within a framework of Earth system simulation using high-performance leadership-class computing.

TES SFA research is an iterative process (Fig. 1) translating mechanisms to ecosystem models with a quantitative understanding of model uncertainties. This process informs priorities for future measurements. Our paradigm is to identify and target critical uncertainties in coupled climate and terrestrial ecosystem processes and feedbacks, prioritized by their influence over global change predictions on decadal and century timescales. New measurements and experiments are employed to obtain new knowledge required to characterize, quantify, and reduce these uncertainties.



Fig. 1. Diagram of the TES SFA research philosophy and flow illustrating an iterative exchange between model projections, question or hypothesis development and the execution of observations and experiments to better understand impacts of multi-factor environmental changes on ecosystems. Terrestrial ecosystem research requires the integration of biophysical, biochemical, physiological, and ecological process understanding. Terrestrial ecosystem models integrate these processes in a mathematically consistent, meta-hypothesis on the coupled operation of the C, hydrological, and energy cycles at hourly to multi-annual timescales and at ecosystem to landscape spatial scales. Terrestrial ecosystem models are built upon, validated by, and constrained by historical and contemporary observations and experiments. Nevertheless, the future of terrestrial ecosystems remains highly uncertain. Further integration of models and experimental manipulations are required to enable reliable projections of ecosystem responses and feedbacks to future climate and other atmospheric forcing.

ORNL's current high-profile environmental change study, the Spruce and Peatland Responses Under Changing Environments experiment (SPRUCE), focuses on the combined response of multiple levels of warming at ambient or elevated CO<sub>2</sub> (eCO<sub>2</sub>) levels in a *Picea mariana–Sphagnum* peat bog in northern Minnesota. The experiment provides a platform for testing mechanisms that control vulnerability of organisms and ecosystem processes to important climate change variables which provide data for model development. The TES SFA also supports smaller-scale, process-level manipulations of ecosystem processes ranging from photosynthesis to root function to mechanistic studies of soil carbon (C) cycling, as well as long-term monitoring of landscape flux and sun-induced chlorophyll fluorescence (SIF) measurements at the Missouri flux (MOFLUX) site. TES SFA research on SPRUCE and MOFLUX covers a wide range of ecosystems in eastern North America from cool wet northern systems to warm and relatively dry systems on the forest-prairie transition in Missouri. This coverage allows for system comparison and for testing whether fundamental process knowledge gained from one system can be applied to the other.

Data from large- and small-scale TES SFA observations and experiments are integrated into models to identify and reduce terrestrial process and parameter uncertainties in the global Earth system. The Multi-Assumption Architecture & Testbed (MAAT) and Energy Exascale Earth System Model (E3SM) provide frameworks for this model-data integration and uncertainty quantification. Model predictions are improved through parameterization, calibration, and the development of new process-based submodels focused on key aspects of wetland, boreal and temperate forest systems, for example the E3SM Land Model (ELM) and the Microbial Enzyme Decomposition (MEND) model, which focuses on the belowground ecosystem.

The TES SFA engages broader scientific communities via multiple channels. FRED is a freely available resource for the broader community of root and rhizosphere ecologists and terrestrial biosphere modelers. Leafweb is a free online service tool for automatic photosynthesis analysis and has been used by photosynthesis researchers around the world. Both SPRUCE and MOFLUX provide site access and resources support to independent scientists from other institutions who conduct research at TES SFA sites with their own funding.

Given the opportunities for serendipity in scientific research, the TES SFA has built-in mechanisms within its overall planned and organized research tasks to allow timely exploration of emerging scientific issues that are unplanned but nevertheless important to the TES SFA vision and relevant to the EESSD's Strategic Plan. These mechanisms facilitate the formation of novel ideas and new research frontiers, as well as the growth of early career staff.

TES SFA research is ambitious in its scope, effort, and resource requirements. It undertakes the challenge of fully utilizing, testing and extending the broad interdisciplinary facilities of a DOE National Laboratory. ORNL's SFA research plans and philosophy attempt to eliminate an artificial distinction between experimental or observational studies and modeling so that the science can be advanced efficiently and effectively.

### 2.0 SCIENCE QUESTIONS, GOALS AND MILESTONES

The following overarching science questions and the subsequent description of key goals and milestones are focused on resolving uncertainties in terrestrial ecosystem response to atmospheric and climate change.

- 1) How will atmospheric and climate change affect the structure and functioning of terrestrial ecosystems at spatial scales ranging from local to global and at temporal scales ranging from sub-annual to decades and centuries?
- 2) How do terrestrial ecosystem processes, and the interactions among them, control biogeochemical cycling of carbon and nutrients, the exchanges of water and energy, and the feedback to the atmosphere, now and in the future?

## **Goals and Milestones**

The TES SFA goals and long-term (5 to 10-year) milestones are summarized below. Details on progress and proposed work are documented in Section 4.

- *Goal 1*: Understand, quantify and model long-term ecosystem responses to the interactive effects of atmospheric and climatic change in an understudied but globally important ecosystem.
  - <u>Long-term milestone</u>: Comprehensive predictive understanding of decadal peatland ecosystem responses to a range of technologically advanced warming and elevated [CO<sub>2</sub>] treatments in an ombrotrophic bog in northern Minnesota.

<u>*Goal 2*</u>: Understand drivers of ecosystem functions and interactions by integrating new process knowledge in the ELM and related models of varying complexity, and the use of state-of-the-art sensitivity analysis, uncertainty quantification, and model evaluation to obtain novel insights leading to new observations and experiments.

• <u>Long-term milestone</u>: Increase confidence in model projections based on improved mechanistic understanding and model representation of ecosystem processes, ranging from canopy photosynthesis to microbially-mediated decomposition, that govern responses to, and interactions with, environmental change.

<u>Goal 3</u>: Improve the understanding and model representation of the linkages among above- and belowground functional traits, their response to changing environmental conditions, and the resulting consequences for ecosystem biogeochemical cycling.

• <u>Long-term milestone</u>: Develop global ecological trait databases, available to the broader community of ecologists and terrestrial biosphere modelers and use these databases to target novel observations and experimental manipulations that fill gaps in our mechanistic understanding and modeling of key ecological traits.

<u>Goal 4</u>: Improve process-based understanding of belowground biogeochemical cycling, including processes occurring at the root-soil interface, to support predictions of small- and large-scale belowground pools and fluxes in terrestrial biosphere models.

• <u>Long-term milestone</u>: Incorporate a flexible, comprehensive, and tested model of the belowground ecosystem into fully coupled ESMs.

<u>Goal 5</u>: Achieve a predictive understanding of ecophysiological, biochemical, and physical processes controlling the exchanges of carbon, water, and energy between land and atmosphere with advanced observational and theoretical approaches.

• <u>Long-term milestone</u>: Develop and demonstrate a mechanistic model of photosynthesis involving light reactions for applications in ESMs, a light reactions-based approach for partitioning net ecosystem changes of carbon and water, and a reliable, easy-to-use SIF measurement system for flux communities around the world.

## **3.0 TES SFA PROGRAM STRUCTURE AND PERSONNEL**

Responsibility for the TES SFA resides within DOE's Climate and Environmental Sciences Directorate and is aligned with associated and related activities of the Climate Change Science Institute (CCSI) at ORNL. The organization chart for the TES SFA is presented in Fig. 2. The TES SFA includes a science and management organization to guide and direct research activities. The TES SFA Leadership Team, comprised of the individuals listed in Fig. 2, provides advice on the yearly SFA plans and budgets, monitors progress, adjusts project plans as appropriate, directs informatics development efforts, and resolves issues in a timely manner. The TES SFA is supported by 24 dedicated ORNL scientific and technical staff including 5 current postdocs. Over 100 individuals from the USDA Forest Service, and various other collaborating universities and laboratories are participating in the SPRUCE and MOFLUX projects. We have brought together exceptional multidisciplinary expertise and are retaining and building staff flexibility to support new research priorities as they are identified.

- Dr. Paul J. Hanson is the Coordinating Investigator and provides integrated leadership across tasks and coordinates financial management.
- Dr. Daniel M. Ricciuto is the Coordinating Investigator for terrestrial C-Cycle modeling activities.
- Terri Velliquette serves as the Data Management Coordinator. She brings expertise and technical skills for data policy, management, and archive planning and implementation.



Fig. 2. Organizational chart for the TES SFA (2022 status).

Individual Task lead responsibilities are as follows:

### Large Scale Manipulations (LSM) and Landscape Observations (LO) Task 1 – SPRUCE Personnel

<u>Experimental design, maintenance, and environmental documentation</u> – Paul Hanson leads operations of the SPRUCE infrastructure together with a team of ORNL structural and electrical engineers. Kyle Pearson (an ORNL employee located full-time in Minnesota) leads day-to-day onsite activities at the SPRUCE site with the help of a support technician (to be hired). Kyle is supported by Jeff Riggs (Lead Instrument Technician) to keep the treatments running and data streams flowing. Misha Krassovski, systems engineer, designed, implemented and now maintains the automated data acquisition systems.

<u>Plant growth, Net Primary Productivity (NPP) and phenology</u> – Paul Hanson is leading tree and shrub growth assessments with the participation of Jana Phillips. David Weston now leads the characterization of growth and community dynamics of the diverse *Sphagnum* communities. Belowground growth measurements are led by Colleen Iversen in collaboration with current postdoctoral staff, and with technical assistance from Joanne Childs and John Latimer. Vegetation phenology efforts are being led by Andrew Richardson (Northern Arizona University) with onsite manual observations collected by Kyle Pearson.

<u>Community composition</u> – Community compositional changes are being led by Brian Palik of the USFS with the participation of Rebecca Montgomery (Univ. of Minnesota). Chris Schadt leads efforts on microbial community changes and coordinates related efforts among the SPRUCE collaborators.

<u>Plant Physiology</u> –Plant physiological carbon and water relations and ecophysiological responses are led by Jeff Warren and current postdoctoral and technical staff. We are actively encouraging external participation in associated tasks: including collaborative research on gas exchange, carbohydrate dynamics, C partitioning, and woody and root respiration assessments.

<u>Biogeochemical cycling responses</u> – Work on hydrologic cycling and porewater biogeochemistry is led by Natalie Griffiths and Steve Sebestyen (USDA FS) with technical

assistance from Keith Oleheiser. Colleen Iversen leads the subtask focused on plant nutrient availability in the shallow rhizosphere with technical assistance from Joanne Childs and John Latimer. C-Cycle observations focused on peat changes and C emissions are coordinated by Paul Hanson. Natalie Griffiths coordinates with Colleen Iversen, Randy Kolka (USFS), and external investigators on extensive decomposition studies. Verity Salmon coordinates compilation of peat, plant and porewater data for analysis of ecosystem-level N and P biogeochemical cycles.

<u>Modeling of terrestrial ecosystem responses to temperature and CO<sub>2</sub></u> – Daniel Ricciuto coordinates efforts to utilize and incorporate experimental results into improved modeling frameworks for understanding the peatland C-Cycle and its feedbacks to climate together with Xiaoying Shi, Jiafu Mao and Xiaojuan Yang.

A coordinating panel is made up of the Response SFA research manager (Hanson), the local USFS contact (Kolka), the Technical Task leaders listed above, and an external SPRUCE advisory committee. The panel serves as the decision-making body for major operational considerations and the decision-making body for vetting requests for new research initiatives to be conducted within the experimental system.

Task 6 – Lianhong Gu leads activities in the Coordinated Eco-physiology, Eddy covariance, and Sun-Induced chlorophyll Fluorescence Studies (CEESIFS) to advance integrative ecosystem science. Jeff Wood (University of Missouri) is subcontracted to operate the MOFLUX on-site activities. Other contributing staff include Colleen Iversen, Melanie Mayes, Anthony Walker, Jana Phillips and Joanne Childs.

## Model Development and Evaluation (M)

- Task 3abc C-Cycle modeling activities are led by Daniel Ricciuto. Subtask contributions are made as follows: Wetlands [Xiaoying Shi, Xiaofeng Xu (San Diego State)], Phenology (Jiafu Mao, Daniel Ricciuto), Photosynthesis (Anthony Walker), rhizosphere (Xiaojuan Yang), ecological forecasting [Daniel Ricciuto, Yiqi Luo (NAU)], wildfire analysis [Mingzhou Jin (UTK), Jiafu Mao], global SIF analysis [Anping Chen (Colorado State University), Jiafu Mao, Daniel Ricciuto], model reduction using representativeness (Kumar), and model intercomparisons (Daniel Ricciuto, Jiafu Mao, Xiaoying Shi).
- **Task 3d** Multi-Assumption Systems Modeling task is being led by Anthony Walker to develop the Multi-Assumption Architecture Testbed (MAAT) for analyzing model process representations and to apply MAAT to specific TES-SFA science questions.

### **Process-Level Studies (PLS)**

- **Task 4c** –Jeff Warren leads the initiative to experimentally link root function to specific root traits in collaboration with Colleen Iversen, post-docs and students.
- **Task 5** Melanie Mayes provides expertise in soil C cycling and Chris Schadt in microbial ecology to investigate microbial processes and develop the MEND model for soil C cycling.

## **Global Trait Databases and Analysis Toos (GT)**

- **Task 4b** Colleen Iversen leads the root trait initiative including the development of a global root ecology database to inform the treatment of belowground processes in ecosystem models.
- **Task 8** Lianhong Gu supports and manages LeafWeb, a global database and analysis tool of photophysical, photochemical, and biochemical properties of leaves.

The TES SFA benefits from a SPRUCE advisory panel that is dedicated to providing guidance on the science and operation of our flagship experiment. In FY2022 the active membership of the SPRUCE Advisory Panel included: Serita Frey (University of New Hampshire), Camilo Rey-Sanchez (University of California - Berkeley), Nigel Roulet (McGill University) and Nate McDowell (Pacific Northwest National Laboratory). We have also engaged with Paul Glaser (University of Minnesota) and Heidi Asbjornsen (University of New Hampshire) for future interactions.

## 4. PERFORMANCE MILESTONES AND METRICS

This section represents a summary of TES SFA activities accomplished since the review document dated June 2020. The material is organized by research theme and task with parenthetical identification of the goals addressed by each task. Task numbers are unchanged for reference across historical planning and review documents even though task titles may have been edited over time.

### Large-scale Manipulations and Landscape Observations

Task 1: Spruce and Peatland Responses Under Changing Environments – SPRUCE (Goals 1,2,3,4,5) Task 6: Coordinated Eco-physiology, Eddy covariance, and Sun-Induced chlorophyll Fluorescence Studies (CEESIFS) to advance integrative ecosystem science (Goal 1,2,3,4,5)

### **Carbon-Cycle Focused work**

Task 3a: Improving process models with site-level observations and experimental data (Goals 1,2,3,4).

Task 3c: Functional Testing (Goals 1,2,3,4)

Task 3d: Multi-Assumption Systems Modeling (Goals 2,3,5)

### **Process-Level Studies (PLS)**

Task 4c: Root function and modeling (Goals 2,3,4) Task 5: Microbial processing of soil carbon (Goals 2,4)

## **Global Trait Databases and Analysis Toos (GT)**

Task 4b: Root traits characterization (Goals 2,3,4) Task 8: LeafWeb data assimilation tool –New Task (Goal 3)

Following the description of progress for each TES SFA science task, a table of anticipated deliverables is provided with annotations regarding progress. Task-specific publications and completed manuscripts are listed by Task. Some citations may be repeated when multiple tasks contributed to the product. The number of new data sets established by each task are also noted with details presented in Appendix B.

## 4AI. REVIEW OF SCIENTIFIC PROGRESS BY TASK

## LARGE-SCALE MANIPULATIONS AND LANDSCAPE OBSERVATIONS

## **Task 1: SPRUCE Infrastructure**

SPRUCE warming treatments ran full time throughout 2021. Warming treatments were maintained day and night throughout the year with only minor interruptions. The  $eCO_2$  exposures are applied only during daytime hours during the active growing season (April through November). Table 1 shows the achieved whole-ecosystem warming treatments and  $eCO_2$  treatments for the 2020 calendar year. Treatment data are archived in Hanson et al. (2016D).

Plot #s	Target	Mean Air	Mean Soil	Ambient Daylight	Elevated	
	Temperature	Temperature	Temperature	Mean Growing	<b>Daylight Mean</b>	
	Differential	at +2 m	at -2 m	Season [CO2]* at	<b>Growing Season</b>	
				+2 m	[CO <sub>2</sub> ]** at +2 m	
	(Delta °C)	(°C)	(°C)	ppm	ppm	
Plots 7 & 21	Ambient	6.0,6.4	5.5,6.2	425		
Plots 6 & 19	+0	7.7,7.8	5.0,6.2	434	825	
Plots 11 & 20	+2.25	9.9,9.6	7.3, 7.5	424	838	
Plots 4 & 13	+4.5	12.1, 12.2	9.6, 9.6	429	851	
Plots 8 & 16	+6.75	14.5 , 14.4	11.7, 11.7	432	907	
Plots 10 and 17	+9.0	16.4, 16.5	13.9, 13.7	434	883	

 Table T1.1. Mean annual air and soil temperatures and CO2 concentrations by SPRUCE plot for

 2021. Plot numbers in red text correspond to elevated CO2 treatments.

\*For the purpose of this 2021 enumeration of  $eCO_2$  exposures the growing season runs from day of the year 110 through 305 to match the active season in the +9 °C treatment plots.

\*\*Elevated CO<sub>2</sub> would approach 900 ppm in all plots if a specific active season definition were used for each warming treatment.

In 2020, the unique isotopic signatures of air in the added CO<sub>2</sub> treatments continued to be approximately -27 to -28  $\partial$  ‰ for <sup>13</sup>C and -540  $\Delta$  ‰ for <sup>14</sup>C. Through 5 full active seasons of eCO<sub>2</sub> exposures new tissue growth under eCO<sub>2</sub> has stabilizing at a new isotopic signatures commensurate with the experimental exposures to eCO<sub>2</sub>. Tissue <sup>13</sup>C and <sup>14</sup>C signatures for *Sphagnum* and *Maianthemum* plants are different that for the taller plant species because they reincorporate respired forms of [CO<sub>2</sub>] from the peat profile. The newest data for 2021 samples are still pending.

### The 2021 Drought

In summer of 2021 the research site experienced an "extreme drought: as characterized by the Drought Monitor product developed and distributed by NOAA scientists (https://droughtmonitor.unl.edu/Maps/MapArchive.aspx).

The driest year and lower water tables for the Marcell Experiment forests were recorded in 1976, a year with only 412 mm of precipitation representing 52% of the annual average precipitation inputs. Precipitation in 2021 was 663 mm. In the warmest plots, the water table depths approached those of the historic 1976 (Figure T1.1).



Fig. T1.1. Normalized water table depths for all plots and warming treatments from 2016 through 2021.

Water Table Recession (Jonathan Stelling, University of Minnesota)

Daily water loss measurements show for the period of 2015-2018 that there was little difference in water table recession (daily lowering rates) across the temperature treatments. Recession rates in 2019-2021 saw a significant response to temperature treatment, with recession rates increasing approximately 0.6 mm/day per °C of warming. When comparing the 2021 drought to historic low water table in the S1 bog of >1 m below the surface (i.e., 1976). This depth was nearly reached by the highest temperature treatment plots. Over the 2021 summer season the +9°C plots saw nearly 80 cm of water table drop, and the +0°C plots approximately half that. The probability of a drought that lowered bog water tables to this severity occurs approximately 11 in 100 years, therefore it can be representative of decadal drought regime.

### **Disruption of Treatments**

The first major disruption of SPRUCE air-warming treatments since the inception of operations in August of 2015 occurred on Friday, 16 June 2022 at 1700 CST. The propane vaporizer serving Transect #3 and warming plots 16, 17 and 20 failed. The cause of the failure is unknown and was unobserved. The resulting "burn" of equipment and small amounts of adjacent surface litter and foliage has been recorded and a safety review is underway. Replacement hardware is being acquired for installation following the recommendations of the ORNL review. A separate set of presentation materials on the vaporizer failure,

burn and safety recommendations will be prepared and shared in due coarse. Environmental monitoring in the affected Plots continues uninterrupted and the quantitative record of the temperature treatments for those plots remains intact. Independent belowground warming using electrical resistance heating continues.

### **Task 1: SPRUCE Response Data**

<u>Carbon Cycle Assessments for the SI Bog</u> – Component annual carbon exchange data (NCE; gC m<sup>-2</sup> y<sup>-1</sup>) for all plots and the first six years of the SPRUCE study show that autotrophic decreases for *Sphagnum* and trees is partially offset by gains by the shrub community and fine root processes belowground. The overall C sink capacity declines with warming driven mostly by heterotrophic C losses, and the combined plot-level estimate of NCE shows that loss rates per degree C are similar through 6-years of manipulation without a strong eCO<sub>2</sub> effect (Fig. T1.2).



Fig. T1.2 Estimated ecosystem net C exchange (NCE; g C  $m^{-2} y^{-1}$ ) for all treatment plots (+0, +2.25, +4.5, +6.75 and +9 °C treatments) plotted against the mean annual air temperature at +2 m for each plot in 2016, 2017, 2018, 2019, 2020 and 2021. NCE was calculated as the difference in measures of above- and belowground net primary production and C losses via heterotrophic CO<sub>2</sub> efflux, net CH<sub>4</sub> efflux, and combined total organic C and dissolved inorganic C efflux. Treatment plots receiving elevated atmospheric CO<sub>2</sub> exposures (EANPP) are indicated by orange symbols.

Because COVID-19 protocols kept us from routine access to the SPRUCE site in 2020 we do not have independent evaluations of all components of NCE for each treatment plots for 2020 and we substitute mean data for 2019 and 2021.

## Dramatic Spring Warming Tree Foliar Tissue Damage in 2022

On 19 and 20 June 2022 midday air temperatures in Plot 10 reached 46 to 48 °C, PhenoCam images document a dramatic transition during this period of healthy Picea foliage to necrotic tissues (Fig. T1.3; red brown foliage). Prior to this event the only similar transition occurred following the spring freeze event of April 2016.



Fig. T1.3 Pre-18 June 2022 (left Image) and post-heat wave (right image) PhenoCam images for the +9 °C treatment Plot 17.

<u>Sphagnum production and Community Response</u> – Our prior research showed that the SPRUCE warming manipulation alters the *Sphagnum* associated microbiome and negatively impacts N2-fixation (Carell et al. 2019). To investigate the role that the microbiome has on *Sphagnum* growth and acclimation to elevated temperatures, we performed microbiome transfer studies to test whether the thermal origin of the microbiome influences acclimation of *Sphagnum* growth to treatment temperature.

We leveraged the SPRUCE whole-ecosystem warming study to collect field-grown *Sphagnum*, mechanically separate the associated microbiome and then transfer onto germ- free laboratory *Sphagnum* for temperature experiments (Fig. T1.4). Host and microbiome dynamics were assessed with growth analysis, Chlorophyll-*a* fluorescence imaging, metagenomics, metatranscriptomics and 16S rDNA profiling.



Fig. T1.4 (a) Experimental approach and design: field-collected donor moss microbiomes collected from ambient or warming conditions were transferred to germ-free recipient moss (Sphangum angustifolium), and the resulting communities were then placed in an ambient or warm growth chamber. (b) Average moss growth rate under ambient or warming treatments, as a function of the thermal origin of the microbiome. Error bars represent standard error of the mean of n = 6 for 2016, n = 12 for 2017. (c) Relative abundance of microbiome phyla, determined by 16S rDNA amplicon sequencing of the starting field-collected inoculum (n = 3 of each composite sample) from ambient or warming experimental plots, and the final compositions of experimental samples (n = 6 for each condition). An asterisk indicates statistical significance (P < 0.05) based on a Tukey's HSD post hoc test of the percentage change of total growth between moss with a microbiome and moss without a microbiome within the same chamber.

Microbiomes originating from ambient +9 °C enclosures imparted enhanced thermotolerance and growth recovery at elevated temperatures (Fig. T1.4). Metagenome and metatranscriptome analyses revealed that warming altered microbial community structure in a manner that induced the plant heat shock response, especially the HSP70 family and jasmonic acid production. The heat shock response was induced even without warming treatment in the laboratory, suggesting that the warm-microbiome isolated from the field provided the host plant with thermal preconditioning.

# Our results demonstrate that microbes, which respond rapidly to temperature alterations, can play key roles in host plant growth response to rapidly changing environments.

<u>Belowground Dynamics</u> - The dynamics and distribution of fine roots in ecosystems underlain by organic soils are some of the least understood processes in belowground ecology. Building on our work to understand fine roots in an ombrotrophic bog prior to initiation of climate change treatments (Iversen et al., 2018, *Plant & Soil*), we leveraged on-going data collection (e.g., from manual and automated minirhizotrons, root ingrowth cores, and ion-exchange resins; data citations: Childs et al. 2019, 2020; Iversen et al. 2017a,b, 2021; Malhotra et al. 2020; paper citations: Malhotra et al. 2020, *PNAS*; Defrenne et al. 2021, *PPP*, Iversen et al. 2022, *Ecosystems*) to address two main questions:

1. How does warming affect root and fungal growth, and how are belowground dynamics related to edaphic and environmental conditions?

Papers in recent years led by SPRUCE ORNL post-docs have highlighted increases in ericaceous shrub root growth with warming (using root ingrowth cores, Malhotra et al. 2020) as well as increases in ectomycorrhizal fungi growth and an extended belowground growing season with warming (using automated minirhizotrons, Defrenne et al. 2021). Post-doc Dr. Camille Defrenne took a position with Eric Lilleskov with the PeatCosm experiment in September 2021, but PhD student Katherine Duchesneau in the Kostka lab continues to collaborate with Camille to incorporate her SPRUCE root and fungal trait data into a manuscript using genomics to investigate rhizosphere community structure assessed using portions of our root ingrowth cores. A new postdoc, Sören Weber, begins in July 2022 and will focus on changes in root phenology and rooting depth distribution with warming (using manual minirhizotron data). We continue to collect all three rooting distribution and dynamics data streams (ingrowth cores, manual and automated minirhizotron images) and we continue to analyze them as new stories emerge. Together these data streams inform the treatment of belowground dynamics in ELM-SPRUCE.

2. Does warming increase plant-available nutrients over time and throughout the peat profile?

This year, we published a paper on the impacts of warming and elevated  $[CO_2]$  on plant-available nutrients from 2014 to 2018 (Iversen et al. 2022, *Ecosystems*, data citation: Iversen et al. 2017b). We asked whether a gradient of whole-ecosystem warming (from  $+0^{\circ}$ C to  $+9^{\circ}$ C) would increase plantavailable nitrogen and phosphorus in an ombrotrophic bog in northern Minnesota, USA, and whether elevated [CO<sub>2</sub>] would modify the nutrient response. We tracked changes in plant-available nutrients across space and through time, and assessed whether nutrient warming responses were captured by a point version of the land-surface model, ELM-SPRUCE. We found that warming exponentially increased plantavailable ammonium and phosphate, but that nutrient dynamics were unaffected by elevated [CO<sub>2</sub>]. The warming response increased by an order of magnitude between the first and fourth year of the experimental manipulation, perhaps because of dramatic mortality of Sphagnum mosses in the surface peat of the warmest treatments. However, neither the magnitude nor the temporal dynamics of the responses were captured by ELM-SPRUCE. Future predictions of peatland nutrient availability under climate change scenarios must account for dynamic changes in nutrient acquisition by plants and microbes, as well as microtopography and peat depth. Building on this data stream, we have analyzed nutrient availability data in surface hollows from 2019 to 2021 for a manuscript on nitrogen fixation in Sphagnum mosses being led by Dr. Caitlin Petro, a post-doc in the Kostka lab; we see a developing interaction between warming and elevated [CO<sub>2</sub>], where NH<sub>4</sub>-N availability is depressed under elevated  $[CO_2]$  in the warmest plots.

<u>Woody Plant Physiology</u> – The primary focus of FY2021/2022 was on intensive hydraulic measurements, processing lab samples, analyzing automated data streams, and development of datasets and publications. We continued our focus on hydraulic stress and its linkages to visible damage to foliar loss and branch hydraulic failure. Initial results from our ongoing non-structural carbohydrate analyses suggest similar magnitudes of available carbon across treatments, indicating that damage to plants is more likely due to hydraulic failure than to carbon starvation. New assessments of multi-year water potential dynamics indicate the warming-induced water stress seen in first few years continues, particularly for *Larix* and *Chamaedaphne*, but also to a more moderate degree in the *Picea*. The % loss of xylem conductivity increased with warming for both tree species, illustrating the mechanism for observed water potential stress. Using stomatal closure curves paired with xylem water potential indicates a non-linear response of stomata to drying conditions, indicating a potential treatment adjustment of stomatal behavior.

We have analyzed four years of sap flow data and are currently analyzing the 2021 dataset. From these data we extracted sap flow initiation and cessation dates for each tree across the treatments and found that the mean active season as defined by tree transpiration was extended by 42 days for both *P*.

*mariana* and *L. laricina* in the +9 plots compared to controls, four weeks earlier in the spring and two weeks later in the fall (Fig. T1.5). Such sapflow data will be compared to a new analysis and manuscript from the PhenoCam dataset (Schädel et al., in review) that defines the active season as a change in foliar greenness of 25% of the seasonal greenness index amplitude; which reflects a shorter active season by that definition. Sap flow data are further going to be used to scaling up to whole tree and plot level tree transpiration and gross primary production.



Fig. T1.5 As pretreatment allometry has likely shifted due to the treatments there is a strong need to establish an alternate method for estimating leaf area index of the trees – a parameter that is required for scaling transpiration from sap flow measurements to the plot level. As such, we have engaged with the Boise State Lidar team to develop algorithms for assessment of leaf area index directly from the Lidar scans. To test the algorithms we will leverage earlier paired measurements of Lidar and destructive allometry on trees from outside the SPRUCE enclosures.

We are also continuing our focus on linking tree water stress to remote sensing based on simultaneous drone imaging and physiological measurements to assess diurnal and seasonal impacts of treatments on individual trees. Imagery will link sap flow, water potential and foliar hydration to canopy temperatures, and multispectral imaging to foliar nutrient status. Successful test samples were collected last summer, and intensive measurements are planned for July 2022.

Initial work with a limited set of branch anatomy samples has demonstrated that the elevated  $CO_2$ , in combination with the warming treatment increased xylem wood production in branches, which can facilitate greater water movement though the stem. A temperature-induced change in hydraulic anatomy or a shift in leaf area to sapwood area has been expected, and comprehensive destructive assessments are planned for the end of the study.

In 2021 we also sampled peat, sphagnum and woody plant xylem water isotopes, which might be linked to evaporative dynamics, water sourcing and the D-excess (strange isotope enrichment) that has been seen in the pore water isotope samples (Stelling). Samples will be processed in FY 23. Data will be linked to soil water drying dynamics, as measured by water table depth, relative measurements of peat volumetric water content, and potentially new measurements of peat water potential as funding allows.

We continue to analyze data from 2017, 2019 and 2021 gas exchange campaigns, and several manuscripts are in development. Acclimation has been seen in trees, where the thermal optimum of net photosynthesis increased with warming in both species and was higher in elevated  $CO_2$  due to suppression of photorespiration. Shrub data are more variable and have strong seasonal changes that will warrant more investments in gas exchange as planned for 2024. These campaign-style measurements will be informed by modeling of current data and will be used to characterize long-term acclimation of photosynthesis and respiration to combined warming and elevated  $CO_2$  in shrubs and the two boreal conifers.

<u>Water Chemistry and Fluxes</u> – Solute concentrations in outflow continue to respond to warming. Higher total organic carbon (TOC) concentrations (Fig. T1.6A), along with some cations and metals (i.e., calcium, aluminum, iron), were observed in warmer enclosures while the responses of total N and total P concentrations to warming were variable. The volume of water leaving the enclosures via lateral flow (i.e., stream flow) was generally lower under the warmer treatments (Fig. T1.6B), likely due to increased evapotranspiration. These low flows were very apparent during the 2021 drought, and flow ceased from

the warmest enclosures for most of the year. While the concentrations of some solutes have increased in outflow with warming, the fluxes of these solutes are generally lower from the warmest enclosures (Fig. T1.6C) as fluxes are driven primarily by changes in flow rather than changes in chemistry. These measurements will continue in FY23 and will focus on quantifying any post-drought changes in solute concentrations and fluxes.



Fig. T1.6 (A) Total organic carbon (TOC) concentrations in lateral outflow (i.e., stream flow), (B) cumulative annual lateral outflow, and (C) cumulative annual TOC flux from SPRUCE enclosures and responses to warming over six years (2016-2021).



Fig. T1.7 Relationships between soil temperature and tensile strength loss (cotton strip decomposition) with depth (from 5 to 135 cm deep) in winter (top left) and summer (top right) and relationships between water level (an indicator of soil moisture) and tensile strength loss in near-surface peats (from 5 to 25 cm deep) in winter (bottom left) and summer (bottom right).

<u>Decomposition</u> – The second-to-last set of litterbags was retrieved from the bog in fall 2021 (after 6 years of incubation), with one final (10-year) retrieval planned in 2025. The "year 6" samples are currently being processed in the laboratory. Biannual cotton strip retrieval occurred in 2021 and these data continue to reveal strong effects of warming on labile carbon decomposition (cotton strips are 95% cellulose), with greater decomposition rate of labile C in warmer enclosures throughout the peat profile (Fig. T1.7). While temperature appears to be a strong control on labile C decomposition, soil moisture is the predominant driver in the near-surface peats (Fig. T1.7), which has implications for interpreting decomposition drivers in our litterbag study (the litterbags are deployed in the top 0-20 cm of peat). Interestingly, the relationship between cotton strip decomposition and temperature was consistent with depth (i.e., from ~35 to 135 cm deep) suggesting a lack of microbial limitation on labile C decomposition at depth. A manuscript describing the cotton strip results is being drafted.

<u>Nutrient Cycling</u> – Across the SPRUCE experiment, multiple independent data streams quantify pools and fluxes of N and P in plants, peat, and water. Availability of these nutrients limits plant productivity in ombrotrophic peatlands (Bridgham et al., 1998; Bubier et al., 2007), so quantifying the impact of warming and eCO<sub>2</sub> on these nutrient cycles will yield important insight into peatland C balance under real and simulated climate change. We therefore are working to compile existing N and P data from the first six years of SPRUCE manipulative treatments as well as quantify how five years of warming impacted turnover of these critical nutrients in rhizosphere soils. These efforts will address two key questions: 1. How do warming and elevated CO2 impact the cycling of N and P through peat, water, and plants at SPRUCE?

A synthesis of nutrients in plants, peat, and water is underway to assess the impact of warming and elevated CO<sub>2</sub> on pools and fluxes of nitrogen (N), and phosphorus (P). For plants, we hypothesized that nutrients in biomass pools and net primary production (NPP) fluxes of N and P would increase with warming due to higher availability of soil N and P in warmed plots (Iversen et al., 2022). To test this hypothesis, we brought together above and belowground biomass and NPP measurements on trees, understory species, and Sphagnum moss from 2016 through 2020. Our preliminary results indicate that C, N, and P in biomass pools decreased with warming (Fig. T1.8) though the decrease in biomass N was somewhat offset by a positive interaction between the elevated CO<sub>2</sub> and warming treatments. The overall pattern of losses was predominantly driven by the decline of Sphagnum cover warmed plots (Norby et al., 2019). Observed increases in understory shrubs (Malhotra et al., 2020; McPartland et al., 2020) were not sufficient to cause net accumulation of N and P in plant biomass in the warmest plots. The observed increase in nutrient availability observed in soils at SPRUCE could be due to release of N and P from recently deceased Sphagnum. Currently, we are processing 2021 plant samples in the laboratory for inclusion in this analysis. We are also working to synthesize N and P in peat, porewater and lateral outflow from the SPRUCE plots to determine whether N and P are being lost under warmed conditions. Once we have compiled this nutrient budget, we plan to compare our empirical results to pools and fluxes of C, N and P in ELM-SPRUCE model output. Such a comparison will highlight areas where improvements can be made to both model representation of this peatland ecosystem as well empirical quantification of N and P pools and fluxes.

2. What is the impact of warming on soil N and P pools, turnover, and rates of mineralization?

The impact of five years of warming on soil N and P pool size, pool turnover, and rate of mineralization are currently being assessed with a one-year soil incubation. We hypothesize five years of warming in the field has accelerated soil decomposition and reduced the size of labile C, N and P. This is supported by both observed losses in ecosystem C (Hanson et al., 2020) and increased availability of mineralized N and P in soils (Iversen et al., 2022). For this incubation experiment, soils were collected in July of 2021 from the top 30 cm of the SPRUCE peat profile. Soils from each 10 cm depth intervals were then distributed into plastic funnels and placed into an incubator at 30 °C in October of 2021. Deionized water has been continuously added to soils to keep them at field capacity. At periodic timepoints during the incubation, C, N and P losses are being quantified in soil gas fluxes and soil leachate (Fig. T1.9). Leachate solution is analyzed for DOC, TN, NH<sub>4</sub>, NO<sub>3</sub>, PO<sub>4</sub> and gas fluxes are capturing production of CO<sub>2</sub>, CH<sub>4</sub>, N<sub>2</sub>O, NH<sub>3</sub>. Paired measurements of soil leachate and gas fluxes have been performed at seven timepoints thus far. Three remaining timepoints are scheduled prior to the incubation's completion. DOE SULI program participant Shabré Eskridge (University of Tennessee, Knoxville, TN) is working on the incubation experiment during the summer of 2022, measuring bulk soil %CNP, performing acid-fluoride digests for initial pools of available P, and processing gas flux data in R. The effluxes of C, N and P from this experiment will be used to quantify labile versus recalcitrant pool sizes and turnover rates for C, N and P using a two-pool kinetic model (Bridgham et al., 1998).



Fig. T1.8 Decreased accumulation of C (panel a), N (panel b) and P (panel c) in plant biomass following five years of manipulative warming at SPRUCE. Delta biomass terms on the y axes were calculated as 2020 values minus pretreatment values of tree, understory, and *Sphagnum* moss. All elements showed significant decreases in response to higher mean annual temperatures (MAT). Decreases in Delta biomass N with warming were partially offset by a positive interaction with elevated CO<sub>2</sub> treatment.



Fig. T1.9 SPRUCE soils from the upper 30 cm of the peat profile have leachate collected (panel a). At ten timepoints throughout the incubation experiment (panel b), leachate and gas fluxes are sampled. The resulting losses of C, N and P will be used to quantify the lability, pool size, and turnover of these elements.

<u>Microbial Community Responses</u> – Initial studies of the responses to in situ experimental warming of the SPRUCE chambers indicated that the peat microbial communities and decomposition rates were resistant to elevated temperatures in the first years of experimental warming (Wilson et al., 2016). A more recent in-depth analysis using combined evidence from metagenomics, proteomics and metabolomic analysis has shown that while abundance profiles have not changed, there has been a distinct shift in the microbial activity toward methanogenic metabolisms (Wilson et al. 2021) that is shifting deeper within the peat profile, with both temperature and CO<sub>2</sub> treatments. Analyses completed over the past year of metagenomes completed in collaboration with JGI from peat sampling in Aug 2018 show that across all years analyzed to date (2014, 2016 and 2018) microbial communities deeper in the peat profile are indeed changing in composition Fig T1.10. Approx. 10% of all the 800+ Metagenome Assembled Genomes (MAGs) recovered from the peat profiles show significant changes in relative abundance with time and treatment. These include most of the MAGs occurring in high abundance and across multiple depth layers.



Fig. T1.10 Non-Metric Multidimensional Scaling (NMDS) ordination of deep peat catolem communities analyzed collectively over three years of metagenomic datasets from 2015, 2016 and 2018.

Concurrently with the above metagenomic analyses, this past year we have been conducting collaborative analyses of overall microbial biomass and abundance profiles using QPCR approaches (ongoing - collaboration w/ M. Mayes) and microbial community patterns during peat decomposition ladder experiments using amplicon based analyses of rRNA genes (completed - w/ N. Griffiths and R. Kolka). In particular, decomposition community analyses show significant and strong effect of peat ladder depth, and more subtly (but significantly) in response to temperature and CO<sub>2</sub> treatment. Peat decomposition communities show decreased diversity with depth however within depths microbial communities shifted and have become more diverse under over the course of the three-year decomposition study. The deepest depth of the decomposition study (40cm) also shows an increase in the abundance of methanogenic archaeal lineages with temperature. Data are currently being compared both with mass loss, carbon/nitrogen content as well as with FTIR based characterization of organic matter remaining after the three year in-situ incubation.

<u>SPRUCE field activities and COVID-19</u> – Operations at SPRUCE are largely back to normal following the 2020 COVID-19 restrictions. The only remaining challenge for the research group is in the area of material acquisitions. Remaining post-COVID issues on instrument electronics supplies and deliveries is having an impact on the planned deployment of the automated, chamber-based flux systems acquired to better address high resolution system fluxes of  $CO_2$  and  $CH_4$ . We are actively working this issue are deploying instruments to treatment plots as soon as analyzers (ABB Los Gatos  $CO_2$ ,  $CH_4$ ,  $H_2O$  analyzers) as they are delivered.

## **SPRUCE Deliverable Progress**

The SPRUCE project has now completed 6 years of whole-ecosystem warming manipulations. Science measurement and modeling tasks represent the dominant effort. The following deliverables cover SPRUCE activities for FY2021 and beyond pending updates in the renewal proposal due February 2023.

Date	Deliverable	Status
September 2021	Publish manuscript on initial sap flow timing and seasonal	Overdue
	dynamics	
December 2021	Manuscript on nutrient responses to warming at SPRUCE	Published
		(Iversen et al. 2022)
October 2021	Publish manuscript on warming induced physiological stress	Overdue
October 2021	Publish manuscript on photosynthetic acclimation to SPRUCE	Overdue
	treatments	
December 2021	Publish manuscript on 5 years of non-structural carbohydrate	Overdue
	dynamics	
September 2022	Publish manuscript on woody plant water stress and hydraulic	Underway
	failure	

Task 1 – SPRUCE Deliverables

October 2022	Publish manuscript on tree photosynthetic acclimation to SPRUCE	Underway
	treatments	
December 2022	Publish manuscript on initial sap flow timing and seasonal	Underway
	dynamics	
Winter 2022	Develop techniques and measure branch and woody fine root	Planned
	respiration at multiple temperatures	
Summer 2023	Measure branch respiration from within SPRUCE enclosures, and	Planned
	woody fine root respiration from excised samples from outside	
	plots	
Summer 2024	Complete 2 <sup>nd</sup> field campaign to collect A-Ci curves to test	Planned
	respiratory and photosynthetic acclimation to temperature	
September 2022	Manuscript on root and fungal strategies in response to warming	Underway
		(Duchesneau et al.)
September 2022	Compete draft manuscript of the responses of labile carbon	Planned
	decomposition to warming.	
September 2023	Manuscript on the impacts of warming and elevated [CO <sub>2</sub> ] on	Planned
	belowground phenology and rooting depth distribution	(Weber et al.)
September 2023	Manuscript on N and P in peat, vegetation, and porewater	In progress
		(Salmon et al.)
September 2023	Complete draft manuscript on the responses of carbon and nutrient	Planned
	concentrations in outflow to the first 5 years of warming.	

### Task 1 SPRUCE Publications

- Baysinger MR, Wilson RM, Hanson PJ, Kostka JE and Chanton JP (2022) Compositional stability of peat in ecosystem-scale warming mesocosms. PLOS ONE 17:e0263994. https://doi.org/10.1371/journal.pone.0263994.
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- Iversen CM, Latimer J, Brice DJ, Childs J, Vander Stel HM, Defrenne CE, Graham J, Griffiths NA, Malhotra A, Norby RJ, Oleheiser KC, Phillips JR, Salmon VG, Sebestyen SD, Yang X, Hanson PJ (2022) Whole-ecosystem warming increases plant-available nitrogen and phosphorus in an ombrotrophic bog. *Ecosystems* (in press), https://doi.org/10.1007/s10021-022-00744-x.
- Kolton M, Weston DJ, Mayali X, Weber PK, McFarlane KJ, Pett-Ridge J, Somoza MM, Lietard J, Glass JB, Lilleskove EA, Shaw AJ, Tringe S, Hanson PJ, Kostka JE (2022) Defining the Sphagnum Core Microbiome across the North American Continent Reveals a Central Role for Diazotrophic Methanotrophs in the Nitrogen and Carbon Cycles of Boreal Peatland Ecosystems. *mBio* 13:e03714-21. https://doi.org/10.1128/mbio.03714-21.

- Ma S, Jiang L, Wilson RM, Chanton JP, Brigham S, Niu S, Iversen CM, Malhotra A, Jiang J, Lu X, Huang Y, Keller J, Xu X, Ricciuto DM, Hanson PJ, Luo Y (2022) Evaluating alternative ebullition models for predicting peatland methane emission and its pathways via data-model fusion. *Biogeosciences 19*(8):2245-2262.
- Ofiti N, Solly E. Hanson P, Malhotra A, Wiesenberg G, Schmidt M (2021) Warming and elevated CO2 promote rapid incorporation and accelerated degradation of plant-derived organic matter in an ombrotrophic peatland. *Global Change Biology* 28:883-898, doi: 10.1111/gcb.15955.
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  I. model structure and sensitivity analyses. *Journal of Geophysical Research -Biogeosciences* 126:e2019JG005468, https://doi.org/10.1029/2019JG005468.
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## Task 1 Data Sets

Five new Task 1 data sets have been prepared and posted (Baysinger et al. 2021D; Rush et al. 2021D; Schädel et al. 2021D; Wilson et al. 2021D all in Appendix B).

## Task 6: Coordinated Eco-physiology, Eddy covariance, and Sun-Induced chlorophyll Fluorescence Studies (CEESIFS) to advance integrative ecosystem science (formerly MOFLUX)

<u>MOFLUX site updates</u> – Field operations are busy at MOFLUX. In collaboration with TES-SFA Task 5, this summer we will be excavating roots (10 pits, to 1 m depth), and initiating a root and leaf litter decomposition study to provide new constraints on important belowground processes in support of modeling activities. We also have extensive plans for leaf level photosynthesis and chlorophyll fluorescence measurements to support photosynthesis modeling activities.

A technician (Mr. Brian Widmer) has just completed his first year of work on the project, and done an excellent job of developing new skills required of the position. MOFLUX Co-PI Dr. Jeff Wood has 2 MS students starting in the summer of 2022; both are funded on faculty startup funds and will be working on research projects that leverage MOFLUX's rich historical data sets.

<u>MOFLUX field activities and Covid-19</u> – We have been able to successfully maintain field activities with minimal disruptions owing to proximity to Columbia Missouri and are taking great care to maintain safety.

<u>CEESIFS science updates</u> – We have continued to integrate our unique observational, modeling, and technology innovation capabilities to develop deeper predictive understandings of photosynthesis and ecosystem responses to drought. Specific avenues of inquiry include (i) Coupling photophysics, photochemistry, and biochemistry for complete modeling and remote sensing of photosynthesis, (ii) coupling of photosynthesis with soil respiration (and its components), and (iii) ecosystem-scale hydraulics and water relations.

# Coupling photophysics, photochemistry, and biochemistry for complete modeling and remote sensing of photosynthesis

Photosynthesis consists of photophysical, photochemical, and biochemical reactions. These three stages of reactions are spatially separated, follow different laws, and operate at vastly contrasting time scales. The photosynthetic machinery uses feedforward and feedback interactions among these reactions to conduct a series of complex tasks to convert light energy from the sun to chemical bond energy in sugar for long-term storage and support of life on earth. Yet we still lack a broadly-applicable model that couples all three stages of reactions to predict photosynthesis. In our early efforts, we developed a model of photophysics of photosynthesis (Gu et al. 2019). In an attempt to develop a coupled model of photophysics, photochemistry, and biochemistry for complete modeling and remote sensing of photosynthesis, we have also developed a photochemical model for modeling the photosynthetic electron transport (PET) along the electron transport chain from photosystem II (PSII) to photosystem I (PSI) (Gu et al. 2022a). How redox reactions control photosynthetic electron transport (PET) is key to predictive understanding of PET regulation and linkage between light and carbon reactions. We lack a steady-state model of redox control of PET and are unable to determine redox conditions of key electron carriers and enzymes of PET in natural environments. To advance, we derived analytical steady-state equations governing the states and redox reactions of complexes and electron carriers along the PET chain. These equations, universal to oxygenic photosynthetic pathways, allow the redox conditions of the mobile plastoquinone pool and the cytochrome b6f complex (Cyt) to be inferred with typical fluorometry. We found the equations agreed well with fluorometry measurements from diverse C3/C4 species across



Fig.T6.1. Examples demonstrating the performance of the open-closed (OC) model of photochemistry for predicting the linear electron transport rate (JPSII) as a function of fraction of open PSII reaction centers (qL) with the lake connectivity of photosynthetic units for a variety of non-crop species. The thylakoid swelling is enabled. Measurements are either from light response only - systematic variation of light intensity at a fixed ambient CO<sub>2</sub> concentration (A, B, and C), or light response in conjunction with CO<sub>2</sub> response – systematic variation of ambient CO<sub>2</sub> concentration at a fixed light intensity (D, E, F, G, and H), or natural diurnal environmental variations (I). JPSII and qL generally vary in the opposite direction for light response but in the same direction for CO<sub>2</sub> response. Colors, circles, and × denote species, measurements, and model fits, respectively. All model fits have  $r^2 > 0.97$  and P < 0.001. (Gu et al. 2022a).

environments in the relationship between the PET rate and fraction of open photosystem II reaction centers, and the oxidation of plastoquinol by Cyt was the bottleneck of PET across species (Fig. T6.1). Redox reactions and photochemical and biochemical interactions are highly redundant in their complex controls of PET. Although individual reaction rate constants cannot be resolved, they appear in parameter groups which can be collectively inferred with fluorometry measurements for broad applications. Genetically improving the oxidation of plastoquinol by Cyt may enhance the efficiency of PET and photosynthesis across species. This photochemical model paves the way of linking the photochemical and biochemical models of photosynthesis.

The successful development of the photochemical model has led us to develop a theory to explain an enduring mystery with respect to granal thylakoid structure and function of higher plants (Gu et al. 2022b). In higher plants, PSII and PSI are found in grana stacks and unstacked stroma lamellae, respectively. To connect them, electron carriers negotiate multi-media tortuous paths, subject to macromolecular blocking. Why does evolution select a seemingly unnecessary, inefficient bipartition? We propose and test a theory to explain this perplexing phenomenon. Acting like bellows in accordions, grana stacks expand the volume of ultrastructural control on photosynthesis through thylakoid swelling/shrinking induced by osmotic water fluxes in coordination with varying stomatal conductance and turgor of guard cells which act like accordions' air buttons. The thylakoid ultrastructural dynamics regulates macromolecular blocking/collision probability, direct diffusional pathlength, duty division of cytochrome b6f complex, luminal pH via osmotic water fluxes, and separation of pH dynamics between granal and lamellar lumens in response to environmental variations. With the functionally asymmetrical PSII and PSI distantly located from each other, the ultrastructural control, non-photochemical quenching, and carbon-reaction feedbacks maximally cooperate to balance electron transport with gas exchange (Fig. T6.2), provide homeostasis in fluctuating light environments, and protect photosystems in drought. Grana stacks represent dry/high irradiance adaptation of photosynthetic machinery to improve fitness in challenging land environments. Our theory unifies many well-known phenomena of thylakoid structure and function of higher plants.



Fig. T6.2. Examples of variations of stomatal conductance with thylakoid swelling function inferred from PAM fluorometry measurements using the OC model. Each curve is marked with a number thatrepresents the species/cultivar: 1, *Bauhinia glauca*; 2, *Solanum lycopersicum*, tomato Basket Vee; 3 *Solanum lycopersicum*, tomato Growdena; 4, *Zea mays*; 5, *Bauhinia purpurea*; 6, *Oryza sativa*, rice IR64; 7, *Cornus racemosa* 'Ottzam'; 8, *Betula alleghaniensis*; 9, *Magnolia henryi*; 10, *Juglans nigra*; 11, *Dichanthelium clandestinum*; 12, *Sorghastrum nutans*. (Gu et al. 2022b)



Fig. T6.3. Seasonal variability of above- ( $R_{above}$ ) and belowground respiration ( $R_{below}$ ) expressed as daily totals for the MOFLUX site.  $R_{above}$ -DNS: estimated with daytime net ecosystem exchange of CO<sub>2</sub> (NEE) and soil respiration (DNS);  $R_{above}$ -NNS: estimated with nighttime NEE and soil respiration (NNS).

Partitioning of ecosystem respiration into above- and below-ground contributions: A three-way CO<sub>2</sub> flux-partitioning algorithm was developed to separate net ecosystem exchange (NEE) into aboveground plant respiration ( $R_{above}$ ), belowground root and soil respiration ( $R_{below}$ ), and gross primary production (GPP) (Liu et al. 2022). This algorithm was applied to a coupled long-term dataset of continuous chamber-measured soil respiration and eddy covariance (EC)-measured NEE of CO<sub>2</sub> at the MOFLUX site located in central Missouri. It was found that on annual time scale,  $R_{below}$  dominated over  $R_{above}$  with the former accounting for 67–86% and the latter 14–33%, respectively, of the total ecosystem respiration ( $R_{eco}$ ) (Fig. T6.3). The ratio of  $R_{below}$  to  $R_{above}$  varied seasonally, ranging from 2 to 7 in growing seasons, and 1 to 5 in non-growing seasons. The temperature sensitivity of  $R_{below}$  was significantly higher than that of  $R_{above}$  and  $R_{below}$  accounted for 21% and 69% of GPP, leaving only 10% to be permanently sequestered in the ecosystem. These results demonstrated the importance of separate accounting of below and aboveground processes in modeling ecosystem respiration and offered unique data for testing modeling predictions.

Ecosystem wilting point as a threshold for transitions in ecosystem functional state: We have further developed the concept of the ecosystem wilting point ( $\Psi_{EWP}$ ), that we previously derived by synthesizing ecosystem flux and predawn leaf water potential and invoking an analogy to the tissue-level pressure-volume (PV) technique. The top-down PV analysis yielded a whole-forest  $\Psi_{EWP}$  of -2.0 MPa. We have since conducted a bottom-up analysis of root density and soil moisture release data to derive a soils-based ecosystem wilting point ( $\Psi_{EWP,soil}$ ) of -2.0 MPa (Fig. T6.4), suggesting that capacity for soil water extraction determines  $\Psi_{EWP}$  and plants coordinate leaf turgor loss points accordingly. We also conducted analyses to determine whether GPP is limited by stomatal or non-stomatal processes during drought, retrieving parameters from models of stomatal conductance and C3 photosynthesis. We found that stomatal limitation dominated prior to ecosystem wilting, with non-stomatal limitations becoming important after wilting (Figure T6.5). These results are reported in a submitted manuscript.



Fig. T6.4. Estimation of ecosystem wilting point from soil moisture release characteristics and the root density distribution.



Fig. T6.5. Weekly estimates of A) apparent  $g_1$ (normalized water use efficiency, inversely related to intrinsic water use efficiency) and apparent V<sub>cmax</sub> during the 2012 growing season; significant quantile (=0.5) regressions are shown as solid lines with the 95% confidence interval of the regression shown as the shaded region; when regressions were not significant the mean  $\pm$  95% CI is shown as a dashed line. Before community predawn leaf water potential ( $\Psi_{pd}$ ) fell below the wilting point  $(\Psi_{\text{EWP}})$  for the first time (blue bars), apparent  $g_1$ decreased over time while apparent V<sub>cmax</sub> was stable, supporting stomatal limitation of GPP. During the wilted period (red bars) apparent  $V_{\text{cmax}}$ decreased strongly, indicating that non-stomatal limitation of GPP became important.

<u>Technology innovation and licensing</u>: The Fluorescence – Auto-Measurement Equipment (FAME), an invention developed under Task 6.1, has been granted patent protection by the United States Patent Office (Patent No. US 11,287,381 B2) on March 29, 2022 (<u>https://patentimages.storage.googleapis.com/bf/5d/e8/3761b318f2ff41/US11287381.pdf</u>). FAME, together with its controlling software Integrated Measurement And Control System for SIF (IMACSS)

has been licensed to Campbell Scientific, INC (https://www.ornl.gov/news/campbell-scientific-licenses-ornl-system-monitor-wide-scale-plant-health).

### Collaborative research activities -

We have collaborated on several multi-site papers that included MOFLUX data and were published in the last year (Benson et al. 2022; Novick et al. 2022a; Novick et al 2022b).

During June/July 2022 a team led by Dr. Lawren Sack (UCLA) is visiting MOFLUX to make detailed measurements of plant traits of tree species that make up ~90% of the basal area. This is part of an NSF macrosystems biology project, and MOFLUX is one of the few non-NEON sites to be included in the sampling plan. MOFLUX was selected because of the unique combination of rich historical data sets at a drought-prone deciduous forest site.

As part of collaborative activities funded by NASA, we anticipate deploying a new spectrometer on the MOFLUX tower in the growing season of 2022 to complement our FAME SIF system. This will enable retrieval of additional information concerning canopy function.

Date	Deliverable	Status
Sept 2021	Manuscript on NEE partitioning based on SIF	Complete
Feb 2022	Manuscript on three-way partitioning of NEE	Complete
March 2022	Physiological basis of using SIF to monitor	Complete
	photosynthesis (added/readjusted deliverable)	
April 2022	Inferring photosynthetic capacity with SIF (added	Complete
	/readjusted deliverable)	
May 2022	Submit 2021 MOFLUX data to AmeriFlux	Complete
July 2022	Manuscript on photochemical modeling of photosynthetic	On track
	electron transport	
Aug 2022	Submit manuscript on MOFLUX litter dynamics	On track
Oct 2022	Manuscript on why high plants have grana stacks	On track
Nov 2022	Manuscripts on ecosystem hydraulics/wilting point	On track
May 2023	Manuscript on coupled photophysical, photochemical, and	Planned
	biochemical modeling of photosynthesis	

#### **Task 6. Deliverables**

### Task 6 Publications/Manuscripts

- Benson MC, Miniat CF, Oishi AC, Denham SO, Domec JC, Johnson DM, Missik JE, Phillips RP, Wood JD, Novick KA (2022) The xylem of anisohydric *Quercus alba* L. is more vulnerable to embolism than isohydric codominants. *Plant, Cell & Environment* 45:329-346, DOI: 10.1111/pce.14244.
- Chang CY, Wen J, Han J, Kira O, LeVonne J, Yu L, Melkonian J, Riha SJ, Zhou R, Skovira J, Wang C, Shan X, Fan Y, Ng S, Gu L, Wood JD, Näthe P, Sun Y (2021) Unpacking the drivers of diurnal dynamics of sun-induced chlorophyll fluorescence (SIF): Canopy structure, plant physiology, instrument configuration and retrieval methods. *Remote Sensing of Environment* 265:112672.
- Gu L, Grodzinski B, Han J, Marie T, Zhang Y-J, Song YC, Sun Y (2022) Granal thylakoid structure and function: Explaining an enduring mystery of higher plants. *New Phytologist* (in review after first round of review).
- Gu L, Grodzinski B, Han J, Marie T, Zhang Y-J, Song YC, Sun Y (2022) Steady-state redox modeling of photosynthetic linear electron transport. New Phytologist (in review after first round of review).
- Han J, Chang CYY, Gu L, Zhang Y, Meeker EW, Magney TS, Walker AP, Wen J, Kira O, McNaull S, Sun Y. 2022. The physiological basis for estimating photosynthesis from Chla fluorescence. *New Phytologist* 234:1206–1219 doi: 10.1111/nph.18045.
- Han J, Gu L, Warren JM, Guha A, Mclennan DA, Zhang W, Zhang Y (2022) The roles of photochemical and non-photochemical quenching in regulating photosynthesis depend on the phases of fluctuating light conditions. *Tree Physiology* 42:848-861.

- Han J, Gu L, Wen J, Sun Y (2022) Inference of photosynthetic capacity parameters from chlorophyll a fluorescence is affected by redox state of PSII reaction centers. *Plant, Cell & Environment* 45:1298-1314.
- Liang J, Wang G, Singh S, Jagadamma S, Gu L, Schadt CW, Wood JD, Hanson PJ, Mayes MA (2021) Intensified soil moisture extremes decrease soil organic carbon decomposition: A mechanistic modeling analysis. *Journal of Geophysical Research: Biogeosciences* e2021JG006392.
- Liu X, Dong W, Wood JD, Wang Y, Li X, Zhang Y, Hu C, Gu L (2022) Aboveground and belowground contributions to ecosystem respiration in a temperate deciduous forest. *Agricultural and Forest Meteorology* 314:108807.
- Meng L, Zhou Y, Román MO, Stokes EC, Wang Z, Asrar GR, Mao J, Richardson AD, Gu L, Wang Y (2022) Artificial light at night: an underappreciated effect on phenology of deciduous woody plants. *PNAS Nexus*, 0:1–10. https://doi.org/10.1093/pnasnexus/pgac046.
- Novick KA, Ficklin DL, Baldocchi D, Davis KJ, Ghezzehei TA, Konings AG, MacBean N, Raoult N, Scott RL, Shi Y, Sulman BN, Wood JD (2022a) Confronting the water potential information gap. *Nature Geoscience* 15:158-164, https://doi.org/10.1038/s41561-022-00909-2.
- Novick K, Jo I, D'Orangeville L, Benson M, Fung Au T, Barnes M, Denham S, Fei S, Heilman K, Hwang T, Keyser T, Maxwell J, Miniat C, McLachlan J, Pederson N, Wang L, Wood JD, Phillips RP. (2022b) The drought response of eastern us oaks in the context of their declining abundance BioScience 72:333-346, https://doi.org/10.1093/biosci/biab135
- Yang Y, Anderson MC, Gao F, Wood J, Gu L & Hain C (2021) Studying drought-induced forest mortality using high spatiotemporal resolution evapotranspiration data from thermal satellite imaging. *Remote Sensing of Environment* 265:112640, https://doi.org/10.1016/j.rse.2021.112640.

### **CARBON-CYCLE FOCUSED WORK**

### Task 3abc: Mechanistic Carbon Cycle modeling

This task incorporates model development and MODEX activities at the point scales (Task 3a), regional to global scales (Task 3b), and at the level of mechanistic functional units (Task 3c) to identify process contributions to the global climate C cycle forcing from terrestrial ecosystems. Brief summaries of progress are presented along with tabular summaries of progress on proposed deliverables.

### Canopy processes

We investigated the potential of using a SIF time series product named contiguous SIF (CSIF) to estimate spring, summer, and autumn phenology in the extratropical Northern Hemisphere (>30°N) and compared the results with those based on Moderate Resolution Imaging Spectroradiometer (MODIS) Normalized Difference Vegetation Index (NDVI) for the period 2001-2018. Overall, we found similar spatial patterns in phenological states; however, specific dates of key phenological events differed when using CSIF vs. MODIS NDVI data (Fig. 3abc.1); and NDVI data indicated that the growing season started earlier (by 10.1 days on average) and ended later (11.5 days on average) relative to CSIF data. This implies that actual periods for photosynthetic activity are shorter (by 21.6 days on average) than those estimated from vegetation indices more directly related to changes in canopy structure. These large differences between results from NDVI and that from CSIF suggest that vegetation indices such as NDVI seem to overestimate the period for active photosynthesis over the extratropical Northern Hemisphere. We conclude that SIF data offers a novel and unique approach for assessing phenological change - one that is more directly tied to the carbon cycle and how it is being influenced by climate change. Relevant work is detailed by Chen et al. (2022a), and model evaluations using these phenological metrics are ongoing.

## Phenology Modeling

We evaluated and improved the above-ground evergreen phenology of ELM using the SPRUCE observations. We tested explicit green-up controlled by temperature and degree-day thresholds in spring and changed the default constant litterfall to intensive offset in autumn via an exponential function. The results show that the new models reduced the uncertainties of start of season (SOS) and end of season

(EOS) compared to the default models; moreover, the new phenological schemes improved the temperature responses of SOS and EOS. Since root growth in boreal regions generally span longer time periods than shoot growth, we fitted SPRUCE minirhizotron-observed root growth data, and soil temperature and moisture data, and made different assumptions on the root onset and offset. For example, for the root onset, length of the onset period is a function of time, all year (365 days) or approximately until summer solstice (171 days); for the onset controlling factors, we had different assumptions related to the control by soil temperature or soil water. For the root offset, it's currently represented using the same scheme as evergreen leaf, meaning the intensive offset in autumn is via an exponential function. Compared to the limited root observations, the revised models can at least reproduce the magnitude of SOS and SOS responses to warming; however, for EOS, we still need more data for validation. The model development is still ongoing and relevant manuscript is under preparation.

### <u>Disturbance</u>

Natural and anthropogenic driving mechanisms underlying the changes of peat fire remain to be explored. We investigated major affecting factors and predictability of peatland fires for the 1997-2016 period using multi-source environmental datasets and the two-step-correcting machine learning framework, a combination of multiple machine learning classifiers, regression models, and a self-correcting technique. We found 1) the oversampling algorithm worked for the unbalanced data and improved the recall rate by 26.88% to 48.62%; 2) the Random Forest performed best in simulating the peatland fire counts with the Area Under the Receiver Operating Characteristic Curve value ranging from 0.83 to 0.93 across multiple fire datasets; 3) temperature, air dryness, seasonality, and frost day frequency dominated the peat fires, overriding the impacts of biomass, soil moisture and human activities; and 4) the seasonality and frost day frequency was further identified as the critical factor that could change the physical characteristics and thermal hydrology in peatland, thus favoring peat fire occurrences. Relevant manuscript (Tang et al., 2022) is finished and will be submitted soon.



Fig. 3abc.1 Vegetation phenology of different plant functional types. The phenological sequences from left to right are SOS, POS, EOS, SOP, POE and GS. ENF: Evergreen Needleleaf Forests; EBF: Evergreen Broadleaf Forests; DNF: Deciduous Needleleaf Forests; DBF: Deciduous Broadleaf Forests; MF: Mixed Forests; OS: Open Shrublands; WS: Woody Savannas; SA: Savannas; GRA: Grasslands.

### SPRUCE CH4 and hydrology modeling

A new paper using the ELM-SPRUCE model has been published describing the hydrological feedbacks on peatland CH<sub>4</sub> emissions under the SPRUCE treatments (Yuan et al., 2021). This study found that the water table is a critical regulator of hydrological feedbacks. Although warming significantly stimulates CH<sub>4</sub> emission, reduced water table levels from increased evapotranspiration mitigates the warming effect. Further work continues to evaluate the ability of ELM-SPRUCE to predict CH<sub>4</sub>

emissions during the 2021 drought, when observations from the large collars indicated a sharp reduction. Improving the predictions of water table height in ELM-SPRUCE is also a high priority for the upcoming year. Another methane model, TECO-SPRUCE, was also used to simulate the effects of the treatments at SPRUCE. An improved approach to simulating ebullition was integrated into TECO-SPRUCE, and the authors found that improved predictions of porewater CH<sub>4</sub> concentrations (Ma et al., 2022). A third study examined the sensitivities of ELM-SPRUCE carbon and hydrology outputs to uncertainties in microtopography parameters including hummock height, horizontal separation, and hollow fraction (Graham et al., 2022). These parameter distributions were informed by terrestrial laser scanning (TLS) observations within the enclosures. The model experiment showed that the model outputs were typically most sensitive to hummock height, and that net ecosystem exchange (NEE) was overall the most sensitive model output to the microtopography parameters. Hummock height also influenced the partitioning of carbon into above and belowground pools. These results will be useful in applying our ELM-SPRUCE model to other peatlands where these microtopography parameters may be different.

### Model-data integration

We developed an invertible neural network (INN) to calibrate ELM with observed flux data from the Missouri Ozark eddy covariance site. The INN in this example is trained on 1000 simulations of ELM with different values of 7 parameters related to allocation, leaf properties and phenology. The INN can efficiently address both forward and inverse modeling simultaneously; in inverse mode, it can perform model calibration by producing posterior distributions for the ELM parameters, while in forward mode, the INN generates ELM predictions of a model output of interest, for example latent heat flux. In past studies, we have used Markov Chain Monte Carlo (MCMC) to perform the model calibration step. However, the INN is much more computationally efficient, producing similar posterior estimates of latent heat flux as the MCMC method but runs 30 times faster. The INN may be used to quickly evaluate the role of new measurements or reduced measurement uncertainty in reducing model prediction uncertainty, potentially speeding up the MODEX cycle at our study sites.



Fig. 3abc.2 Box plots summarizing latent heat (LH) predictions at MOFlux from the parameter posterior samples of INN and MCMC; MAP\_INN represents the prediction from the maximum a posteriori (MAP) estimate of INN and MAP\_MCMC represents the prediction from the MAP estimate of MCMC.

Date	Deliverable	Status
2021	Complete ELM forcing factor simulations and publish results	Complete
2021	Publish results of improved ELM-SPRUCE model	Complete (Shi et al., 2021; Ricciuto et al., 2021; Yuan et al., 2022)
2021	Integration of new model features into E3SM code base	Ongoing
2021	Publish multi-model SPRUCE intercomparison	Underway (Hou et al., submitted)

2022	Enable data assimilation capability with ELM-SPRUCE using surrogate modeling approach	Complete
2022	Publish manuscript on using EcoPAD for SPRUCE intercomparison	Underway
2022	Develop root function, nutrient cycling, disturbance functional units/surrogates	Underway
2022	Regional benchmarking of ELM including new improvements	Underway
2022	Complete manuscript on ELM fire evaluation	Underway

## Task 3abc Publications

Chen A, Meng F, Mao J, Ricciuto D, Knapp A (2022a) Photosynthesis phenology, as defined by solarinduced chlorophyll fluorescence, is overestimated by vegetation indices in the extratropical Northern Hemisphere. *Agricultural and Forest Meteorology* 

323:109027, https://doi.org/10.1016/j.agrformet.2022.109027

- Chen A, Ricciuto D, Mao J, Wang J, Lu D, Meng F (2022b) Improving E3SM land model photosynthesis parameterization via satellite SIF, machine learning, and surrogate modeling. *Journal of Advances in Modeling Earth Systems (in review)*.
- Graham JD, Ricciuto DM, Glenn NF, Hanson PJ (2022) Incorporating microtopography in a land surface model and quantifying the effect on the carbon cycle. *Journal of Advances in Modeling Earth Systems* 14:e2021MS002721. https://doi.org/10.1029/2021MS002721.
- Lu, D., D. Ricciuto, J. Zhang (2022) Invertible neural networks for E3SM land model calibration and simulation. *International Conference on Learning Representations* (virtual).
- Ma S, Jiang L, Wilson RM, Chanton JP, Brigham S, Niu S, Iversen CM, Malhotra A, Jiang J, Lu X, Huang Y, Keller J, Xu X, Ricciuto DM, Hanson PJ, Luo Y (2022) Evaluating alternative ebullition models for predicting peatland methane emission and its pathways via data-model fusion. *Biogeosciences 19*:2245-2262.
- Meng L, Zhou Y, Román MO, Stokes EC, Wang Z, Asrar GR, Mao J, Richardson AD, Gu L, Wang Y. (2022) Artificial light at night: an underappreciated effect on phenology of deciduous woody plants. *PNAS Nexus* 0:1–10. https://doi.org/10.1093/pnasnexus/pgac046.
- Tang R, Mao J, Jin M, Ricciuto D (2022) Tackling key drivers and predicting fires in boreal peatland with a two-step-correcting machine learning framework. (Finished and to be submitted).

### Task 3d: Multi-Assumption Systems Modeling

Hundreds of models have been developed to describe the formation and decomposition of soil organic carbon (SOC), with recent models employing ever more complicated representations of microbial and mineral interactions. Unsurprisingly, this has resulted in an explosion of potential model configurations with different models making divergent predictions about SOC responses to global change. The Multi-Assumption Architecture & Testbed (MAAT; Walker et al., 2018) is an open source-software that was developed during the previous phase of the TES-SFA as a tool for formal and informal evaluation of alternative model process representations. In this current phase, Task 3d is primarily focused around understanding how alternative process hypotheses lead to variable model predictions in below-ground processes.

To this end, we have developed a multi-assumption soil carbon modelling tool. In FY22, we have continued developing this tool, which will allow users to explore the sources of process-level uncertainty among contemporary soil C models. Our framework and a first version of the multi-assumption soil C model was described in Craig et al. (2021) and in last year's report. Since last year we have updated four contemporary soil C models, many from the DOE sphere: (MEND, Wang et al., 2013; MIMICS, Wieder et al., 2014; MILLENIAL, Abramoff et al., 2022; and CORPSE, Sulman et al., 2014) – added a conventional model as a point of comparison (CENTURY; Parton et al. 1987), added numerical solvers to speed up steady-state simulations, and we are currently implementing a sixth model, RESOM (Tang and Riley 2015).

These models make different predictions about a key question in soil biogeochemistry: how do the rate and quality of organic inputs affect soil C? For example, model predictions about how soil C should change with increasing input rates range from a strong linear increase (CENTURY and CORPSE) to complete insensitivity (MEND; Fig 1a). We are using MAAT to probe the structural sources of this uncertainty. For example, there is some disagreement among models about the extent to which microbial decomposers can access mineral-associated forms of soil C. Though this pool is often viewed as highly resistant to microbial attack, Matt Craig (scientist on Task 3d) recently published empirical results suggesting that microbial decay of this pool can be stimulated by organic inputs (Fig. 2; Craig et al. 2022). Using MAAT, we unified the representation of this process among the four contemporary models, allowing microbes to access mineral-associated C. We also unified the representation of microbial turnover and mineral saturation as described in last year's report and in Craig et al. (2021). We find that these microbial-mineral interactions account for a substantial portion of the inter-model variability (Fig. 1b). We plan to submit a manuscript describing this work in the coming year.

Collaborations with the Global Carbon Project to provide multiple assumptions to their model ensemble of the global carbon budget are ongoing. This has resulted in three co-authorship publications, two that evaluate global carbon cycle models in drylands ecosystems in the southwestern US (MacBean et al., 2021) and the Australian continent (Teckentrup et al., 2021). Both analyses found that models simulate a variety of dryland ecosystem functions poorly, generally worse than many other ecosystems, and provide ideas for how simulations may be improved. The other analyzes how net land-use change emissions calculated with global carbon cycle models differs from calculations from bookkeeping models (Obermeier et al., 2021). The analysis focused on the "loss of additional sink" capacity that arises from reduced capacity of modified ecosystems to respond to global change, primarily increasing CO<sub>2</sub>, finding that since 1850,  $40\pm15$  Pg of carbon remains in the atmosphere due to the loss of additional sink capacity from land use change.



Fig. T3d.1 Simulated response of total soil carbon at equilibrium to altered organic input rates for five models in (a) their default configuration and (b) after unifying the representation of three processes (mineral-associated C decay, mineral saturation, and microbial turnover) among the four microbially explicit contemporary models (CORPSE, MIMICS, MILLENNIAL, and MEND).



Fig. T3d.2 Path analysis results from Craig et al. (2022) showing that stimulation of microbial activity by high quality inputs drives decomposition of mineral associated soil C (MA-SOC). "Microbial physiological traits" is an index representing the combined effects of microbial growth, carbon use efficiency, and turnover. Numbers above and below paths represent standardized coefficients during early- and intermediate-stage decomposition, respectively, with significance levels indicated (\*p < 0.1, \*\*p < 0.05, and \*\*\*p < 0.01). Thickness and color of lines correspond to coefficient magnitude and sign, respectively.

	Г	ask	3d	Deliverables	
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Date	Deliverables	Status
April 2019	1. State-of-the-art MCMC routines enabled in MAAT	Complete
December 2021	2. Parameter estimation and hypothesis testing in SPRUCE Sphagnum photosynthesis and respiration data 2014-2018	On hold
April 2020	3. pbdr enabled MAAT to run on OLCF	In progress
April 2020	4. Multi-assumption soil decomposition model	Near complete
September 2021	5. Multi-assumption root resource acquisition model	On hold
April 2021	6. Structural and parametric uncertainty quantification of soil decomposition model, *see below Craig et al 2021	Complete
Sept 2021	7. Structural and parametric uncertainty quantification of root nutrient acquisition and nutrient competition models	Reconfigur ed

## Task 3d Publications/Manuscripts

- Craig ME, Geyer KM, Beidler KV, Brzostek ER, Frey SD, Stuart Grandy A, Liang C, Phillips RP (2022). Fast-decaying plant litter enhances soil carbon in temperate forests but not through microbial physiological traits. *Nature Communications*, *13*:1–10. https://doi.org/10.1038/s41467-022-28715-9
- Craig ME, Brzostek ER, Geyer KM, Liang C, Phillips RP (2021D): Data for "Fast-decaying plant litter enhances soil carbon in temperate forests, but not through microbial physiological traits". The Carbon-Nutrient Economy of the Rhizosphere: Improving Biogeochemical Prediction and Scaling Feedbacks From Ecosystem to Regional Scales, ESS-DIVE repository. Dataset. doi:10.15485/1835182
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## **PROCESS-LEVEL STUDIES (PLS)**

### **Task 4c: Linking Root Traits to Function**

The root function task was developed to improve understanding of root function and implications for modeling. Our research focuses on assessing root function in situ, including a focus on the root rhizosphere, where there are dynamic interactions and exchanges between roots, soil pores and soil surfaces. Of particular interest is how hydraulic conductivity from the bulk soil to the root changes as soils dry and if there is a loss of preferential pathways, or development of air gaps that prevent water uptake. Such data are critical for modeling water availability to plants, especially in process-based models

such as FATES. We also continue to be interested in linking root traits to root function, especially through root, mycorrhizal and soil respiration rates.



Fig. T4c.1 Testing ultrafast neutron computed tomography and machine learning in July 2021 using maize with deuterium injections. Over 160,000 images were collected to assess capability to measure threedimensional soil-root water dynamics in situ (1.8 terabytes of data). Initial results suggest a 90 second run with 0.1 s image exposure may be an optimal setting. Our earlier research with NCT collected images over multiple hours, precluding understanding of short-term dynamics.

We continue to leverage the unique capabilities of the neutron imaging facilities at ORNL, including a new focus on ultrafast computed tomography of water dynamics in the soil and uptake by roots (Fig T4c.1). We also have continued building on our earlier analyses that linked water uptake rates to root traits (such as diameter or order (Dhiman et al., 2019, DeCarlo et al. under revision). Subsequent modeling of these data revealed that roots and mycorrhizae can affect soil hydraulic properties, which we measured in a follow-up study (Fig. T4c.2; Marcacci et al. 2019), and which have a direct impact on ELM and other process-based modeling efforts. Our involvement with neutron imaging continues to evolve, including development of a comprehensive review manuscript draft of the use of neutron imaging in Geosciences. We have also been hosting workshops focused on how BER research interests might addressed using neutrons. In June, 2022, Jeff Warren hosted one exciting session on future application of neutrons to soil, rhizosphere processes and plant-soil interactions. Results will be used to inform development of future beamlines, e.g., CUPI2D (Brügger et al. 2022) that could enable unprecedented insight in soil, rhizosphere and root function in situ.



Fig. T4c.2 (A) Based on earlier neutron imaging work, we determined that root-free hydraulic parameters do not fit the data (Dhiman et al. 2019). Here we illustrate how one soil hydraulic parameter (saturated water content) varies from soil only (CON), soil with living switchgrass roots (RTS) and soil with roots or mycorrhizal hyphae (MYC) (Marcacci et al. 2022).

We continue to focus on assessing linkages between root traits, mycorrhizae and root respiratory (Rs) response to environmental dynamics in order to improve autotrophic vs heterotrophic Rs modeling. In collaboration with the Root Traits Task, and to provide additional physiology data to FRED, we have established a relationship with The Morton Arboretum to assess root, hyphal and soil respiration dynamics for tree species with divergent root traits. 108 root/fungal exclusion collars were built, installed and are currently being measured in different monoculture stands at Morton. In a complementary study

funded by the DOE SCGSR program, we focused on assessing root traits and their linkages to seasonal respiration *in situ* using a novel measurement system that assessed a single 3<sup>rd</sup> or 4<sup>th</sup> order root system while attached to mature temperate tree species. This manuscript received good reviews at Geoderma and has been revised and resubmitted (Hogan et al. under review).

Results will provide novel root functional data for models that increasingly consider root function. This effort has synergy with MAAT, MOFLUX and NIST research efforts.

Date	Deliverable	Status
July 2021	Publish manuscript on above/belowground physiological acclimation to	Completed
	warming in Populus trichocarpa	
July 2021	Use neutron beamtime to further develop analysis techniques for assessing	Completed
	root function <i>in situ</i> using fast neutron imaging	
August 2021	Installation of root/hyphal separation chambers at Morton Arboretum	Completed
June 2022	Publication on root and mycorrhizal impacts on soil hydraulic properties	Completed
August 2022	DOE-SCGSR PhD student-led publication of seasonal patterns of in situ	Underway
	respiration of intact root systems with divergent traits and simultaneous	
	publication of completed dataset	
December 2022	Develop comprehensive review article on neutron imaging in the	Underway
	Geosciences (including root, rhizosphere and soil dynamics)	
December 2022	Complete first year of root/hyphal respiration at Morton Arboretum,	Underway
	develop manuscript framework	
December 2022	Analysis of data and manuscript draft of root function in situ using fast	Planned
	neutron imaging	
Spring 2023	Revise and resubmit manuscript on root rhizosphere water dynamics based	Planned
	on neutron imaging and automated image analysis	
Spring 2023	Develop and test measurement techniques of root respiration of woody root	Planned
	systems at SPRUCE (outside plots)	

**Task 4c. Deliverables** 

### Task 4c New Datasets and Publications

- Brügger A, Bilheux HZ, Warren J et al. (2022) CUPI<sup>2</sup>D: Complex, Unique and Powerful Imaging Instrument for Dynamics. *Review of Scientific Instruments* (in review)
- DeCarlo KF, Warren JM, Caylor K, Bilheux H (2022) Integrating fine root morphology and soil distance mapping to characterize the plant-soil interface. (in revision).
- Hogan JA, Labbé JL, Carell AA, Franklin J, Hoyt KP, Valverde-Barrantes OJ, Baraloto C, Warren JM (2022D) 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
- Hogan JA, Labbé JL, Carrell AA, Franklin J, Hoyt KP, Valverde-Barrantes OJ, Baraloto C, JM. Warren JM (2021) Functional variability in specific root respiration translates to differences in belowground CO<sub>2</sub> efflux in a temperate deciduous forest. *Geoderma (*revised and under review)
- Marcacci KM, Warren JM, Perfect E, Labbe, JJ (2022) Influence of living grass roots and endophytic fungal hyphae on soil hydraulic properties. *Rhizosphere* 22:1-13. https://doi.org/10.1016/j.rhisph.2022.100510

### Task 5: Microbial Processing of Soil C

Experimental and modelling studies continued to focus on the key role of microbial activity in influencing  $CO_2$  emissions and soil carbon cycling, and in particular, to understand the role of soil moisture which has been neglected in the literature in comparison to temperature.

We continue to work to understand connections between below-ground microbial respiration and canopy photosynthesis at MOFLUX (Wood et al. under preparation) in association with Task 6. We continued quarterly soil core sampling and analysis for soil moisture, microbial biomass C and N, total C and N, pH, texture, and root length and density. We are actively collaborating with Jeff Wood at MU in planning a pedon sampling activity at MOFLUX. We will conduct similar analyses as above from the soil

pits for the project, and we have also helped design a root litter decomposition experiment to begin summer 2022. We will also assist with soil analyses on the decomposition activity.

In 2017, four LiCor soil chambers were trenched to isolate heterotrophic and total respiration measurements under Task 6. Using 17 years of soil respiration data (2004-2021) of which 4 years (2017-2021) include trenched measurements to isolate heterotrophic respiration, we applied artificial intelligence and wavelet coherence analysis to examine the effects of environmental factors and their timescales on soil respiration and its partitioning. Random Forest machine learning models were used to identify the relative importance of different environmental drivers of heterotrophic and autotrophic respiration, including soil temperature, soil moisture, LAI, and date and time information. Wavelet coherence analysis was used to identify the timescales (e.g., daily, weekly, monthly, or seasonal) of importance for key drivers and whether they are in-phase or out-of-phase. Key results are (1) heterotrophic respiration is most responsive to soil temperature at daily and seasonal timescales, while autotrophic respiration is most responsive to above ground productivity (using LAI as a proxy) and the time of the year; (2) Heterotrophic and autotrophic respiration have similar responses to temperature but not to LAI or time of year, and as a result, the time of year and LAI are most influential for determining the partitioning between heterotrophic and autotrophic respiration (R<sup>2</sup>=0.67), suggesting that the partitioning between heterotrophic and autotrophic respiration can be inferred from these variables; (3) Soil moisture is most important to soil respiration on synoptic weekly-to-monthly timescales; and (4) Soil temperature effects are generally in-phase (Fig. T5-1), but soil moisture effects are sometimes out of phase, depending on the event (Fig. T5-2).



Fig. T5.1 Wavelet coherence analysis on 3-hourly measurements of soil temperature (°C) and heterotrophic (left) and autotrophic (right) respiration. Color bar indicates the strength of coherence between temperature and respiration. The red band which occurs at Scale = 8 represents the daily timescale (8\*3-hourly measurements = 24 hourly), indicating that there is strong coherence between soil respiration and temperature at daily timescales for both autotrophic and heterotrophic respiration.



Fig. T5.2 Wavelet coherence analysis on 3-hourly measurements of soil moisture (m<sup>3</sup> m<sup>-3</sup>) and heterotrophic (left) and autotrophic (right) respiration. Color bar indicates the strength of coherence between temperature and respiration.

In collaboration with Task 3d (Multi-Assumption Architecture and Testing; MAAT), we have set up the MAAT modeling platform to run multiple models at MOFLUX in order to test different model formulations of soil moisture and carbon cycle interactions at the site. We aim to understand the effects of seasonal drought on plant root and microbial activity and to use model constrained with long-term measurements to project C responses and their uncertainty to future climate. We have completed a run on MOFLUX data with the Millennial model, which was originally published by Abramoff et al. (2022).

We completed the microbial biomass C and N analyses from chloroform fumigation extraction (CFE) on soils from two coring activities at SPRUCE (June and August 2021), and have nearly completed the qPCR analyses. There is one additional planned coring activity at SPRUCE (June 20 2022). We are working with Jessica Gutknect at the University of Minnesota to compare our results to those using PLFA, and we are planning to hire a summer M.S. student with expertise in PLFA data. The student will analyze PLFA data at UMN, and they will compare their findings with those from ORNL's qPCR and CFE analyses. Task 5 has an existing dataset from different soil types comparing the 3 kinds of microbial analyses (PLFA, CFE, qPCR) and we initially found a high degree of correlation between the different soil types, with one exception—organic soils. The SPRUCE samples are also organic samples and will complete the pre-existing dataset. Quantifying a correlation between the datasets would greatly expand the kinds of microbial data available for modeling the SPRUCE site, and would be an important contribution in the literature by providing a validated relation for different kinds of simple microbial analyses.

Dr. Singh defended her PhD at the University of Tennessee in December 2020 and she was supported by Task 5 throughout her dissertation work. One paper remains to be published, where she investigated the "Birch effect". This spike in respiration is thought to be attributed to: (1) release of cellular solutes (metabolites) accumulated during drought due to rapid increase in water potential upon re-wetting; (2) sudden death of certain microbes serving as C sources for surviving microbes, leading to microbial community shifts; and (3) release of physically-protected C due to aggregate breakdown upon repeated drying-wetting. The relative importance of these mechanisms may change in different soil textures. In this study, the effects of repeated drying and wetting cycles (transient state moisture conditions) on Birch effect were evaluated to elucidate the underlying mechanisms in different textured soils in comparison to a steady state moisture condition at 55% water holding capacity (WHC). Soils of three distinct textures (sandy, loamy, and clayey) were incubated for 140 days under five alternate cycles of drying (10% WHC) and wetting (100% WHC) conditions. Soil respiration was measured periodically, and destructive samplings occurred over time. Microbial biomass C, extractable organic C, metabolites, microbial community structure, and changes in aggregate-associated C were determined at each time point. Results showed that soil texture strongly influenced respiration rates throughout the incubation as among the soils, and sandy soil showed the lowest respiration rate followed by loamy and clayey soils. Also, cumulative CO<sub>2</sub> loss was higher under transient moisture state compared to steady state. Our results also conclusively showed that the relative contribution of various mechanisms to Birch effect were different in different soil types. In sandy and clayey soils, changes occurred in bacterial abundance while in loamy soil, the release of aggregate protected C controlled the Birch effect. Deciphering the mechanisms underpinning increased soil organic C turnover from different soils upon wetting and drying is of interest to improve the understanding of terrestrial C cycling in response to extreme events.

Dr. Siyang Jian defended his PhD at Tennessee State University (note, TSU is HBCU/MEI) in July of 2020, and he was also supported by Task 5 throughout his dissertation work. His last paper explores the extent to which model microbial parameters from ORNL incubation experiments (dataset: Kluber et al. 2020) can be generalized across multiple sites. Initial results suggest that model parameters are highly site-specific if only the 5 major microbial parameters controlling microbial uptake, growth and maintenance are optimized in model fitting. However, if microbial parameters relevant to enzyme production and turnover are optimized as well, the model parameters can be generalized based on soil-series regardless of plantation or experimental treatments within the incubation study—2 each paired grassland and forest soils, involving Alfisols from Missouri and Ohio, an Ultisol from Tennessee, and a Mollisol from Iowa. This suggests that a common parameterization for microbial carbon cycle models are applicable to a diversity of ecosystems and environments based on soil type, which will aid in the broader application of microbial models.

<u>Future Plans</u> - We are implementing a modeling strategy for the MOFLUX site and are currently working on data analysis and early modeling. We will complete the plan for seasonal sampling of different measures of microbial biomass at SPRUCE to complement and enhance existing site data, and to relate our results to soils from around the globe. We have established a strong collaboration with the microbiology task in SPRUCE and the University of Minnesota to compare microbial data using qPCR, chloroform fumigation, and PLFA. We will prepare an initial data publication so that our results are available for the modeling team. We have also become involved with the SPRUCE site – assisting with the installation of the new ABB greenhouse gas analyzers and fluxchambers. This equipment was recently provided through a supplement from BER, and new analyzers continue to arrive. The postdoctoral research associate is not yet on board, but the team has taken three trips to SPRUCE to begin installation and establish procedures.

Date	Deliverable	Status
2022	Understanding of depth and spatial distribution of simple microbial metrics for	In progress
	modeling at SPRUCE	
2023	Machine learning and data analysis of soil respiration at MOFLUX	In progress
2023	Development and initial offline testing of microbial models at MOFLUX	In progress
September 2023	A plan for applying microbial modeling at SPRUCE	09/23

### Task 5. Deliverable Status FY2021 and FY2022

### Task 5 Publications/Manuscripts

Singh S, Jagadamma S, Kivlin S, Mayes MA Soil organic carbon mineralization under transient and steady state moisture conditions in different soil types. (in prep)

Wood JD, Mayes MA, Gu L Environmental and photosynthetic regulation of soil heterotrophic and autotrophic respiration. (in prep)

Zhang H, Deng Q, Schadt CW, Mayes MA, Zhang D, Hui D (2021) Precipitation and nitrogen application stimulate soil nitrous oxide emission. *Nutr Cycl Agroecosys* <u>https://doi.org/10.1007/s10705-021-10155-4</u>.

## Task 5 Datasets

Liang J, Wang G, Singh S, Jagadamma S, Gu L, Schadt CW, Wood JD, Hanson PJ, Mayes MA (2021D) MOFLUX Intensified Soil Moisture Extremes Decrease Soil Organic Carbon
 Decomposition: Modeling Archive. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/ornlsfa.023/1804106

## GLOBAL TRAIT DATABASES AND ANALYSIS TOOLS (GT)

## Task 4b: Leveraging root traits to inform terrestrial biosphere models

<u>The Fine-Root Ecology Database</u> – FRED has been and will continue to be a freely available resource for the broader community of root and rhizosphere ecologists and terrestrial biosphere modelers. Building on the recent release of FRED 3.0 (Iversen and McCormack 2021, *New Phytologist*) and our work to understand fine-root trait variation across the globe (data citations: Guerrero-Ramirez et al. 2021; Iversen et al. 2016; Iversen et al. 2018; Iversen et al. 2021, and papers: Bergmann et al. 2020, *Science Advances*; Guerrero-Ramirez et al. 2021, *Global Ecology and Biogeography;* Freschet et al. 2021a and b, *New Phytologist*; Iversen et al. 2017, *New Phytologist*; McCormack et al. 2018, *Eos*; Laughlin et al. 2021, *Nature Ecology & Evolution;* McCormack and Iversen 2019, *Frontiers in Plant Science*; Weigelt et al. 2021, *New Phytologist*), we are focused on asking four main questions:

1. How can we leverage data in FRED to improve our understanding of root trait variation across the globe?

Dr. Yao Liu, an ORNL post-doc co-advised by Drs. Anthony Walker and Colleen Iversen, accepted a position as a senior lecturer at Northumbria University in the UK in September 2020, but continues to complete a manuscript on a hierarchical Bayesian assessment of the variation in root traits in FRED in relation to environmental conditions, phylogeny, and mycorrhizal symbiosis (Liu et al., *in preparation*). Furthermore, a number of groups have also used FRED as a resource to answer their own scientific questions (https://roots.ornl.gov/publications).

The FRED team has continued to be involved in international collaborations (https://roots.ornl.gov/synthesis-activities). For example, the sROOT working group within the German Centre for Integrative Biodiversity Research (iDiv) from 2018 to 2020 leveraged FRED 2.0 to develop a 'Ready to Use' database that is a species-specific subset of FRED and TRY (Guerrero-Ramirez *et al.* 2021, *Global Ecology and Biogeography*, data citation: Guerrero-Ramirez *et al.* 2021), resulting a series of papers focused on the global variation in fine-root traits within the multidimensional Root Economics Space (Bergmann et al. 2020, *Science Advances*) and along environmental gradients (Laughlin et al. 2021, *Nature Ecology & Evolution*), and within and among above- and belowground plant traits (Weigelt et al. 2021, *New Phytologist* Tansley Review). This collaboration continues as 'RootFUN', focused on the links between root traits and ecosystem function (Barry et al., *in preparation*).

2. What root trait data are missing from FRED (e.g., root traits from the tundra and the tropics) and what additional data synthesis activities or data harvest from the literature are needed to fill these gaps?

The root trait framework developed by the FRED team has also been leveraged to develop a 'Root Traits Handbook' and companion paper advocating for increased quantification of functional root traits (Freschet et al. 2021a and b). FRED is also providing the foundation for new, biome-specific working groups in the Arctic (the 'Arctic Underground', PIs Hewitt and Mack) and the tropics ('Tropical Forest Root Traits', PI Cusack).

We released the third version of the Fine-Root Ecology Database (FRED 3.0) in March 2021 (data citation: Iversen *et al.* 2021), and data have been filtered and downloaded via the new user interface at <u>https://roots.ornl.gov/public-release</u> nearly 300 times through June 15 2022. FRED 3.0 has more than 150,000 observations of more than 330 root traits, with data collected from more than 1400 data sources. FRED is focused on fine roots (roots traditionally defined as less than 2 mm in diameter) but accepts data collected from roots of all sizes.

We highlighted FRED 3.0 as a community resource for belowground ecologists and modelers alike in an editorial that accompanies a Virtual Special Issue in *New Phytologist*, where we compiled more than 40 recent papers on the topic 'Filling gaps in our understanding of belowground plant traits across the world' (Iversen and McCormack 2021, *New Phytologist*).

**3.** What new measurements are needed to understand root trait variation within and among plant functional types?

The data in FRED are necessarily a mixture of root traits and ancillary data, quantified using a variety of methodologies. These observations need to be placed into a comprehensive framework that will allow us to more effectively use the diversity of traits in FRED to understand above- and belowground linkages across the globe. To advance this framework, we are making new observations across a phylogenetically and ecologically diverse suite of tree species from mature forestry plots at The Morton Arboretum, Lisle, IL (in collaboration with M.L. McCormack, Fig. T4b.1). These observations of linkages among root, leaf, and wood phenology as measured across plant functional types at Morton Arboretum can also improve the of fine roots and their symbiotic mycorrhizal partners in ELM.


Fig. T4b.1 The timing of leaf, wood, root, and rhizomorph growth, assessed in forestry plots at Morton Arboretum using multiple methodologies.

4. How can we inform and improve model representation of root form and function?

Leveraging FRED, post-doc Dr. Bin Wang, co-advised by Drs. Dan Ricciuto and Colleen Iversen, is preparing a manuscript that seeks to tame fine-root system complexity to reduce Earth System Model uncertainty. Although improving leaf and canopy processes has been the focus of model-data integration since the 1970s, the representation of belowground processes has lagged. In contrast, theoretical and empirical advances in recent decades have improved our understanding of structural and functional differentiation and cooperation underlying fine-root function. To close this model-data gap, we have replaced the current 1-pool representation of fine roots in the E3SM Land Model (ELM) with a 3-pool belowground model structure representing both Transport and Absorptive fine roots, as well as Mycorrhizal fungi (TAM) to model vertically- and temporally-resolved fine-root systems. A demonstration of TAM in a big-leaf model (ELM) in temperate forests shows robust impacts on model predictions regardless of forest type (i.e. evergreen and deciduous). We argue that TAM should be adopted by the broader community of empiricists and modelers an effective approximation of the high-dimensional variability in fine-root systems because it balances realism and complexity with empirical support from databases of explicit root and fungal traits (e.g., FRED).



Fig. T4b.2 TAM impacts against the 1-pool homogenization on temperate evergreen (A-D) and deciduous forests (E-H), preserving the fine-root population C/N ratio of 42. The preservation is differentiated between two partitioning patterns: TAM\_descend (the partitioning of photosynthates allocated to fine roots descends from Transport roots first through Mycorrhizal hyphae last) and TAM\_ascend (the partitioning ascends from T through M, where hyphae receive the most photosynthate), on top of which comparisons are made by further adding a dynamic rooting depth distribution that changes in response to soil nitrogen availability (TAM\_descend\_dd and TAM\_ascend\_dd). Note these comparisons were made under an assumption of existing ELM model phenology. Fluxes of GPP (B, F) and heterotrophic respiration (C, G) are aggregated

annual values, while pools of fine-root mass and soil organic carbon (SOC) are averaged daily values. The variation (95% confidence interval) is attributed to parameter uncertainty with respect to root longevity and fine-root chemistry. Note different y-axis scales across panels are for distinguishing between different scenarios.

Task 4b. Delive	rable Status
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Date	Deliverable	Status
September	Manuscript on improving the representation of root form and function in	Underway
2022	ELM	(Wang et al.)
September	Manuscript on above- and belowground phenology linkages from 18 plant	Underway
2022	functional types at Morton Arboretum	(McCormack et al.)
September	Manuscript on representativeness of FRED root traits across edaphic and	Planned
2023	environmental conditions	(Iversen et al.)
September	Release FRED 4.0 with improved representation of tundra and tropical	Planned
2024	root traits	

# Task 4b Publications/Manuscripts

Weigelt A, Mommer L, Andraczek K, Iversen CM, Bergmann J, Bruelheide H, Fan Y, Freschet GT, Guerrero-Ramírez NR, Kattge J, Kuyper TW, Laughlin DC, Meier IC, van der Plas F, Poorter H, Roumet C, van Ruijven J, Sabatini FM, Semchenko M, Sweeney CJ, Valverde-Barrantes OJ, York LM, McCormack ML. (2021) An integrated framework of plant form and function: The belowground perspective. *New Phytologist* 232:42 - 59.

# Task 8: LeafWeb

We have updated Leafweb to accept joint measurements of Pulse Amplitude Modulated (PAM) fluorometry and gas exchange. Both fluorometry and gas exchange measurements are essential for photosynthesis research both in laboratory and field environments, and are routinely measured by plant scientists around the world. Leafweb was initially designed to fit the Farquhar model which uses only gas exchange data, and thus did not provide options for submitting and accepting PAM fluorometry data. With the new development of photophysical and photochemical models by Task 6, it is now possible to use joint measurements of PAM fluorometry and gas exchange to improve photosynthesis modeling. With the enabling of joint collection and analysis of PAM fluorometry and gas exchange, Leafweb will become a much more powerful web tool for supporting photosynthesis research and carbon cycle modeling.

In conjunction with enabling joint submission and analysis of PAM fluorometry and gas exchange measurements, we have been conducting offline tests on ways to increase the flexibility in data formats that Leafweb users are allowed to use. The current operational version of Leafweb (i.e, www.leafweb.org) has a strict requirement on data formats, e.g., which variable appears in which column in the submission file and in which lines where metadata and actual data are stored must be exact; otherwise, the background processing algorithms of Leafweb will not work. In offline tests, we now use keywords to define variable names. Users are free to place a variable in any column as long as the keyword for that variable is used as the header name for that column. Leafweb automatically searches for that keyword to determine the column in which the variable value is stored. Also, the start and end of the metadata and actual PAM fluorometry and gas exchange data are automatically determined. These added flexibilities will reduce the time users need to prepare for data submission and analyses by Leafweb.

Also in offline efforts, we have started to integrate codes for the models of photophysics of Gu et al. (2019) and photochemistry (Gu et al. 2022a) into the Leafweb code system for both C3 and C4 photosynthetic pathways. This integration will enable the joint optimizations of photophysical, photochemical, and biochemical parameters of photosynthesis by Leafweb when

joint measurements of PAM fluorometry and gas exchange are submitted by users. Previously, only the parameters in the biochemical model of photosynthesis (i.e., the Farquhar model) can be estimated. With the new efforts, we aim to enable Leafweb to estimate essentially all photophysical, photochemical, and biochemical parameters of photosynthesis that are of interests to ecophysiologists, modelers, and remote sensing scientists.

We are also working to update the C4 photosynthesis model in Leafweb. The current version of Leafweb uses a simple empirical C4 model that is commonly used in large scale carbon cycle models. But this simple C4 model is too simplistic and cannot represent full responses of C4 photosynthesis to climate change. Our aim in this aspect is to replace it with the state-of-the art biochemical model of C4 photosynthesis commonly used by plant scientists. Furthermore, we intend to couple the state-of-the-art C4 biochemical model with the photophysical model (Gu et al. 2019) and the photochemical model of Gu et al. (2022a) to enable Leafweb for complete analyses of C4 photosynthesis (C3 and C4 share the same photophysics and photochemistry but differ in biochemistry).

Date	Deliverable	
September 2021	LeafWeb manuscript synthesizing the dynamics of NPQ and qL for modeling photosynthesis.	
September 2022	Addition of LeafWeb support capability for integrated light and carbon reaction modeling	On track
Oct 2022	Release of the quality-controlled subset of leafweb data	On track
June 2023	Complete the coding for the coupled photophysical, photochemical, and biochemical (PPB) model of photosynthesis for leafweb for C3 photosynthetic pathway	Planned
July 2023	Invitation for community test of new leafweb with the coupled PPB modeling capability for C3	Planned
January 2024	Formal announcement and launch of the new leafweb with the coupled PPB capability for C3	Planned
July 2024	Code development of the coupled photophysical, photochemical, and biochemical model for leafweb for C4 photosynthetic pathway	Planned

Task 8. Deliverable status FY2021 through FY2022

# Task 8 Publications/Manuscripts

- Han J, Gu L, Warren JM, Guha A, Mclennan DA, Zhang W, Zhang Y. 2022. The roles of photochemical and non-photochemical quenching in regulating photosynthesis depend on the phases of fluctuating light conditions. *Tree Physiology* 42: 848-861.
- Han J, Gu L, Wen J, Sun Y. 2022. Inference of photosynthetic capacity parameters from chlorophyll a fluorescence is affected by redox state of PSII reaction centers. *Plant, Cell & Environment* 45: 1298-1314.

## TES SFA DATA SYSTEMS, MANAGEMENT, AND ARCHIVING UPDATE

The open sharing of all data and results from the TES SFA research and modeling tasks among researchers, the broader scientific community, and the public is critical to advancing the mission of DOE BER's Earth and Environmental Systems Sciences Division. Active data sharing facilitates delivery of TES SFA products to our stakeholders. TES SFA researchers continue to develop and deploy the data systems, repositories, tools, and integration capabilities needed for the collection, QA, storage, processing, sharing, analysis, and archiving of data and model products.

These capabilities facilitate model-data integration and provide accessibility to model output and benchmark data for analysis, visualization, and synthesis activities in support of the TES SFA Vision. Task-specific web sites (e.g., SPRUCE: https://mnspruce.ornl.gov and FRED: <u>https://roots.ornl.gov),m</u> access to web-based tools (e.g., LeafWeb: https://leafweb.org) and links to external products (e.g., microbial metagenomes) enable these interactions.

All results of laboratory experiments and sample analyses, synthesis of information, genomics analyses, and model products (inputs, codes, outputs) developed in support of TES SFA tasks and data collected specifically at the SPRUCE experiment facility, are submitted to the respective SPRUCE or TES SFA data archive in a timely manner such that data will be available for use by project scientists and collaborators and, following publication, the public, thru the SPRUCE (https://mnspruce.ornl.gov) and TES SFA (https://tes-sfa.ornl.gov) websites.

Data acquisition and real time display of SPRUCE experimental plot monitoring data are fully implemented. More than 1,100 sensors are deployed across 16 instrumented plots. Real-time visual displays of selected monitoring and infrastructure operational control parameters are provided using Campbell Scientific's Real-Time Monitor and Control (RTMC) software. Vista Data Vision (VDV) software has been implemented for performance monitoring, data visualization, and data review by the SPRUCE Team. Data are stored and will be accessible through web-based search and download applications to the project and public.

In addition, ongoing SFA task data products continue to be archived at program-specific archives (e.g., MOFLUX at AmeriFlux). New SFA task data products are publicly available on the ORNL TES-SFA web site: https://tes-sfa.ornl.gov.

See **APPENDIX B** for the list of newly and previously archived datasets and details of data sharing, archiving, and fair use.

# **AFFILIATED TES SFA-SUPPORTED PUBLICATIONS**

Staff supported by the TES SFA continue to collaborate and complete work funded by US DOE BER in prior fiscal years that may not explicitly be funded under Tasks 1 through 8. The following listing shows additional manuscripts completed since the June 2021 with some TES SFA support.

- Ardón M, Zeglin LH, Utz RM, Cooper SD, Dodds WK, Bixby RJ, Burdett AS, Follstad Shah JJ, Griffiths NA, Harms TK, Johnson SL, Jones JB, Kominoski JS, McDowell WH, Rosemond AD, Trentman MT, Van Horn DJ, Ward AK (2021)0: A global meta-analysis from streams and rivers. *Biological Reviews* 96:692-715.
- Benson MC, Miniat CF, Oishi AC, Denham SO, Domec JC, Johnson DM, Missik JE, Phillips RP, Wood JD, Novick KA. 2022. The xylem of anisohydric *Quercus alba* L. is more vulnerable to embolism than isohydric codominants. *Plant, Cell & Environment* 45: 329-346, DOI: 10.1111/pce.14244.
- Costello DM, Tiegs SD, Boyero L, Canhoto C, Capps KA, Danger M, Frost PC, Gessner MO, Griffiths NA, Halvorson HM, Kuehn KA, Marcarelli AM, Royer TV, Mathie DM, and 79 coauthors (listed alphabetically) (2022) Global patterns and controls of nutrient immobilization on decomposing cellulose in riverine ecosystems. *Global Biogeochemical Cycles* 36:e2021GB007163.
- Griffiths NA, Mulholland PJ (2021) Long-term hydrological, biogeochemical, and climatological data from Walker Branch Watershed, east Tennessee, USA. Hydrological Processes 35:e14110.

# **4AII. SCIENCE HIGHLIGHTS SINCE JUNE 2021**

- ORNL TES SFA staff authored 45 papers that have been published or are in press/accepted status since June 2021. Notable papers include the following high impact totals: 1 in *PNAS Nexus*, 3 in the *Nature* family of journals. An additional 2 manuscripts are in peer review.
- SPRUCE The overall C sink capacity declines with warming driven mostly by heterotrophic C losses, and the combined plot-level estimate of NCE shows that loss rates per degree C are similar through 6-years of manipulation without a strong eCO2 effect.

- SPRUCE Our results demonstrate that microbes, which respond rapidly to temperature alterations, can play key roles in *Sphagnum* host growth response to rapidly changing environments.
- SPRUCE We found that warming exponentially increased plant-available ammonium and phosphate, but that nutrient dynamics were unaffected by elevated [CO<sub>2</sub>]. The warming response increased by an order of magnitude between the first and fourth year of the experimental manipulation, perhaps because of dramatic mortality of *Sphagnum* mosses in the surface peat of the warmest treatments.
- SPRUCE Solute concentrations in outflow continue to respond to warming. Higher total organic carbon (TOC) concentrations (Fig. T1.5), along with some cations and metals (i.e., calcium, aluminum, iron), were observed in warmer enclosures while the responses of total N and total P concentrations to warming were variable.
- SPRUCE Preliminary results indicate that C, N, and P in biomass pools decreased with warming though the decrease in biomass N was somewhat offset by a positive interaction between the elevated CO<sub>2</sub> and warming treatments. The overall pattern of losses was
- MOFLUX/SIF Chlorophyll a fluorescence is the only optical signal that is functionally tied to the operation of photosynthetic machinery and observable with remote sensing. However, whether and how photosynthesis can be inferred by observing chlorophyll a fluorescence, which is the basis for applying Solar-Induced Chlorophyll Fluorescence (SIF) to monitor gross primary productivity (GPP) at different scales, are still not clear. To illuminate these issues, we coupled the photophysical model previously developed by us with a biochemical model and applied the coupled model on diverse species from different biomes around the world. It was shown that by using chlorophyll a fluorescence as observed inputs, the coupled model is less sensitive to parameter uncertainties and can produce more accurate estimates of photosynthetic rate as compared with conventional models because of the photosynthetic functional information contained in the observed fluorescence.
- MOFLUX/SIF Photosynthetic capacity parameters control gross primary productivity for a given set of environmental conditions. Their precise determination is key to understanding and predicting ecosystem functioning and productivity. Previous studies have reported inconsistent relationships between photosynthetic capacity parameters and solar induced chlorophyll a fluorescence (SIF). To determine the source of these inconsistencies, the photophysical light reaction model, previously developed by us, was coupled to the Farquhar von Caemmerer Berry biochemical model of photosynthesis and used to model how variations in photosynthetic capacity parameters affect fluorescence emission critically depends on the redox condition of photosystem II (PSII) reaction centers as indicated by the fraction of open PSII reaction center (qL). Further, it was shown that photosynthetic capacity parameters can be estimated from the product of fluorescence intensity and qL.
- MOFLUX/SIF Ecosystem respiration returns most of the carbon sequestered by photosynthesis to the atmosphere and is the second largest carbon flux after gross primary production (GPP). Many processes contribute to ecosystem respiration. These processes have drastically different respiratory characteristics. Understanding their relative contributions is essential for predicting net ecosystem carbon balance. This study first developed a novel three-way CO2 flux partitioning algorithm and then applied it to the long-term flux and soil respiration datasets collected at MOFLUX to partition observed fluxes into aboveground plant respiration, belowground root and soil respiration, and GPP.
- CC Modeling An improved methane modeling framework was integrated into ELM-SPRUCE and TECO-SPRUCE, allowing for improved predictions of the seasonal cycle of CH4 fluxes and concentration profiles. The impact of hydrological feedbacks under the warming treatment was also found to be significant.
- CC Modeling A new method for model-data assimilation using surrogate modeling was developed, allowing for a more efficient cycle of model improvement and for identifying which types of new observations would be most effective in reducing model uncertainty.

 Soil C – Tasks 6 and 5 continue to use the high-resolution measured soil chamber respiration data and SIF information from MOFLUX over the last 5 years to examine how seasonal and diurnal patterns and responses to drought influence the proportions of heterotrophic versus autotrophic respiration. New findings show how heterotrophic respiration is sensitive to soil temperature at daily to seasonal timeframes, while autotrophic respiration responds to aboveground productivity (i.e., LAI), consequently, the time of year and LAI are most influential for determining the partitioning between heterotrophic and autotrophic respiration and suggest methods for partitioning respiration using these variables.

## **4AIII. ANALYSIS OF PUBLICATIONS**

Through senior and coauthored effort, TES SFA staff produced 47 publications or completed manuscripts since our last summary report. This total includes 45 published/in press/accepted journal articles and 2 working through the review process. This level of 1-year productivity over  $(47 \text{ y}^{-1})$  just exceeds our average paper production rate from March 2015 through June 2021 of 46 y<sup>-1</sup>. A TES SFA cumulative publication summary since 2015 is provided in **Appendix A** with the most recent publications from the current reporting period listed first. This listing duplicates the Task-specific summaries already provided.

The TES SFA group published in 35 different peer-reviewed publications in this reporting period including one in *PNAS Nexus* (Meng et al. 2022) and 3 in the *Nature* family of journals (Craig et al. 2022; Helbig et al. 2022; Novick et al. 2022a). We also published 5 papers in *New Phytologist*, 3 papers in *Journal of Geophysical Research – Biogeosciences, 2 papers in Global Change Biology* and 2 in *Plant Cell and Environment*.

Journal selection for publication of TES SFA work is at the discretion of the author team. Journals are selected to achieve maximum exposure of the research results for the science community. We focus on journals having high impact factors, but that is not necessarily the primary criteria for the selection of a journal for publication of a given research result. High-profile journals (e.g., *Science, Nature* family of journals, *PNAS*) are pursued for the publication of results anticipated to be of general interest to a wide audience. We find that solid and well-presented scientific results are well received and cited in all of our chosen journals.

We also continue to place significant and sustained effort on the production of archived data sets based on TES SFA work. A complete and cumulative summary of TES SFA data sets is provided in **Appendix B**.

### **4B. FUTURE SCIENCE GOALS AND PLANS**

Future science plans for the TES SFA were detailed in the quadrennial review document submitted to DOE BER in February 2019. We are in the process of adding high-temporal resolution  $CO_2$  and  $CH_4$  flux observations for all SPRUCE treatments.

The TES SFA plans to enhance efforts to leverage knowledge gained from past and ongoing process studies, manipulative experiments, and ecosystem observations (e.g., SPRUCE, PiTS, belowground fundamentals, landscape fluxes, EBIS, and TDE) to improve ecosystem models. Future, experimental studies will be used to test key mechanistic processes in ELM. To improve the modeling of gross primary production, we will apply new understanding of canopy processes enabled through the development of the SIF instrument and use and photophysical and photochemical models to improve the modeling of gross primary production in ELM.

<u>Improved Sphagnum modeling</u> – Within the ELM framework we will continue to improve a mechanistic model of *Sphagnum* photosynthesis based on in situ assessment of GPP, *Sphagnum* production, capitula water content and environmental conditions. The data and modeling results will also provide critical information for latent heat and energy balance calculations.

<u>Coupled photophysical, photochemical, and biochemical modeling</u>. We will continue our current effort in the coupled modeling of photophysical, photochemical, and biochemical reactions of photosynthesis. The goal is to develop a coupled model with all parameters that can be estimated from typical leaf-level measurements of PAM fluorometry and gas exchange and can be implemented in global models such as ELM with adding parameter burdens and computation cost.

<u>Measures of microbial biomass at SPRUCE</u> – Three coring activities at SPRUCE in 2021-2022 have focused on the distribution of microbial properties as a function of depth, using three different methods (qPCR, chloroform fumigation, and PLFA). The team will complete the analyses for all methods and will compare their findings with other environments, potentially providing quantitative relationships to maximize the use of more kinds of microbial data for use in model simulations.

## 4C. NEW SCIENCE FOCUS AND IDENTIFIED KNOWLEDGE GAPS

We have clearly seen warming induced changes in the phenology of the bog vegetation with the expected acceleration of spring growth activities, but also a clear extension of the autumn growing season. In many current ecosystem models including SPRUCE-ELM, the predominant driver for fall senescence based on the interpretation of observational interannual variation is day length. The SPRUCE data show that algorithms will need to be modified to include warming influences on fall phenology changes. We have added new phenology cameras to better capture dynamics of ground layer vegetation.

Carbon cycle analysis clearly shows major losses of C from the SPRUCE ecosystem proportionate to the level of warming. At this time models are overestimating the influence of  $eCO_2$  on C uptake and may not be adequately accounting for nutrient and water limitations.

Early year results from the SPRUCE study suggest further investment in the biogeochemical cycling of both nitrogen and phosphorus are needed to adequately capture long-term nutrient feedbacks within the bog with warming. Several key areas identified include N and P mineralization, nutrient resorption during fall senescence, and the availability of organic N and P in soils. Field measurements investigating the temperature response of N and P mineralization, as well as organic nutrient availability were originally planned for the summer of 2020 but we are considering making these observations on laboratory incubations instead because of COVID-19 – related travel restrictions. Resorption of N and P during fall senesce is being quantified for litter samples collected in the fall of 2019. Models are being used to evaluate the potential feedback magnitudes from P limitations and N<sub>2</sub> fixation inputs to better define the need for future measurements.

Identified knowledge gaps also drive model developments for land surface development of the E3SM model, and within NGEE project tasks that are complementary to efforts within the TES SFA. The TES SFA will continue to contribute new science to E3SM code base, including new algorithms for phenology and responses to extremes, and *Sphagnum* processes described above. TES SFA modeling efforts will benefit from developments in the other projects, such as improved hydrology through the ELM-PFLOTRAN coupling in NGEE-Arctic and the inclusion of a global phosphorus cycling model in E3SM.

We have obtained conclusive evidence that photophysical and photochemical processes, in addition to biochemical processes, are important for modeling photosynthetic responses to environmental change including extreme events. The photophysical and photochemical models that have been developed by the TES SFA need to be tested and parameterized for species across major biomes and climates to prepare for their eventual incorporation into ELM. To support this effort, we will update the Leafweb processing code to enable automatic joint analyses of photophysical, photochemical, and biochemical reactions of photosynthesis and to gather measurements of PAM fluorometry and gas exchange from photosynthesis researchers from around the world.

Feedback from review panels and science advisory committees have highlighted the lack of continuous ground-flux measurements at SPRUCE. In 2021, BER provided substantial equipment funding to relieve this important knowledge gap. As of 2022, greenhouse gas analyzers and 2 large chambers are being installed in each treatment chamber to measure CO<sub>2</sub> and CH<sub>4</sub> dynamics twice hourly. The SPRUCE team and Task 5 are working to install and test the systems in advance of the start of a postdoctoral researcher to onboard in FY22.

#### **4D. COLLABORATIVE RESEARCH**

We continue to encourage key external groups to develop complementary research tasks for the benefit of TES SFA research tasks. Support for the following independently funded research groups is being provided through the use of SPRUCE leased office/lab facilities and access to the SPRUCE experimental site on the S1 Bog:

- Dr. Joel Kostka (Georgia Tech), Jeff Chanton (Florida State) and colleagues have received new support from DOE BER for new studies of microbial ecology at SPRUCE and we look forward to continuing to work with them.
- Dr. Andrew Richardson (Northern Arizona University) leads the task on phenology with his funding for the PhenoCam network and made this a funded subtask in FY2018 and beyond.
- Drs. Brandy Toner, Ed Nater and colleagues from the University of Minnesota, are examining mercury and sulfur dynamics in the SPRUCE experiment using funding provided through the USDA Forest Service.
- Dr. Karis McFarlane and colleagues at LLNL-CAMS provide measurement support for <sup>14</sup>C isotopic composition of air, plant tissues and peat from the S1-Bog and SPRUCE experimental plots.
- Dr. Nancy Glenn (Boise State) is contracted through SPRUCE to provide ground-level LIDAR observations as a supplement to our destructive woody harvests and *Sphagnum* production estimates. Jake Graham is executing the onsite work.
- Dr. Yiqi Luo's group (now at Cornell) is utilizing new high-temporal-resolution, model-data iterative analyses to better define measured ecosystem responses with the intention of helping the research group apply measurement efforts to critical processes.
- Dr. Xiaofeng Xu (San Diego State University) continues work with the modeling group on improved biogeochemical cycling models for methane flux.
- Dr. Danielle Way (University of Western Ontario) is an unfunded collaborator who has been
  providing expertise and multiple PhD students to assist with seasonal assessment of *Picea* and *Larix* photosynthetic and respiratory thermal and CO<sub>2</sub> acclimation.
- Dr. Jalene M. LaMontagne (DePaul University) joined the SPRUCE group in 2017 to study mast seeding patterns in response to climate change.
- Dr. M. Luke McCormack (Morton Arboretum) is making new observations across a phylogenetically and ecologically diverse suite of tree species from mature forestry plots at The Morton Arboretum, Lisle, IL to advance our framework of root trait understanding gained from FRED.
- Dr. Al Kovaleski (University of Wisconsin) is an unfunded collaborator that plans to evaluate how cold hardiness changes in the tree and shrub species due to warming treatments, and the interaction with a longer growing season and warmer winters, and how different chilling models perform in predicting dormancy stage in woody perennials.
- Dr. Anping Chen (Colorado State University) continues work with the modeling group on the global SIF analysis and the use of observational SIF to constrain the ELM GPP and phenology.
- Prof. Mingzhou Jin (University of Tennessee at Knoxville) and his PhD student, Rongyun Tang, continue work with the modeling group on the wildfire analysis and evaluation for ELM.
- Prof. Lawren Sack (UCLA) is visiting MOFLUX to make detailed measurements of plant traits for an NSF macrosystems biology project.
- Prof. Christian Frankenberg (Caltech) is conducting PAM fluorometry monitoring at the MOFLUX site as part of collaborative activities funded by NASA.
- Dr. Jessica Gutknecht (University of Minnesota) has independently measured microbial biomass using PLFA at SPRUCE for a number of years; and is now collaborating with Task 5 on multiple measures of microbial biomass.
- Dr. Jean Philippe Gibert (Duke University) is funded through DOE BER Genomics Sciences to investigate the neglected controls on peatland C cycling: bacterial predation by protists and viral infections.
- Dr. Michael Gundale (SLU, Swedish University of Agricultural Sciences) is an unfunded collaborator investigating SPRUCE feathermoss responses.

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# 5. STAFFING AND BUDGET SUMMARY

# 5A. FY2022 FUNDING ALLOCATION BY PROGRAM ELEMENT

Total expected available funding for ORNL's TES SFA in FY2022 included \$7,117K carryover from FY2021 (including \$332K for external commitments) and expected \$8,300K of new budget authorization. FY2022 spending to date is summarized in the following table.

include prior year carryover amounts.						
Task	Cost Through 14 June 2022 (\$K)	Commitments Through 14 June 2022 (\$K)	Remaining Funds 14 June 2022 (\$K)			
T1: SPRUCE Science	\$2,861K	\$702K	\$2,215K			
T6: MOFLUX, etc.	378	226	300			
T3ab: Carbon Cycle Modeling	650	131	509			
T3d: MAAT	226	0	288			
T4c: Process Study - Root Func.	81	0	12			
T5: Soil C Studies	192	2	292			
T4b: Process Study - Root traits	143	78	215			
T8: LeafWeb	52	0	92			
T1.1: SPRUCE – Operations	622	354	1,024			
T1.2: SPRUCE – Materials	509	558	64			
T1.3: SPRUCE – Reserve	0	0	685			

# FY2022 Budget expenditures by TES SFA Program Element through 14 June 2022. The data include prior year carryover amounts.

We are currently spending at rates consistent with the spending plans outlined in the February 2019 TES SFA renewal proposal budgets for FY2021 extended into the extra funded years under COVID-19 protocols. We anticipate unspent carry over funds to be approximately \$2,500K across all TES SFA tasks and external SFA commitments.

0

\$5.714K

0

\$2.051K

1.126

\$6.822K

## **5B. FUNDING ALLOCATION TO EXTERNAL COLLABORATORS**

SFA Contingency

**SFA Totals** 

A variety of collaborations are maintained and funded by the TES SFA to provide necessary commodities, and disciplinary expertise and effort in areas critical to the completion of research tasks. In FY2021 we directly funded the following individuals or groups.

**The University of Missouri (\$170K)** is subcontracted to provide MOFLUX on site execution of the following measurements: stand-level eddy covariance, soil CO<sub>2</sub> efflux, belowground production via repeated minirhizotron image collections, stem allometric increment data, and litter basket net primary production. Since June 2016, J.D. Wood serves as Missouri site-PI of the MOFLUX site.

**Yiqi Luo- Northern Arizona University (\$150K)** – Dr. Luo's research group at NAU is developing an ecological forecasting capability at SPRUCE. Using the TECO model as a demonstration, data assimilation capabilities are being developed and applied using SPRUCE observations, and forecasts were made for the 10 experimental plots using a range of future scenarios. A methane model was also added to TECO. **Xiaofeng Xu - San Diego State University (\$25K)** - In a joint subcontract with NGEE-Arctic, Dr. Xu is developing and testing a CH<sub>4</sub> modeling capability for the CLM and ELM-SPRUCE modeling efforts. This work has contributed to two manuscripts. Work to refine and optimize the model with SPRUCE observations is continuing.

**M. Luke McCormack – The Morton Arboretum (\$88K)** – The Morton Arboretum (and Dr. M. Luke McCormack) have been subcontracted to quantify changes in above- and belowground phenology among plant functional types at Morton Arboretum to inform the development of belowground phenology algorithms in ELM and add to the limited root phenology observations in the Fine-Root Ecology Database.

**RhizoSystems, LLC (\$28K)** – The company who designed and built the automated minirhizotrons (AMRs) is being subcontracted for support and maintenance of these systems. This includes off-site repair and maintenance of all AMR and RhizoSystems-installed equipment and remote assistance with field repair on-site. It also includes assistance with and support of the RV3n software as well as AMR-related software updates to and routine maintenance of RhizoSystems-installed computers.

**Interagency Agreement with the USDA Forest Service (\$40K)** – This agreement allows Forest Service employees to help with the operation, planning and execution of the SPRUCE experimental infrastructure and science tasks. It also provides some coverage for the use of the USDA FS bunk house on the Marcell Experimental Forest.

Andrew Richardson – Northern Arizona University (\$98K) – This contract allows Dr. Andrew Richardson's group to maintain the automated phenology observations and greenness calculations for all treatment and ambient plots on the SPRUCE site. Dr. Richardson also leads the phenology task for the SPRUCE project.

**Nancy Glenn – Boise State University (\$60K)** This contract provides twice annual terrestrial lidar scans of the SPRUCE experimental plots to help assess vegetation growth and microform elevation change (hummock and hollow distributions).

**Karis McFarlane – Lawrence Livermore National Laboratory (\$161K)** – We contract with LLNL to provide isotopic analyses ( $^{14}$ C and  $^{13}$ C) for air (x5 events per year) and tissue analyses (x1 per year) to provide a record of the application and accumulation of unique isotopic tracers into the SPRUCE ecosystem. Funding is also provided in for her group to return to the site to evaluate the isotopic composition of emitted CO<sub>2</sub> and CH<sub>4</sub>.

**Infrastructure subcontracts** in support of the SPRUCE project in FY2020 include funds and funding for site maintenance (**Pokegama Electric \$65K**), electrical service (**Lake Country Power \$147K**), propane supply (**Lakes Gas Co. \$407K**), eCO<sub>2</sub> supply (**PRAXAIR Inc. \$135K**), fiber internet connections (**\$10K**), and leased space in Minnesota (**\$41K**). The amounts required for each of these operational contracts are reevaluated annually as actual usage rates and prices change.

# **5C. PERSONNEL ACTIONS AND PROCEDURES**

<u>New Hires and Promotions</u> – Terri Velliquette and Tom Ruggles were brought on board to replace the skills for data management following Leslie Hook's retirement. Brandi Maki was hired as full-time technical support staff to work in Minnesota with Kyle Pearson to operate the SPRUCE experiment.

<u>Anticipated Future Hires</u> – Looking ahead to FY2023, the TES SFA is in the process of hiring replacement postdoctoral fellows to sustain effort and to supplement full time staff positions and as the budget allows.

<u>Retirements and Releases</u> - Les Hook retired at the end of FY2021.

<u>Procedures for advancing new and developing investigators</u> - New TES SFA staff members are commonly first hired through postdoctoral research associate positions and their performance and contributions to task activities are tracked. Our postdocs are vetted for potential future roles as task leads and are hired as staff into leadership roles as appropriate for our needs.

Where identified disciplinary needs are established (and for which adequate funding is available) the TES SFA also has the capacity to hire established staff persons directly into a task leadership role. When a need for new staff is identified but funding is insufficient to initiate a new hire, ORNL internal funds may be requested through a strategic hire program to bring individuals on board. This internal program

allows for a 1 to 2-year transitional period to enable the TES SFA group to establish an appropriate, stable, and fully funded position.

Within the TES SFA, task accomplishments and budget management is executed at an overarching level by the Principal Investigator with feedback from all Task leads. Individual Task leads are given the responsibility to track scientific progress and the responsibility for managing their fiscal resources within an annual cycle. Training to allow new staff to understand ORNL procedures, accounting systems, and managerial activities is available and provided when appropriate. Such training, in addition to one-on-one mentoring with established staff, provides developing staff with the information and skill sets required to transition into leadership roles. At the institutional level, ORNL has formal programs for mentoring high-potential early career staff, and we use informal mentoring at the personal level to ensure that staff with potential leadership qualities are identified and helped with career development.

## 5D. NATIONAL LABORATORY INVESTMENT IN THE PROGRAM IN FY2022

Concepts for the belowground warming technologies used for the SPRUCE Experiment (Task R1) were initiated with ORNL LDRD funds totaling \$480K in FY2008 and FY2009. In FY2014, ORNL provided the equivalent of \$1000K staff support from internal funds to allow completion of the SPRUCE warming aboveground infrastructure. No ORNL funds were requested or have been needed in FY2022.

The Climate Change Science Institute brings together all ORNL Climate Change staff including members of the TES SFA to foster day-to-day interactions among modelers, experimentalists and data management specialists. The TES SFA is supported by world-class capabilities at ORNL. The National Leadership Computing Facility provides an open, unclassified resource that we will use to enable breakthrough discoveries in climate prediction. We continue to be engaged with neutron sciences through cutting-edge root and rhizosphere imaging research at the High Flux Isotope Reactor (HFIR) and active participating in science development teams for the future VENUS beamline at SNS and proposed beamlines at the potential Second Target Station. We work with the DOE BER data center ESS-DIVE as our primary permanent data repository (see also Appendix B).

We also use other facilities at collaborating DOE National Laboratories. The Lawrence Livermore National Laboratory – Center for Accelerator Mass Spectrometry (LLNL-CAMS) provides large volume, high precision <sup>14</sup>C measurements for ecosystem tracer studies. Pacific Northwest National Laboratory's Environmental Molecular Science Laboratory combines advanced instrumentation such as high-throughput mass spectrometry, advanced microscopy instruments, and NMR instruments with high performance computing.

## **5E. CAPITAL EQUIPMENT**

Since the threshold amount of funds needed to define a capital expenditure is high, no ORNL TES SFA funds have been used to acquire capital equipment in FY2022. Funding for SPRUCE experimental infrastructure maintenance and development at the S1 Bog are not classified as capital expenditures but represent an analogous investment for the decadal duration of the experiment.

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# **APPENDIX A: COMPLETE PUBLICATION LIST – ORNL TES SFA**

Published, accepted and in review papers since the June 2021 annual Report

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#### **APPENDIX B: TES SFA DATA SETS AND POLICIES**

#### Data Policy for Archiving, Sharing, and Fair-Use

The open sharing of ORNL TES-SFA data, modeling products, and documentation among researchers, the broader scientific community, and the public is critical to advancing the mission of DOE BER's Earth and Environmental Systems Sciences Division. The policy is applicable to all TES-SFA participants including ORNL, cooperating independent researchers, and to the users of data products. Data collected by TES-SFA researchers, results of analyses and syntheses of information, and model algorithms and codes will be quality assured, documented, and archived and will be made available to the public.

Archived data products are freely available to the public. Users should acknowledge the contribution of the data provider with the citation (with DOI) as provided in the documentation and acknowledge the U.S. DOE BER Earth and Environmental Systems Sciences Division. Find the complete document "ORNL TES-SFA Data Policy: Archiving, Sharing, and Fair-Use" last updated May 15, 2019 at: <u>https://mnspruce.ornl.gov/sites/default/files/ORNL TES-SFA Policy 2090515.pdf</u>

TES-SFA data management plans and policies align with the most recent DOE policies for "Public Access to the Results of DOE-Funded Scientific Research"

<u>https://mnspruce.ornl.gov/sites/default/files/DOE\_Public\_Access%20Plan\_FINAL.pdf</u> and the "Statement on Digital Data Management"

https://science.osti.gov/Funding-Opportunities/Digital-Data-Management

#### **Data Products Archived**

There are usually multiple data sets in the development que at any one time. For efficiency, the data management (DM) team implemented a collaborative work management tool – Trello - to track data sets from initial contact to publication and track tasks related to the website. The DM team established weekly check-in meetings and regularly presented updates at project meetings and the all-hands meetings.

For ongoing TES-SFA tasks, 30 products are available to the public. The products include regularly updated time-series of SPRUCE environmental data, peat analyses, modeling archives, results of laboratory incubations, links to genomic products at JGI, "supporting validation data" for specific publications (e.g., organic matter characterization), web-based tools (e.g., LeafWeb), historical Walker Branch data, literature compilations (e.g., FRED 3.0), and characterization of SPRUCE plots (e.g., elevation).

SPRUCE to date has archived and shared 82 data products with the public and 5 products available to the project team only typically because associated manuscripts are awaiting publication.

Persistent identifiers (digital object identifiers, DOIs) for all products are registered through the OSTI (DOE Office of Scientific and Technical Information) E-Link System. SPRUCE products as 10.25581/spruce.0XX/zzzzzz and TES SFA products as 10.25581/ornlsfa.0XX/zzzzzz. Comprehensive metadata can be entered that facilitate the transfer of metadata, documentation, and data to the DOE's Environmental Systems Science Data Infrastructure for a Virtual Ecosystem (ESS-DIVE) archive.

#### **Transfer of Data Products to DOE ESS-DIVE Archive**

Data management (DM) staff of the TES SFA worked with the ESS-DIVE staff to increase the number of mirrored data packages at the repository with 10 TES-SFA and 21 SPRUCE data packages to date. These first data packages are publicly accessible and considered final, that is, they will not be updated with periodic new data additions. These data packages are available through the ESS-DIVE repository search tool and within portals developed by DM staff that provide information about the TES SFA and SPRUCE with filtered views of associated data. DM staff continued to review and update the project data and metadata to meet the repository requirements. In this past year, DM staff began efforts to implement some of the new repository reporting formats which included inviting ESS-DIVE staff to present at two of the SPRUCE monthly meetings.

#### **Code Sharing**

Public release of SPRUCE-specific E3SM code will be managed by the E3SM project as part of a collaboration agreement between the ORNL TES SFA and E3SM and subject to E3SM policies and licensing (https://e3sm.org/resources/policies/). Development branches of the E3SM code for research purposes will also be available through https://github.com/E3SM-Project/E3SM/. The most recent release was assigned a DOI through DOECODE (10.11578/E3SM/dc.20210927.1) with a 3-Clause BSD license. Code developments will be discussed and agreed upon by the TES SFA modeling team, with the understanding that our goal as a group is to make the developments here available to the larger community as soon as possible. For reproducibility, publications using model output will include information about the specific release or development branch used in the simulations. Public release removes the 'rights' of code developers to be automatically considered for co-authorship. However, we encourage users of the released model to consider informing or including those developers to the extent it would benefit the users' analyses.

## **Collaboration across DOE BER Projects**

TES-SFA participant Terri Velliquette serves as Co-lead for Data Management on the DOE ESS Cyberinfrastructure Working Group, serves as a member of the ESS-DIVE Archive Partnership Board and builds relationships with colleagues both within DOE and externally through interactions with the ESS Community, Earth Science Information Partners (ESIP), DOE Office of Scientific and Technical Information (OSTI), ESS-DIVE, etc. From these interactions, the TES SFA can stay current with community archiving expectations, maintain awareness of data management best practices, and can present project challenges and requests for improvements.

#### Websites Upgrades

The FRED, SPRUCE and TES SFA Websites are all now upgraded to the latest version of the website content management system Drupal 9. With this upgrade came improved security measures, changes to look of the websites, and some technical issues to resolve. The DM team worked with and enlisted the ORNL Drupal Team to improve the file access and download capabilities of the SPRUCE website by transferring the file host from Drupal to Dropbox and installing a Dropbox module directly on the data set landing page. Dropbox also improved the backend workflow and the ability to transfer large data files to the DM team.

## TES SFA Software:

1. The Multi-Assumption Architecture and Testbed (MAAT v1.0) is now open source, available at <u>https://github.com/walkeranthonyp/MAAT</u>.

## SPRUCE Public Data Sets (\*\*New or ##Upgraded since June 2021):

- \*\*Baysinger MR, Wilson RM, Hanson PJ, Kostka JE, Chanton JP (2021D). SPRUCE Compositional Stability of Peat in Ecosystem-Scale Warming Mesocosms, 2014 and 2019. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.093/1820162
- Childs J, Defrenne CE, Brice DJ, Woodward J, Holbrook KN, Nettles WR, Taggart M, Iversen CM (2020D) SPRUCE High-Resolution Minirhizotrons in an Experimentally-Warmed Peatland Provide an Unprecedented Glimpse at Fine Roots and their Fungal Partners: Supporting Data. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.081/1637336
- Dusenge ME, Ward EJ, Warren JM, McLennan DA, Stinziano JR, Murphy BK, King AW, ChildsJ, Brice DJ, Phillips JR, Stefanski A, Villanueva R, Wullschleger SD, Cruz M, Reich PB, Way DA (2020D) SPRUCE Photosynthesis and Respiration of *Picea mariana* and *Larix laricina* in SPRUCE Experimental Plots, 2016-2017. Oak Ridge National Laboratory, TES SFA, U.S.

Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.056/1455138</u>

- 4. Dusenge ME, Stinziano RJ, Warren JM, Ward EJ, Wullschleger SD, Hanson PJ, Way DA (2018D) SPRUCE Whole Ecosystem Warming (WEW) Photosynthesis and Respiration of *Picea* and *Larix* in Experimental Plots, 2016. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.056/1455138</u>
- Fernandez CW, Heckman K, Kolka R, Kennedy PG (2019D) SPRUCE Fungal Necromass Litter Bag Decomposition Study in SPRUCE Experimental Plots, 2016-2018. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.063/1503589</u>
- Finzi AF, Giasson MA, Gill AL (2016D) SPRUCE Autochamber CO<sub>2</sub> and CH<sub>4</sub> Flux Data for the SPRUCE Experimental Plots. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/SPRUCE.016</u>
- Furze ME, Jensen AM, Warren JM, Richardson AD (2018D) SPRUCE S1 Bog Seasonal Patterns of Nonstructural Carbohydrates in Larix, Picea, Rhododendron, and Chamaedaphne, 2013. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.037/1473917
- Graham JD, Glenn NF, Spaete LP (2019Da) SPRUCE Terrestrial Laser Scanning of Experimental Plots Beginning in 2015. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.067/1515552</u>
- Graham JD, Glenn NF, Spaete LP (2019Db) SPRUCE Microtopography of Experimental Plots Derived from Terrestrial Laser Scans Beginning in 2016. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.068/1515553
- Griffiths NA, Hook LA, Hanson PJ (2016Da) SPRUCE S1 Bog and SPRUCE Experiment Location Survey Results, 2015 and 2020. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.3334/CDIAC/spruce.015</u>
- 11. Griffiths NA, Sebestyen SD (2016Db) SPRUCE S1 Bog Porewater, Groundwater, and Stream Chemistry Data: 2011-2013. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.018</u>
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- Griffiths NA, Sebestyen SD (2017D) SPRUCE Hollow Elevation Data for Experimental Plots Beginning in 2015. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. http://dx.doi.org/10.3334/CDIAC/spruce.035
- 14. Gutknecht J, Kluber LA, Hanson PJ, Schadt CW (2017D) SPRUCE Whole Ecosystem Warming (WEW) Peat Water Content and Temperature Profiles for Experimental Plot Cores Beginning June 2016. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.041</u>
- Hanson PJ, U.S. Forest Service Staff, and SPRUCE Team (2012D) SPRUCE S1-Bog Vegetation Survey and Peat Depth Data: 2009. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.003</u>.
- Hanson PJ, Brice D, Garten CT, Hook LA, Phillips J, Todd DE (2012D) SPRUCE S1-Bog Vegetation Allometric and Biomass Data: 2010-2011. Carbon Dioxide Information Analysis

Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.004</u>.

- Hanson PJ, Krassovski MB, Hook LA (2015D) SPRUCE S1 Bog and SPRUCE Experiment Aerial Photographs. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.3334/CDIAC/spruce.012</u>
- Hanson PJ, Nettles WR, Riggs JS, Krassovski MB, Hook LA (2021D) SPRUCE CO<sub>2</sub> and H<sub>2</sub>O Data Beginning In 2015. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.092/1784060</u>
- Hanson PJ, Phillips JR, Brice DJ, Hook LA (2018Da) SPRUCE Shrub-Layer Growth Assessments in S1-Bog Plots and SPRUCE Experimental Plots beginning in 2010. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.052/1433837</u>
- 20. Hanson PJ, Phillips JR, Brice DJ, Hook LA (2018Db) SPRUCE Bog Surface Elevation Assessments with SET Instrument Beginning in 2013. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.055/1455014
- 21. ##Hanson, PJ, Phillips JR, Riggs JS, Nettles WR (2017D) SPRUCE Large-Collar in Situ CO<sub>2</sub> and CH<sub>4</sub> Flux Data for the SPRUCE Experimental Plots: Whole-Ecosystem-Warming. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <a href="https://doi.org/10.3334/CDIAC/spruce.034">https://doi.org/10.3334/CDIAC/spruce.034</a>
- 22. Hanson PJ, Phillips JR, Riggs JS, Nettles WR, Todd DE (2014D) SPRUCE Large-Collar in Situ CO<sub>2</sub> and CH<sub>4</sub> Flux Data for the SPRUCE Experimental Plots. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.006</u>.
- 23. ##Hanson PJ, Phillips JR, Wullschleger SD, Nettles WR, Warren JM, Ward EJ (2018Dc) SPRUCE Tree Growth Assessments of Picea and Larix in S1-Bog Plots and SPRUCE Experimental Plots beginning in 2011. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.051/1433836</u>
- 24. Hanson PJ, Phillips JR, Nettles WR, Pearson KJ, Hook LA (2020D) SPRUCE Plot-Level Water Table Data Assessments for Absolute Elevations and Height with Respect to Mean Hollows Beginning in 2015. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.079/1608615</u>
- 25. Hanson PJ, Riggs JS, Dorrance C, Nettles WR, Hook LA (2015D) SPRUCE Environmental Monitoring Data: 2010-2016. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. doi: http://dx.doi.org/10.3334/CDIAC/spruce.001. (Includes recent additions of annual data files.)
- 26. Hanson PJ, Riggs JS, Hook LA, Nettles WR, Dorrance C (2015D) SPRUCE S1-Bog Phenology Movies, 2010-2106. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. http://dx.doi.org/10.3334/CDIAC/spruce.011.
- 27. Hanson PJ, Riggs JS, Nettles WR, Krassovski MB, Hook LA (2015D) SPRUCE Deep Peat Heating (DPH) Environmental Data, February 2014 through July 2105. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. http://dx.doi.org/10.3334/CDIAC/spruce.013
- 28. ##Hanson, P.J., Riggs, J.S., Nettles, W.R., Krassovski, M.B., Hook, L.A. (2016D) SPRUCE Whole Ecosystems Warming (WEW) Environmental Data Beginning August 2015. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.3334/CDIAC/spruce.032
- 29. Haynes KM, Mitchell CPJ, Kolka RK (2019D) **SPRUCE Total Gaseous Mercury Fluxes and Peat Mercury Concentrations, 2014-2015.** Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.066/1512288</u>

- 30. Hofmockel KS, Chen, J, Hobbie EA (2016D) SPRUCE S1 Bog Pretreatment Fungal Hyphae Carbon and Nitrogen Concentrations and Stable Isotope Composition from In-growth Cores, 2013-2014. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, Oak Ridge, Tennessee, USA. <u>http://dx.doi.org/10.3334/CDIAC/spruce.025</u>
- 31. Hopple AM, Pfeifer-Meister L, Zalman CA, Keller JK, Tfaily MM, Wilson RM, Chanton JP, Bridgham SD (2019D) SPRUCE Does dissolved organic matter or solid peat fuel anaerobic respiration in peatlands? Supporting Data. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. https://doi.org/10.25581/spruce.062/1500027
- 32. Iversen CM, Hanson PJ, Brice DJ, Phillips JR, McFarlane KJ, Hobbie EA, Kolka RK (2014D) SPRUCE Peat Physical and Chemical Characteristics from Experimental Plot Cores, 2012. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.005</u>.
- 33. Iversen CM, Brice DJ, Childs J, Vander Stel HM, Salmon VG (2021D) SPRUCE S1 Bog Production of Newly-Grown Fine Roots Assessed Using Root Ingrowth Cores in 2013. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.25581/spruce.091/1782483</u>
- 34. Iversen CM, Childs J, Norby RJ, Garrett A, Martin A, Spence J, Ontl TA, Burnham A, Latimer J. (2017D) SPRUCE S1 Bog fine-root production and standing crop assessed using with minirhizotrons in the Southern and Northern ends of the S1 Bog. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.019</u>.
- 35. Iversen CM, Garrett A, Martin A, Turetsky MR, Norby RJ, Childs J, Ontl TA (2017D) SPRUCE S1 Bog tree basal area and understory community composition assessed in the Southern and Northern ends of the S1 Bog. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. http://dx.doi.org/10.3334/CDIAC/spruce.024.
- 36. ##Iversen CM, Latimer J, Burnham A, Brice DJ, Childs J, Vander Stel HM (2017D) SPRUCE plantavailable nutrients assessed with ion-exchange resins in experimental plots, beginning in 2013. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.036</u>.
- Iversen CM, Ontl TA, Brice DJ, Childs J (2017D) SPRUCE S1 Bog plant-available nutrients assessed with ion-exchange resins from 2011-2012 in the Southern end of the S1 Bog. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.022</u>.
- 38. Jensen, AM, JM Warren, PJ Hanson, J Childs and SD Wullschleger. (2015D) SPRUCE S1 Bog Pretreatment Photosynthesis and Respiration for Black Spruce: 2010-2013. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.007</u>
- 39. Jensen AM, Warren JM, Hook LA, Wullschleger SD, Brice DJ, Childs J, Vander Stel HM (2018D) SPRUCE S1 Bog Pretreatment Seasonal Photosynthesis and Respiration of Trees, Shrubs, and Herbaceous Plants, 2010-2015. Oak Ridge National Laboratory, TES SFA, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>https://doi.org/10.3334/CDIAC/spruce.008</u>
- 40. Kluber LA, Allen SA, Hendershot JN, Hanson PJ, Schadt CW (2017D) SPRUCE Deep Peat Microbial Diversity, CO<sub>2</sub> and CH<sub>4</sub> Production in Response to Nutrient, Temperature, and pH Treatments during Incubation Studies. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S. Department of Energy, Oak Ridge, Tennessee, U.S.A. <u>http://dx.doi.org/10.3334/CDIAC/spruce.040</u>
- 41. Kluber LA, Phillips JR, Hanson PJ, Schadt CW (2016D) SPRUCE Deep Peat Heating (DPH) Peat Water Content and Temperature Profiles for Experimental Plot Cores, June 2014 through June 2015. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, U.S.

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