# **2022 LTER Standard Sampling Report**

# Main Cropping System Experiment (MCSE)

### Soil

Soil was regularly sampled on the LTER to a depth of 25cm at each of 5 stations in all replicates (R) of all treatments (T) 1-8, CF, DF, and SF. Samples were taken once or twice each month, for a total of twelve times between March 30 and November 9. Soil was sampled using a 2cm diameter push corer when soil moisture conditions allowed. Two cores were taken from each station with all 10 cores composited for each plot. Bucket augers, with 5cm diameter, were used when soil was too dry for push coring on June 22. In addition, T3 and T4 individual stations and prairie strip soil was sampled using push corers three times in 2022 (May 2, July 18, September 28). Inorganic nitrogen extractions and soil moisture determinations were done on subsamples from all soil sampled. Analysis of the extracts for nitrate and ammonium should be completed by Alyssa Trethewey during winter 2022-2023.

The April 25 soil samples and May 2 T3, T4 station and prairie strip soil samples were air-dried and archived. A fresh subsample from each of the July 13 soil samples and July 18 T3, T4 station and prairie strip soil samples was incubated in the field for 21 days and then inorganic nitrogen extracted to evaluate mineralization potential.

A post-harvest extensive grid soil sampling was done in all replicates of T1-6 and T8NT October 3-4. This soil was air-dried and, along with soil from T3 and T4 station and prairie strip (sampled 9/28), T7 (8/2), and DF (10/18), sent to both the MSU Soil and Plant Nutrient Lab and A&L Great Lakes Laboratories for analysis in November 2022. This was done to compare analysis results. The MSU lab closed at the end of 2022, so future analysis will likely be done at A&L. The subsamples sent to the MSU lab underwent their standard analysis which includes pH, lime requirement, P, K, Ca, Mg, and estimated cation exchange capacity (CEC). Analysis of the subsamples sent to A&L for their S1 soil analysis package included pH, buffer pH, P, K, Ca, Mg, CEC, and percent base saturation of cation elements.

Deep soil cores (1.2m depth, 7.6cm diameter) were sampled from all 55 MCSE plots between August 12 and November 11. Two cores were taken from each station. Core 1 was sampled at about 0.7m east and 4m south of each station while core 2 was sampled at about 0.7m east and 2m south of each station. With two cores per station, from all five stations in every plot, there were 550 total cores sampled. The cores were stored in a walk-in refrigerator until they can be cut open and the soil sectioned into 0-10, 10-25, 25-50 and 50-100cm depths. From each section, soil moisture, bulk density, root mass, total carbon and nitrogen, and carbon fraction data will be collected. An oven-dried subsample of all sections for all cores will archived.

# Gas

Greenhouse gases (N<sub>2</sub>O, CO<sub>2</sub>, CH<sub>4</sub>) were sampled throughout the LTER using cylindrial static chambers between April 7 and November 3. With a sample frequency of once or twice each month, gas samples were taken eleven times during the 2022 field season. MCSE plots that were sampled include T1-7 R1-4, as well as the fertilized, untilled microplots in T7 R1-4 and prairie strips in T3, T4 (see MCSE Microplot section). Static chambers were also sampled in T8 R1-4 and all replicates

of successional and forested sites, CF, DF, and SF. Sampling in the forest sites included the Nitrogen Deposition Study microplots. The MCSE (including T8) was sampled in the morning and forest sites sampled in the afternoon of the same day. Winter sampling began on December 7 and should continue monthly through March 2023. This sampling includes only T1-7 and DF. All chamber samples were put into Labco exetainers and transported back to the lab for analysis. They were analyzed for nitrous oxide, carbon dioxide and methane by Kevin Kahmark using the Agilent/Gerstel automated GC in Academic 330.

Soil temperature and soil samples to determine moisture content were taken near each chamber when gas was sampled during the field season. HOBO dataloggers were not used in 2022 to measure air temperatures.

# Water

Soil water samplers, or lysimeters, were used to collect water from the soil at about a 1m depth in T1-7 R2-4 and all replicates of CF, DF, and SF. Samplers were evacuated, and leachate collected once or twice each month. Twelve collections were made between March 17 and November 10. The volume of leachate collected was recorded. When 20mL or more of leachate was collected, the sample was filtered and frozen for analysis and archive. Analysis for nitrate and ammonium of all stored leachate samples should be completed by Alyssa Trethewey during winter 2022-2023.

# Plants

Plant biomass samples were taken from plots prior to tillage, harvest, weed-control measures and at peak biomass. Plant species separations were completed for most but not all samplings. The understory in T5 was sampled June 24 and 27, ahead of mowing between poplar tree rows. Wheat was sampled ahead of harvest in T1-4 on July 7-8 (T1, T2) and July 11-12 (T3, T4). At peak biomass between August 9 and September 30, plant samples were taken from T8, T7, prairie strips in T3 and T4, T6 and then SF (in that order). Ahead of mowing to control weeds in T4, on September 8, plants were sampled at the approximate mowing height of 6 inches. Poplar leaf litter was collected in ground traps from August 5-November 29. Leaf litter was collected in traps in CF, DF, and SF between September 15 and December 14. Leaf litter traps were taken from all T7 plots. All plant samples were dried at 60°C for at least 48 hours. All dried biomass will be weighed, ground and archived. Subsamples will be analyzed for carbon and nitrogen by Stacey VanderWulp.

In all replicates of CF and DF, all trees with a dbh (diameter at breast height) greater than or equal to 5cm are marked with a unique numbered tag. The dbh of all tagged trees was measured in January and February 2022. The diameter of the leading stem of ten randomly chosen poplar trees in each T5 plot was measured at 15cm above the ground on December 5.

All replicates of T7 were burned on March 29 to help control woody growth. To further control woody plant species, herbicide was applied to the basal bark of black locust trees, and to a lesser extent, sumac and weedy poplar species. Mark Hammond and Jamie Smith applied the herbicide obtained from Stu Bassett (KBS Grounds) between April 20 and May 4, 2022 using specialty hand-pump sprayers. The herbicide was a mixture of Garlon 4 (20% by volume) and Basal oil (80% by volume). The dead woody stems were then cut near ground-level later in May and left where ever they fell. A gas-powered weed whip with a metal toothed blade attachment was used.

Switchgrass stand frequency was determined in May with stand counts done in all replicates of T6.

Yield data was collected from all mechanically harvested plots.

The Basso Lab used a drone (DJI M600 Pro) to fly from May to November 2022. Approximately every 2 weeks, at least once per month, LTER MCSE T1-7 were flown. The UAV flew at 122m (400ft) and was equipped with a visual camera and Micasense Altum-PT sensor.

### Insects

The Landis lab began to phase responsibility for monitoring generalist insect predators to the lab of Christie Bahlai (Kent State University). Sampling continued to monitor spatial and temporal dynamics using the 2019 protocol, https://lter.kbs.msu.edu/protocols/192. Coordination of that research was done by Stacey VanderWulp through communication with Doug Landis and Julia Perrone (KSU). Weekly surveys were completed by two temporary technicians. Being a wheat year, T-posts and sticky cards were installed beginning May 16 and weekly sampling started on May 23. Sampling concluded on August 22. Agronomic work occasionally interrupted the survey, but T-posts were removed and replaced in a timely fashion and noted on datasheets.

Monitoring was conducted by attaching one yellow sticky card (Great Lakes IPM, PHEROCON AM No-Bait Traps) to a T-post about 1.2m above the ground at each of the five stations within each replicate of T1-T7 and the nine forest plots.

This was the fourth year for surveying insects within the prairie strips of T3 and T4. The survey is conducted concurrently with the main survey, but data are recorded separately. The prairie strip sticky cards are located at the 0-meter mark (strip center) on the north, central, and south transects through all replicates of these two treatments.

The target taxa consisted of 14 common to rare ladybug species (Order: Coleoptera, Family: Coccinellidae); soldier beetles (Cantharidae); fireflies (Lampyridae); scorpion flies (Order: Mecoptera); and other non-target generalist predators (e.g. Twenty-spotted ladybug).

A 22-foot-tall aphid suction tower onsite is part of a network of towers used to monitor regional aphid migration patterns in the Midwest. The fan on the tower was turned on and the first collection cup deployed on April 15. The collection cups were collected and replaced every Friday through October 21 at which time the tower was turned off. The samples were mailed to the University of Illinois at Urbana-Champaign for analysis.

### **MCSE Microplot Experiments**

**Prairie strips (T3 R1-6, T4 R1-6):** Planted with a 22 species prairie mix in 2019, these are 15-footwide strips down the center of each T3 and T4 plot. On April 12, the strips were burned. There was 10' at the north end of each strip that was left unburned. Non-destructive plant species composition data, not percent cover, was collected from five 1m<sup>2</sup> areas in each prairie strip by the Haddad lab between June 28 and August 12. Between August 26 and September 2, at peak growth in the prairie strips, three plant samples were collected from each strip and sorted to species. T3 and T4 individual station and prairie strip soil was sampled three times in 2022 (May, July, September), see MCSE soil section. Surface soil sampled on May 2 was air-dried and archived. Soil from September 28 was sent for analysis, see MCSE soil section. Greenhouse gases were sampled eleven times in the prairie strips, as detailed in MCSE Gas section above.

**Brome grass (T6 R1-6):** When the main T6 plots were planted to switchgrass in June 2019, a 30-foot-wide by 30-foot-long area in the northwest corner of each plot was left unplanted. Brome grass was planted there September 22, 2020. Adjacent to the brome grass microplot, to the east, is a microplot of the same size. That area is planted to switchgrass and remains unfertilized when the main plot receives fertilizer. Yield data was collected from mechanical harvest of a strip through the unfertilized microplots.

**Disturbance/Fertilization Microplots (T7 R1-6):** Started in 1989, these microplots are in the northwest corner of each T7 plot. There are four microplots in each plot; each microplot measures 5x5m. They combine disturbance/tillage and nitrogen fertilization in a full factorial design. Greenhouse gases were sampled eleven times in the untilled, fertilized microplots of T7 R1-4, as detailed in MCSE Gas section above.

Fertilizing and plant sampling was led by Mark Hammond, working with Jen Lau. The fertilized microplots received nitrogen in the form of urea on June 30. This was delayed a bit longer than normal to wait for Jose Waterton's experiment's field plants to gain size. The untilled microplots were sampled for aboveground biomass (ANPP) the week of August 17. The tilled microplot ANPP was sampled near its peak the week of September 21. A reason for the difference in sample dates was to help disperse sampling across a greater time frame while integrating REX ANPP harvest. Also, the decision was made to sort to species the untilled plots. This requires about 80 to 100 hours of labor, and the aim was to finish this before harvesting REX. It was decided not to sort to species the tilled microplots because that was done last year (2021). The general goal is to sort to species at least once every 5 years, while it would be nice to have both tilled and untilled plots. Thus, not requiring sorting in the same year.

Rainfall Exclusion Experiment (REX), T1 R1-4, T2 R1, 3, 4 and 6, T7 R1-6: Sampling summarized in a separate document.

# Nitrogen Deposition Study

Nitrogen is applied to subplots within each of the three replicated sites of CF, DF, and SF. Fertilizer solutions were applied to the 1F, 3F and 10F subplots in this study three times in 2022. Urea (46% N) was applied on April 19, July 26-27 and October 24-25. Rates of fertilization are  $1\text{gN/m}^2/\text{year}$  for the 1F subplots,  $3\text{gN/m}^2/\text{year}$  for the 3F subplots, and  $10\text{gN/m}^2/\text{year}$  for the 10F subplots. Gas was sampled on a routine basis in this study; see the Gas section under MCSE.

### **Resource Gradient Experiment**

Soil samples were not taken in 2022. Sampling is planned for fall of odd-numbered years.

Soil water samplers were not sampled here in 2022. The LTER Resource Gradient experiment automated trace gas system remained dormant in 2022.

Yield data was collected from mechanical harvest. A subsample of the wheat grain and stover was collected from each plot at the time of harvest. Those subsamples were dried and will be ground, archived and analyzed for carbon and nitrogen by Stacey VanderWulp.

## **Interaction Experiment**

Mesocosms installed in 2018 were used to help quantify annual nitrogen inputs from fixation in untilled, unfertilized switchgrass plots 3 and 8. In June and July 2022, 15N<sub>2</sub> isotopic gas additions were made to help answer fixation questions on three mesocosm footprints in plots 3 and 8 (six total) and 13CO<sub>2</sub> isotopic gas for DNA stable isotope probing in one mesocosm in plots 3 and 8 (two total). Additionally, two mesocosms were used as controls and one mesocosm for testing. A total of eleven mesocosms were used during this phase of the project. No mesocosms in plot 6 were used. GPS coordinates for each mesocosm sull be collected in 2023. No rainout shelters were deployed in 2022 except as covers over the mesocosms during testing. This work was led by Sarah Roley and Carmella Vizza.

Switchgrass stand frequency was determined in May with stand counts done in all plots 1-16.

Yield data was collected from mechanical harvest. A subsample of the harvested biomass was dried and will be ground and analyzed for carbon and nitrogen by Stacey VanderWulp.

# **Cellulosic Biofuel Experiment**

Yield data was collected from mechanical harvest.

### **Biodiversity Study**

This experiment was retired after harvest in 2019 and planted to sorghum sudangrass in 2020. There was no data collected from this study after 2020.

Written by Stacey VanderWulp with contributions from Christie Bahlai, Mark Hammond, Kevin Kahmark, Jamie Smith, Ruben Ulbrich and Dave Weed

# Agronomic Soil Analysis

Experiment	2008	2009	2010	2011	2012	2013	2014	2015
LTER MCSE								
LTER Resource Gradient								
GLBRC BCSE main								
GLBRC BCSE micro								
GLBRC BCSE deep core								
GLBRC Scale-up								
GLBRC MLE								
Experiment	2016	2017	2019	2010	2020	2021	2022	
Experiment	2010	2017	2010	2019	2020	2021	2022	
LTER MCSE	2010	2017	2010	2019	2020	2021	2022	
••	2010	2017	2018	2019		2021	2022	
LTER MCSE		2017		G1-3	G1-3		G1-3	
LTER MCSE LTER Resource Gradient			2018					
LTER MCSE LTER Resource Gradient GLBRC BCSE main				G1-3	G1-3			
LTER MCSE LTER Resource Gradient GLBRC BCSE main GLBRC BCSE micro				G1-3	G1-3 G1-3			
LTER MCSE LTER Resource Gradient GLBRC BCSE main GLBRC BCSE micro GLBRC BCSE deep core				G1-3 G1-3	G1-3 G1-3		G1-3	

# **Archived Material**

Experiment	sample type	1988	1989	1990	1991	1992	1993	1994	1995	1996
LTER MCSE	leachate									
LTER MCSE	plants									
LTER MCSE	T7 microplot plants									
LTER MCSE	surface soil									
Experiment	sample type	1997	1998	1999	2000	2001	2002	2003	2004	2005
LTER MCSE	leachate									
LTER MCSE	plants									
LTER MCSE	T7 microplot plants									
LTER MCSE	surface soil									
LTER MCSE	T7 microplot surface soil									
LTER MCSE	deep core soil									
LTER Biodiversity Study	surface soil									
Experiment	sample type	2006	2007	2008	2009	2010	2011	2012	2013	2014
LTER MCSE	leachate									
LTER MCSE	plants									
LTER MCSE	T7 microplot plants									
LTER MCSE	surface soil									
LTER MCSE	T7 microplot surface soil									
LTER MCSE	deep core soil									
LTER Biodiversity Study	surface soil									
LTER Resource Gradient	surface soil									
Experiment	sample type	2015	2016	2017	2018	2019	2020	2021	2022	
LTER MCSE	leachate									
LTER MCSE	plants									
LTER MCSE	surface soil					T0.4	<b>B</b> . <b>P</b> . <b>P</b> .			
LTER MCSE	deep core soil					T3,4 prairie strips	Biodiversity Study			
LTER Biodiversity Study	surface soil						deep core			
LTER Resource Gradient	surface soil									