Utilizing ecosystem function multifunctionality to assess supporting ecosystem services of three land-use types in northwestern Virginia

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Abstract

Supporting ecosystem services are vital in the maintenance of other ecosystem services, yet are often overlooked due to their less obvious and longer-term impacts on human society. They also do not have agreed upon metrics, making them difficult to quantify and apply to environmental decision-making. A suite of aboveground, belowground, and aboveground-belowground interface ecosystem properties, processes, and functions that inform the supporting services of soil formation, nutrient cycling, and primary productivity were measured in three land-use types under variable management in northwestern Virginia over the course of two growing seasons (2017 and 2018): an unmanaged early successional field, a semi-managed native prairie, and a heavily managed agricultural field. The ecosystem functions measured (net nitrogen (N) mineralization, soil respiration, primary productivity, decomposition, and N leaching) were compared across land-use types and then were applied to an ecosystem function (EF)-multifunctionality framework based on associations with carbon, nitrogen, and supporting services. During the 2017 and 2018 growing seasons, NNM did not differ among fields. Decomposition was greatest in the early successional field in both 2017 and 2018 (3.73 ± 1.08 year^{-1} and 2.70 ± 0.48 year^{-1}, respectively). Primary productivity was greatest in the agricultural field (1250 ± 155 g m^{-2}) in 2017 when it was planted; in 2018 when the agricultural field was left fallow, primary productivity was greatest in the early successional field and native prairie in 2018 (265 ± 42 g m^{-2} and 262 ± 31 g m^{-2}, respectively). During 2018, mean soil respiration was greatest in the early successional field (0.24 ± 0.03 g CO_2 m^{-2} h^{-1}). The native prairie, with higher belowground root mass, had the lowest leaching potential index in the top 20 cm of soil. The sum of C and N multifunctionality differed in both years; the agricultural field had the greatest level of multifunctionality when planted but least when not planted. The native prairie and early successional field appear to be comparable in terms of their total C and N multifunctionality, though N multifunctionality was greatest in the early successional field in 2018. While EF-multifunctionality associated with supporting ES was greatest in the agricultural field in 2017 when it was planted, its mean multifunctionality over both years balanced out to near zero (near the overall mean multifunctionality across all fields), suggesting that the agricultural field provides a suite of supporting ES when planted, but when accounting for interannual variability associated with crop rotations, it does not appear to consistently provide those supporting ES. The EF-multifunctionality of the native prairie over both years was several standard deviations

below the overall mean, suggesting that it provides supporting ES at a lower rate than the other land-use types. The early successional field demonstrated the highest mean EF-multifunctionality across both growing seasons, suggesting that it consistently provides a suite of supporting ES. Including the ecological cost of human management alters the provision of supporting services; the agricultural field and native prairie failed to demonstrate greater overall multifunctionality than the early successional field in order to compensate for their ecological cost of management, so they do not appear as valuable in terms of the net supporting services they provide. This study is the first of its kind to implement an EF-multifunctionality approach to inform supporting ES, while also accounting for disservices and cost of management. Assessing multifunctionality with z-scores raises the question of whether the importance and variation can be equated across all functions. Applying multifunctionality in this way unveiled some shortcomings of the framework: equating importance and variation across all functions as well as the undervaluation of conservative nutrient cycling. This work is, however, a step toward elucidating the difficult task of quantifying supporting services in order to inform sustainable land-cover decisions.

Introduction

Ecosystem services (ES) are generally defined as benefits people derive from ecosystems and are commonly classified into four categories: provisioning services, regulating services, supporting services, and cultural services. Of these four categories, some benefit humanity in direct, obvious ways, while some are less apparent. Provisioning services and cultural services both provide direct, noticeable benefits to societies, while regulating services and supporting services benefit societies indirectly and are therefore less noticeable. Supporting services in particular are often overlooked due to their subtle, longer-term impacts on human society. They are particularly difficult to quantify, and therefore are less considered in environmental decision-making. They are, however, vital in the maintenance of other services, providing the foundational inputs for final goods and services, and therefore, crucial for life on Earth and humanity's continued existence. The overall focus of this thesis is to acknowledge, elucidate a framework for quantification, and begin to correct the undervaluation of supporting services. Supporting services were also an attractive research topic as they are more objective than the other types of ES, the measurement and evaluation of which can be much more subjective.

Conversations around the ES framework frequently point out the unclear definitions of, and distinctions among, processes, functions, and services. This project sets a clear definition of processes (fluxes of energy and matter among pools within and between ecosystems that are often considered to be a single transformation; e.g.: photosynthesis, gross primary productivity, autotrophic and heterotrophic respiration, gross N mineralization, and litterfall) and functions (consequence of more than one process; e.g.: decomposition, ecosystem respiration, net primary productivity, net N mineralization, N leaching, and soil respiration). I also acknowledge two commonly mentioned gaps in the current ES framework: functions with detrimental impacts and the ecological cost of human management. Though there are multiple ways to measure supporting services, this is the first approach to utilize a suite of functions and multifunctionality analysis to inform supporting services.

I measured and compared a suite of properties, processes, and functions that relate to supporting services in three land-use types in northwestern Virginia at Blandy Experimental Farm over the course of two growing seasons (2017 and 2018): an early successional field, a native prairie, and an agricultural field. I also applied a suite of five ecosystem functions, net nitrogen mineralization (NNM), soil respiration, primary productivity, decomposition, and nitrogen (N) leaching, to a multifunctionality framework in two ways. In Chapter 1, an ecosystem function multifunctionality analysis was utilized, and functions were organized into two groups: carbon-based (C multifunctionality) and nitrogen-based (N multifunctionality). In Chapter 2, multifunctionality analysis was utilized to inform the supporting ES of soil formation, nutrient (N) cycling, and primary production, by including all functions associated with each supporting service. The ecological cost of management and unwanted impacts of ecosystem functions were also considered.

This study applies the ecosystem function multifunctionality framework in a novel way to inform supporting ES, while also considering shortcomings previously acknowledge in the ES literature: disservices and cost of management. Figure 1 is a graphical representation of this thesis as a whole.

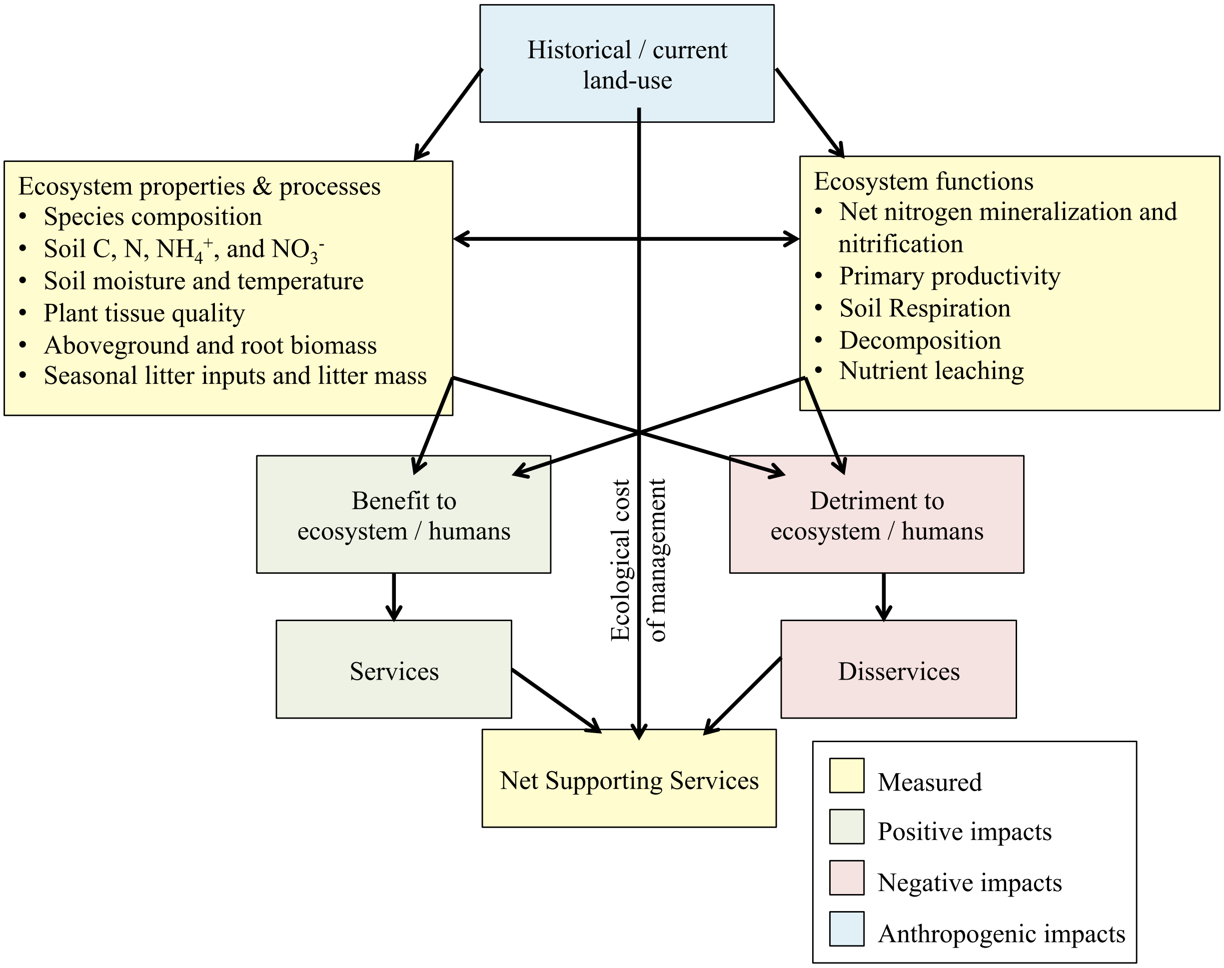

Figure 1. A graphical representation of Chapters 1 and 2 of this thesis. Chapter 1 predominantly covers the top 2 yellow boxes, "Ecosystem properties & processes" and "Ecosystem functions" and considers whether they are beneficial or detrimental (the green and red boxes below, respectively). Chapter 2 acknowledges the blue box of anthropogenic impacts associated with "historical / current land-use" as well as the positive and negative impacts, which, when all taken into consideration, yield "net" supporting services.

Chapter 1: Utilizing ecosystem function multifunctionality to assess benefits derived from a suite of functions among land-use types

1. Abstract

Supporting ecosystem services are vital in the maintenance of other ecosystem services, yet are often overlooked due to their less obvious and longer-term impacts on human society. A suite of ecosystem properties, processes, and functions that inform the supporting services of soil formation, nutrient cycling, and primary productivity were measured in three land-use types in Virginia over the course of two growing seasons (2017 and 2018): an early successional field, a native prairie, and an agricultural field. A suite of five ecosystem functions, net nitrogen mineralization (NNM), soil respiration, primary productivity, decomposition, and nitrogen (N) leaching, were compared across the land-use types, using a variation on the ecosystem function-multifunctionality framework, in which carbon and nitrogen multifunctionality were estimated based on normalized means of multiple carbon-based and nitrogen-based functions, respectively.

During 2017, NNM did not differ among fields, but decomposition was greatest in the early successional field (3.73 ± 1.08 year^{-1}). Primary productivity was highest in the agricultural field (1250 ± 155 g m^{-2}). During 2018, mean soil respiration was greatest in the early successional field (0.24 ± 0.03 g CO_2 m^{-2} h^{-1}). Decomposition was 2.70 ± 0.48 year^{-1} in the early successional field and 1.05 ± 0.12 year^{-1} in the native prairie, and the agricultural field lacked aboveground litter inputs. Primary production was greater in the early successional field and native prairie (265 ± 42 g m^{-2} and 262 ± 31 g m^{-2}, respectively) than in the fallow agricultural field. The native prairie, with its higher belowground root mass, had the lowest leaching potential index in the top 20 cm of soil. C multifunctionality, estimated as the sum of normalized decomposition and primary productivity minus normalized soil respiration, differed significantly among the land-use types in both 2017 and 2018. N multifunctionality, estimated as the sum of normalized net N mineralization (NNM), decomposition, primary productivity, and soil respiration minus normalized leaching potential, was significantly greater in the early successional field than the other fields in 2018 but not in 2017. Total multifunctionality differed in both years, suggesting that the agricultural field has the greatest level of multifunctionality when planted but least when not planted. The native prairie and early successional field appear to be comparable in terms of their multifunctionality. While multifunctionality does not appear to capture everything necessary for valuation, such as giving credit to more conservative nutrient cycling, this work is a step toward elucidating the difficult task of quantifying supporting services in order to inform sustainable land-cover decisions.

2. *Introduction*

2.1 Ecosystem Services

Anthropogenic changes to the world's ecosystems since the mid 20th century have outpaced those of any other point in human history. Additionally, an unsustainable standard of living and large world population have led to a growing demand for clean water, food, and fuel (Millennium Ecosystem Assessment (MEA), 2005). Society's demands have altered ecosystems globally, resulting in losses of biodiversity and changes in carbon and nutrient cycling. As humanity's extensive impacts on Earth's natural systems become more apparent, so does the importance of these systems in humanity's continued existence. The ecosystem services (ES) framework acknowledges the critical role that ecological functions play in human wellbeing (Seppelt et al., 2011) and acts as a bridge between humans and nature (Braat and de Groot, 2012), bringing together multiple perspectives to shape strategies for a more sustainable society (van den Belt and Stevens, 2016).

The ES concept is relatively new, first discussed in the late 1960s by King (1966) and Helliwell (1969), who noted how nature's functions serve human society. Since its inception, the ES concept has served to acknowledge the many ways in which nature benefits society and to steer environmental policy. The MEA (2005) broadly defines ES as "the benefits people derive from ecosystems." Costanza et al. (1997) acknowledge the foundation of ecosystem functions in their definition of ES as "the benefits human populations derive, indirectly or directly, from ecosystem functions." ES are commonly classified into four categories, based on the Millennium Ecosystem Assessment's definitions: provisioning services, regulating services, supporting services, and cultural services. Provisioning services are tangible goods for human consumption, including food, fiber, fuel, and water supply. Regulating services provide benefits to humans through the control of potentially harmful or damaging ecosystem processes, including flood and disease control. Supporting services maintain the conditions for life on Earth, including soil formation, nutrient cycling, and primary production. Cultural services fulfill human societies' spiritual and recreational needs (MEA, 2005).

Of these four ES categories, some benefit humanity in direct, obvious ways, while some are less apparent. Provisioning services and cultural services both provide direct, noticeable benefits to societies, although cultural services are quite dependent on social context (MEA, 2005). Regulating services and supporting services both benefit societies indirectly and are therefore less noticeable. Regulating services assimilate waste and lead to clean air and water supplies for human populations. Supporting services are vital in the maintenance of other services; they provide the foundational inputs for the final goods and services, yet they are often overlooked due to their less obvious and longer term impacts on human society (Villamagna et al., 2013).

2.2 Supporting Services, Ecosystem Processes, and Ecosystem Functions

The Millennium Ecosystem Assessment (2005) discusses the lack of data on supporting services. Whereas provisioning services can easily be quantified, for example, as per capita food production, supporting services are often poorly described and understood. This lack of data can be attributed to the subtlety of supporting services, as well as the indirect role they play in human wellbeing. As a result, decisions are being made with greater focus on provisioning and cultural services for which benefits to society are direct, and values are easily quantifiable. Therefore, the undervaluation of supporting services, which are vital to life on Earth and humanity's continued existence, needs to be acknowledged and improved.

While soil formation, nutrient cycling, and primary production are consistently categorized as supporting services, they are difficult to measure. Their underlying components,

Table 1. A summary of conflicting definitions and examples of processes and functions provided by commonly cited authors in the context of ecosystem services.

Author	Processes	Functions
Costanza et al. (2017)	Do not explicitly differentiate between processes and functions; groups processes and functions together, under the description of "biophysical relationships that exist regardless of whether or not humans benefit"	
Lovett (2006)	Fluxes of energy and matter among pools within and between ecosystems *ex: primary and secondary production, decomposition, autotrophic and heterotrophic respiration, evapotranspiration*	Attributes related to ecosystem performance, resulting from one or more ecosystem processes *ex: nutrient cycling, productivity, nutrient leaching*
Manning et al. (2018)	Do not define processes but do acknowledge that there is not consensus on whether processes should be categorized as functions.	Extension of ecosystem processes
Pettorelli et al. (2017)	Accept Lovett's (2006) definition of ecosystem processes. *ex: Mineralization (further distinguished as an* ecological *process), primary production, decomposition*	Accept Lovett's (2006) definition of ecosystem functions, adding "direct and indirect benefits of ecosystem processes for a range of species, including humans" *ex: Soil and sediment formation and retention*

processes and functions, are more easily quantifiable, but they are inconsistently defined and differentiated in the literature, and there is little consensus on specific examples of processes and functions (Table 1). In part as a result of this lack of consensus, processes and functions are not frequently explicitly integrated into analyses of ES.

The differentiation between process and function used in this study is derived from Lovett's (2006) definitions; I define processes as fluxes of energy and matter among pools within and between ecosystems that are often considered to be a single transformation, whereas functions are the consequence of more than one process. Based on this definition of ecosystem processes, examples would include photosynthesis, gross primary productivity, autotrophic and heterotrophic respiration, gross N mineralization, and litterfall. Examples of ecosystem functions would include decomposition, ecosystem respiration, net primary productivity, net N mineralization, N leaching, and soil respiration.

2.3 Ecosystem Function Multifunctionality

While ecosystem processes and functions contribute to ES, they are not necessarily services in and of themselves (De Groot, 2002). They are, however, critical to the supply of ES (Costanza et al., 2017). In addition to the lack of distinction made between ecosystem process and function, the distinction between ecosystem function and service is not always made within the ES conversation (Barot, 2017). Ecosystem processes and functions are linked through interactions and shared drivers in complex, non-linear ways, which may or may not lead to ecosystem services (Manning et al., 2018). Some functions have positive consequences for other functions, other ecosystems, and human societies and therefore contribute to services. Some functions, however, are detrimental to other functions, other ecosystems, or to human societies and can be considered disservices. Therefore, equating function and service, as many previously have, may be misleading.

The impacts of biophysical drivers, such as climate, soil conditions, and species diversity on *individual* functions is commonly quantified in a systematic and standardized way to compare among ecosystems. Processes and functions, however, are less frequently considered in concert with one another. Individual ecosystem functions, such as specific soil carbon fluxes and biomass production, can be quantified in a standardized way, but this potentially misses nuances of the interactions and tradeoffs among different functions and processes (Sala et al., 2000).

Ecosystem function (EF)-multifunctionality (Manning et al., 2018), the fact that ecosystems simultaneously conduct multiple functions (Byrnes et al., 2014), may be a helpful lens through which to more holistically evaluate a system's overall functionality and therefore its ability to provide supporting ES. The goal of this research was to compare a suite of biogeochemical properties, processes, and functions among different ecosystems.

3. Research Goals and Methods

3.1 Study System

Blandy Experimental Farm (BEF), a 280-hectare research facility in northwestern Virginia, contains multiple land-uses and therefore provides the opportunity to fill some of the current gaps in the ES conversation. Ecosystem properties, processes, and functions that underlie supporting services were measured and compared across three different land-use types under variable human management. Ecosystem properties, including species composition, plant tissue quality, soil organic matter, soil moisture and temperature, soil inorganic ammonium (NH_4^+) and nitrate (NO_3^-), aboveground biomass, root biomass, and mass of the litter pool, were measured in three different land-use types: an agricultural field (cultivating millet for the duration of the first growing season of the study and left bare for the duration of the second growing season), a restored native C_4 tallgrass prairie, and an approximately 16-year-old early successional field. Additionally, ecosystem processes and functions of net nitrogen (N) mineralization and nitrification, seasonal litter inputs, soil respiration, aboveground primary productivity, and N leaching were estimated over the course of 1-2 growing seasons.

3.2 Research Objectives

It is imperative to determine productive and helpful ways to measure and evaluate ecosystem processes, functions, and their drivers, in order to improve assessments of ES (Costanza et al., 2017). The multiple land-use types at BEF under the same climate and geological conditions provide an opportunity to comprehensively do so. The questions this research aims to answer are:

1. How do individual properties of soil moisture and temperature, soil organic matter, root biomass, plant tissue quality, live and standing dead biomass, species composition and diversity, litter mass, and inorganic NH_4^+ and NO_3^- vary across land-use types?
2. How do individual processes and functions of seasonal litter inputs, net nitrogen mineralization (NNM), net nitrification, soil respiration, primary productivity, decomposition, and nutrient leaching vary across land-use types?
3. Can utilizing an ecosystem function multifunctionality approach provide useful information for assessing and comparing supporting ES among land-use types?

While rigorously quantifying supporting ES is challenging, the suite of properties, processes, and functions that underlie them *can* be quantified. Likewise, assessing EF multifunctionality is not trivial, but it is a potentially worthwhile pursuit (Manning et al., 2018). A better understanding of the role of functional mechanisms in ES can improve the ability to make sustainable land-use/land-cover decisions (Power, 2010).

3.3 Study Design

Blandy Experimental Farm (BEF) is located in northwestern Virginia (39.06 ^{o}N, 79.07 ^{o}W). It has a mean annual precipitation of 975 mm, a mean annual temperature of 12 ^{o}C, and a mean maximum July temperature of 31.5 ^{o}C. The facility is underlain by limestone, dolomite, and sandstone from the Conococheague formation (Hubbard 1990).

Research plots were located in three different land use types: restored native prairie, early successional field, and agricultural field. The area of focus within each field was approximately 1 hectare. Five representative 5 m x 5 m plots were randomly chosen in each of the three fields. Within each 5 m x 5 m plot, three 1 m^2 subplots were randomly chosen for replicate sampling.

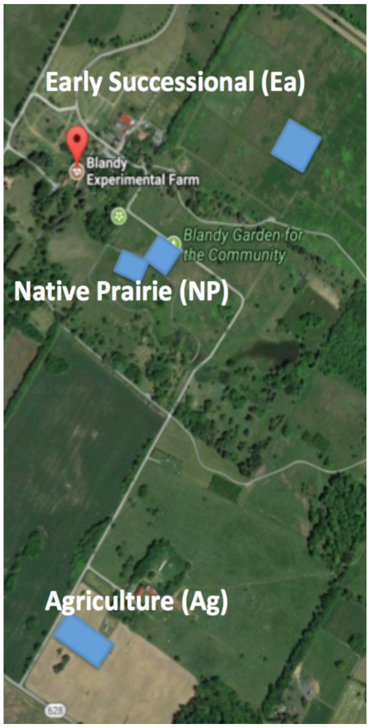

Figure 1. Five research plots, each approximately 1 ha, were located in three different land use types at BEF: native prairie (NP), early successional field (Ea), and agricultural field (Ag).

The native prairie is meadow-like, dominated by native C_4 grasses with some forbs, scattered shrubs, and a few larger trees. Though the meadow is on a mild topographic gradient, there is no significant spatial variability in soil moisture (Yeatman and Epstein, 2001). BEF planted a variety of seed combinations of native species in the prairie restoration; the areas of focus for this research were originally planted with native forbs and six native warm season grass species: *Andropogon gerardii* (big bluestem), *Bouteloua curtipendula* (sideoats grama), *Schizachyrium scoparium* (little bluestem), *Andropogon virginicus* (broom sedge), *Sorghastrum nutans* (Indian grass), and *Panicum virgatum* (switchgrass).

The early successional field is dominated by forbs and shrubs, and has many more exotic species than the native prairie. It is located on low-relief topography. Prior to the establishment of the early successional field 16 years ago, the area was used for small mammal population research. Various early successional plant species began to establish in the area following the end of the research. Common species in the early successional field include *Symphoricarpos orbiculatus* (coralberry), *Rubus spp.* (raspberry and blackberry), *Carduus acanthoides* (spiny plumeless thistle), *Solidago altissima* (goldenrod), and *Galium verum* (yellow bedstraw).

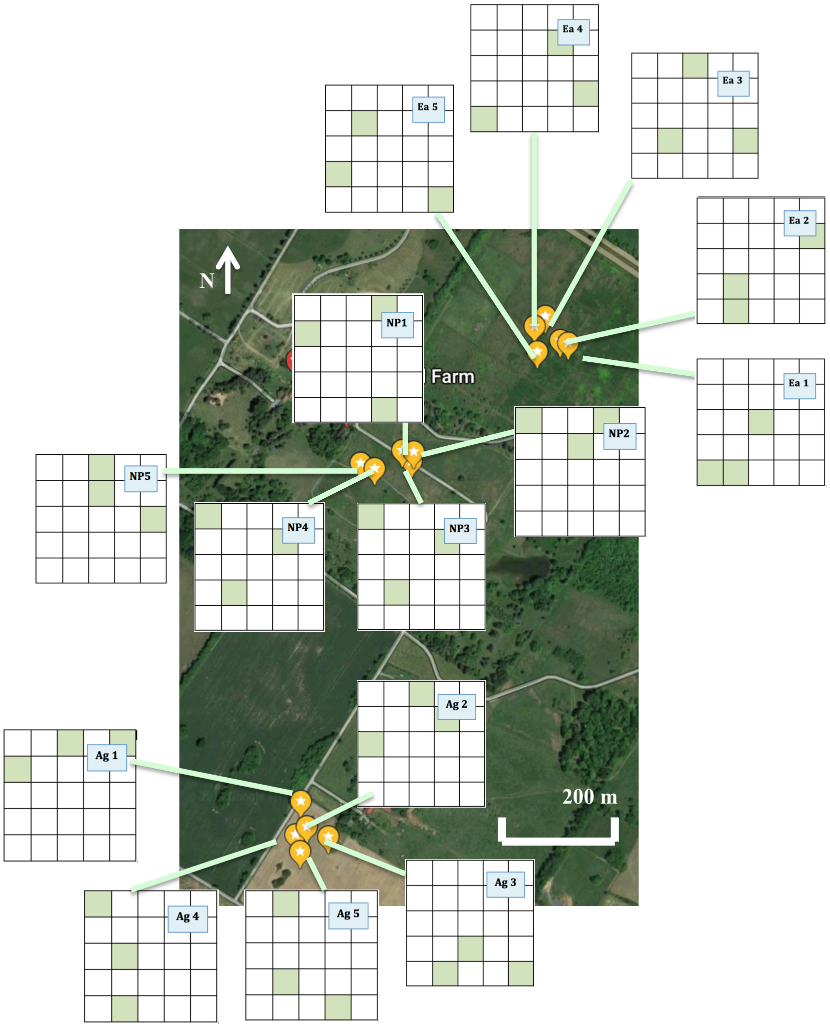

Figure 2. Each field had five 5m x 5m plots, with three randomly selected replicate 1m^2 subplots. The replicate subplots within each plot are shaded light green. Each plot is labeled in a light blue square.

Winter wheat was harvested from the agricultural field in mid-June prior to the beginning of data collection for this study in both 2017 and 2018. During 2017 a monoculture of millet was planted in mid-July following the winter wheat harvest, grown for about 50 days, and harvested at the end of August. During 2018 the field was left bare due to a lack of alignment between the farmer's schedule and weather. Data for this study were collected during both the 2017 and 2018 summer growing seasons.

3.4 Ecosystem Properties

3.4.1 Soil Moisture & Temperature

Soil moisture measurements, recorded as volumetric percent water within the top 12 cm of soil, were taken using a Hydrosense handheld time domain reflectometer probe (Campbell Scientific, Inc., Logan, UT). Soil temperature measurements were taken at a depth of 10 cm using a digital thermometer. Measurements of both were taken at multiple times throughout the summer 2018 growing season in the three replicate subplots within the five plots of the native prairie and early successional field. Because plot establishment in the agricultural field was delayed due to planting schedule, measurements were delayed at that location until late July. Local landscape-scale precipitation data were used to supplement the soil moisture and temperature dataset.

3.4.2 Soil Organic Matter

Soils were sampled to a depth of 30 cm at 10 cm increments to measure total carbon (C) and nitrogen (N), as well as soil bulk density. Soil samples were passed through a 2 mm sieve to homogenize, and rocks and roots were removed and weighed. The remaining homogenized soil sample was also weighed, and a representative subsample was taken and oven-dried until each sample reached a constant weight. The wet versus the dry weight of the subsample was used to estimate the dry weight of the full soil sample. Soil bulk density was calculated as the ratio of soil dry mass to volume by subtracting the density-based volume of rocks and roots from the volume of the entire soil core, and subtracting the mass of rocks and roots from the mass of the full soil sample. Approximately 20 mg of soil were taken from the representative oven-dried subsample for dry combustion in a CN analyzer (Flash 2000; Thermo Fisher Scientific, Inc., Milan, Italy). Using %C and %N, C:N ratios in each 10 cm incremental depth were determined, as well as absolute amounts of C and N using bulk density.

3.4.3 Root Biomass

As described above, soils were sampled to a depth of 30 cm at 10 cm increments. Soil samples were passed through a 2 mm sieve, and roots were removed, cleaned, and weighed. The total mass of roots within each 10 cm incremental depth to 30 cm was determined for each sample.

3.4.4 Plant Tissue Quality

Aboveground biomass harvesting was performed at peak seasonal biomass in each field: the early successional field in mid-August of 2017 and 2018, the native plant meadow in late August 2017 and 2018, and individual millet plants immediately before harvest of the whole field in 2017. Because the agricultural field was left bare during the summer of 2018 due to weather, there was no biomass to harvest. In the early successional field and native prairie, an undisturbed corner of each of the three 1 m^2 subplots was harvested by clipping all live and standing dead plants at the soil surface within a 0.5 m x 0.5 m quadrat (0.25 m^2). In the agricultural field, two average-sized individual millet plants were harvested from each plot.

Figure 3. Three replicate biomass samples were harvested from an undisturbed area in each 1-m^2 subplot using a 0.25m^2 quadrat. All live and standing dead biomass was clipped at the soil surface.

In the laboratory, all clipped samples of aboveground biomass were oven-dried for 72 hours at 60 °C in a forced-air oven. Standing dead material harvested from the early successional field and native prairie was separated from the live plants, and live plants were sorted into plant functional types of graminoids, forbs, and shrubs. Shrubs were then separated into woody and foliar components. From the 2017 harvested biomass, representative subsamples of foliar components from each functional type were ground. Approximately 5 mg were taken from the ground subsamples for dry combustion in a CN analyzer. Using %C and %N, the C:N ratio of live foliage of each functional type was determined.

3.4.5 Aboveground Live and Standing Dead Biomass

The range of total biomass of each plot in the early successional field and native prairie was determined by scaling up the average mass of the three 0.25 m^2 subplot samples to the 5 m x 5 m plot. Because only two millet plants could be sampled from the agricultural field, the numbers of plants within multiple 1 m^2 areas were counted, then multiplied by the mean mass of the two harvested individuals. Dry weights were recorded for the total standing dead and live material of each plant functional type for each of the three replicate subsamples from each plot in the early successional field and native prairie.

3.4.6 Species Composition and Diversity

Species composition and diversity were determined for each plot in the native prairie and early successional fields using the point-intercept sampling method (Caratti, 2006). A 5 m x 5 m grid was established with 0.5 m intervals, including plot edges, for a total of 121 points of intersection. At each of the 121 points, a dowel was dropped, and overstory and understory species touching it were identified. The total observation frequency of each species in each plot was recorded and compared to the total number of hits (the inclusion of overstory and understory species resulted in a total number of hits greater than 121; the total ranged from 196 to 286) to calculate relative frequency of each species in each plot. Species richness, Simpson's diversity index, Shannon's diversity index, and Shannon's evenness index were calculated for each plot. Observed species were separated into native and non-native groups as well as plant functional types in order to calculate the relative frequency of each (Concilio et al., 2016).

3.4.7 Litter Mass

Litter was collected from an undisturbed 0.25 m^2 corner of each of the three 1m^2 subplots in each plot at the beginning of the 2018 growing season. Each sample was oven-dried and weighed in order to estimate the litter pool, established from litter fall of multiple previous growing seasons.

3.4.8 Soil NH_4^+ and NO_3^-

An initial 5.08 cm diameter soil sample was taken to a depth of 10 cm at the start of net N mineralization and nitrification incubations and immediately frozen to minimize any microbial activity. In the laboratory, frozen soil samples were thawed and passed through a 2 mm sieve, and two field-moist 10 g soil subsamples were taken from each. One subsample was oven-dried for 48 hours at 115 ^{o}C, and the other was combined with 40 mL of 2 M KCl. The oven-dried subsamples were reweighed to determine gravimetric soil water content to adjust values to the dry weight of soil. The 2 M KCl solutions of each soil sample were agitated on a shaker at 200 rpm for one hour and then set to rest for one hour, allowing supernatant to collect at the surface. The supernatant was then passed through 11 µm medium-flow Whatman filter paper (Baer and Blair, 2008).

All samples were run through an autoanalyzer (Lachat QuikChem 8500; Hach Company, Loveland, CO) for detection of NO_3^--N and NH_4^+-N. The concentrations of NO_3^--N and NH_4^+-N were compared to the response curves from the analyzer of standards of known concentration, including pure KCl samples. Because sample concentrations often exceeded the threshold of the response curves for the analyzer, all samples were diluted with 2 M KCl to fall within the range of the response curves. The detection limit for this method was 0.005 mg/L for NO_3^--N and NH_4^+-N, so any concentration below that was assumed to be zero.

3.5 Estimated Ecosystem Processes

3.5.1 Seasonal Litter Inputs

Seasonal litter inputs were estimated based on the amount of live foliar biomass (there were no evergreen species present in the plots used in this study) in each land-use type, under the assumption that all foliar biomass would become litter at the end of a growing season. The maximum possible seasonal litter inputs were also estimated by adding the standing dead biomass to the total foliar biomass.

3.6 Measured Ecosystem Functions

3.6.1 Net N Mineralization and Nitrification

Three one-month-long net N mineralization (NNM) *in situ* resin-core incubations were conducted according to the resin-core method of DiStefano and Gholz (1986) to a depth of 10 cm; Persson and Wiren (1995) found that 78% of NNM takes place in the top 10 cm of the soil. Incubations were performed in each of the three replicate subplots in the five plots in all three land-use types. Two of the three incubations were conducted during the 2017 growing season: the first near peak plant growth from mid-June to mid-July (though the agricultural field was excluded from this incubation, as it had not been planted yet) and the second near peak biomass from mid-July to mid-August. The last incubation was conducted during the 2018 growing season near peak biomass from mid-July to mid-August.

An initial 5.08 cm diameter soil sample was taken to a depth of 10 cm at the start of each incubation and immediately frozen to minimize any microbial activity. A second soil sample was taken in a plastic 5.08 cm diameter sleeve and placed back in the ground with an anion-cation exchange resin bag attached to its base at 10 cm depth, according to *in situ* NNM incubation methods established by DiStefano and Gholz (1986) and further supported by Binkley et al. (1992), Kolberg et al. (1997), and Bhogal et al. (1999). The resin bag acted to capture any NO_3^- or NH_4^+ that leached from the soil core, which also gives an estimate of vertical nitrogen flux. The resin bags were constructed of nylon and contained a mixture of 1-tablespoon each of cation (USF C-211 resin cation, Na form) and anion (USF A-464 resin, Type I anion, Cl form) resins, allowing water percolation (Schnabel, 1983) and nutrient adsorbance to the resin surface for later extraction. Soil samples and resin bags were retrieved after one month in the field and immediately frozen to minimize further microbial activity.

Soil samples were handled using the methods outlined above in section 3.4.8, and resin bags were extracted in the same way, following a rinse using deionized Nanopure® water. Initial cores and those incubated *in situ* were used to calculate NNM rates (Robertson et al., 1999). NNM was calculated as the change in NO_3^--N and NH_4^+-N in the cores after the one-month incubation, plus any NO_3^--N and NH_4^+-N detected in the resin bag. Net nitrification was calculated as the change in NO_3^--N after the one-month incubation, plus any NO_3^--N detected in the resin bag (Chapman et al., 2013).

3.6.2 Soil respiration

In situ soil CO_2 efflux was measured with a portable infrared gas analyzer with attached soil respiration chamber (EGM-4; PP Systems, Amesbury, MA), to serve as a proxy for soil respiration. Measurements were taken mid-day between 10h and 14h once per week during May and June in the three replicate subplots within the five plots of both the native prairie and early successional field. Once plots were established in the agricultural field in late July, measurements were taken in the three replicate subplots within the five plots of each land-use type on a weekly basis through August. The time of day at which land-use types were sampled was rotated in order to eliminate any time of day sampling bias.

3.7 Estimated Ecosystem Functions

3.7.1 Primary Productivity

Peak season aboveground biomass components were used to estimate aboveground net primary productivity (ANPP), according to Briggs and Knapp (1991). The mass of all live graminoids, forbs, and the foliar component of deciduous shrubs (evergreen shrubs were absent from research plots) were used to estimate ANPP. Total herbaceous foliar biomass at the end of growing season is assumed to be equivalent to aboveground productivity. Most field measures of NPP account for only new plant biomass produced, likely underestimating NPP by about 30% (Chapin et al., 2011).

3.7.2 Litter Decomposition

The leaf litter pool, sampled at the beginning of the growing season, when compared to the estimated seasonal litter input, provided insight into the rate of aboveground litter decomposition according to the equation $k = \frac{litter\ fall}{litter\ pool}$, where *k* is the decomposition rate. The use of this equation, however, assumes steady-state conditions of the litter pool in these land-use types.

3.7.3 Nutrient leaching

DiStefano and Gholz (1986), Binkley et al. (1992), and Bhogal et al. (1999) state that resin-core incubations not only indicate NNM but also potential leaching of plant-available nitrogen. The resin bag positioned at the base of the soil cores during the *in situ* incubations in 2017 captured NO_3^- and NH_4^+ that leached from the soil core, providing an estimate of soil-controlled vertical nitrogen flux within the top 10 cm of soil.

In order to gain greater insights into leaching capacity beyond the top 10 cm of soil, soil samples were collected from each plot across 10 cm increments to a 30 cm depth. The majority of the root zone was found to be in the top 20 cm of soil in the native prairie (94% of root mass was found in the top 20 cm of soil on average, with 79% of root mass in the top 10 cm). The root zone was assumed to be largely in the top 10 cm in the early successional and agricultural field (89% and 100% of root mass was found in the top 10 cm of soil on average, respectively). Samples were collected twice during 2018: in the middle of the growing season (mid-July), then again after the end of the growing season (mid-October), as the greatest leaching tends to occur following the growing season (Chapin et al., 2011). The samples were immediately frozen to minimize any microbial activity and were treated in the laboratory in the same manner as in the NNM analysis.

A leaching potential index was then calculated using total soil NO_3^--N and NH_4^+-N quantities in each 10 cm soil layer in relation to the mass of roots within and below that soil layer. Quantities of soil NO_3^--N and NH_4^+-N were divided by the mass of roots within and below that soil layer; the greater the root mass present within and below the layer, the lower the potential for leaching. Therefore, for example, a lower leaching potential index value represents a lower potential for leaching, through a combination of lower quantities of available soil NO_3^--N and NH_4^+-N and a greater quantity of roots to inhibit leaching of reactive nitrogen.

3.8 Evaluating Multiple Functions vs. Multifunctionality

Whereas the definition of EF-multifunctionality used by Manning et al. (2018) is relatively straightforward (the ability of ecosystems to simultaneously provide multiple ecosystem functions), how to conceptualize and measure it is unresolved (Allan et al., 2015; Byrnes et al., 2014; Mastrangelo et al., 2014). One generally accepted method for quantifying EF-multifunctionality is the "averaging" approach, which takes the average of standardized values of each function (Maestre et al., 2012; Mouillot et al., 2011). The other is the "threshold" approach, which counts the number of functions that have passed a prescribed threshold (typically a percentage of the highest observed level of a given function in a study (Byrnes et al., 2014; Gamfeldt et al., 2008). Because the choice of a threshold value seems somewhat arbitrary, the "averaging" approach was used in this study to assess multifunctionality. Standardized z-scores were determined for each measured function in each plot during both the 2017 and 2018 field seasons based on the overall mean of each function across plots and years. Calculated z-scores were then used to compare individual functions across land-use types.

EF multifunctionality provides a more holistic evaluation of a system's ability to provide ecosystem functions simultaneously, which can also have implications for the system's ability to provide supporting ES. In order to determine EF multifunctionality, functions quantified were grouped into two categories: carbon (C)-based functions and nitrogen (N)-based functions. These categories embody the concepts of the three widely accepted categories of supporting services, soil formation, nutrient cycling, and primary production. This approach does, however, assume all variables are weighted equally, which may not always be the desired formulation, because certain functions may be more important in particular contexts (Manning et al., 2018).

Some functions have positive consequences for other functions, other ecosystems, and human societies and therefore contribute positively to services. Some functions, however, are detrimental to other functions, other ecosystems, or to human societies and can be considered disservices. When calculating EF-multifunctionality, the normalized rates of beneficial functions were considered positive, while the normalized rates of detrimental functions were considered negative. For example, soil respiration is positively correlated with nutrient (e.g. N) availability so is therefore beneficial for N-based function and would be considered positive. On the other hand, soil respiration is associated with a net loss of carbon from an ecosystem, which has negative implications for C-based function and would therefore be considered negative in the context of C multifunctionality. Another example of a detrimental function is N leaching, which has the potential to occur when the rate of net N mineralization (NNM) exceeds the rate of plant N-uptake. N leaching would be considered negative in the context of N multifunctionality, as it generally has detrimental impacts on other systems, though these impacts are system-dependent. Net nitrification could be considered beneficial or detrimental; while the function creates plant-available N, nitrate leaches more readily from the soil column than other species of N, and its utilization by plants requires more energy than NH_4^+ use requires. Therefore, net nitrification was not used to determine N multifunctionality.

Nitrogen (N) multifunctionality was estimated by subtracting the normalized leaching potential index from the sum of normalized net N mineralization (NNM), decomposition, primary productivity, and soil respiration. For the 2017 growing season, N multifunctionality consisted of the sum of normalized NNM, primary productivity, and decomposition, because soil respiration and leaching were not measured prior to 2018. Carbon (C) multifunctionality was estimated by subtracting normalized soil respiration from the sum of normalized productivity and decomposition. The C multifunctionality of the 2017 growing season did not account for soil respiration rates, because soil respiration was not measured.

As decomposition, soil respiration, and NNM are typically highly correlated, counting each function separately may overestimate contributions to N and C multifunctionality. Decomposition, soil respiration, and NNM are considered separately in our estimate of N multifunctionality, as the three are distinct functions, but with the caveat that they may be correlated and therefore over-emphasized. Likewise, decomposition and soil respiration were considered separately in our estimate of C multifunctionality for 2018, as they are respectively beneficial and detrimental in terms of C.

Additionally, although I measured several ecosystem properties and one process (based on our distinction between process and function), our multifunctionality analysis focuses solely on ecosystem functions. Properties and processes are supplemental information, which, in addition to multifunctionality, can be vital in land-use decisions. For example, soil and plant tissue quality provide context for rates of net N mineralization, soil respiration, and decomposition. Biodiversity and plant species composition can also provide context for nutrient cycling functions, productivity, and provision of habitat and resources for fauna.

3.9 Statistical Analyses and Data Visualization

Analyses of variance (ANOVA) and repeated measures ANOVA (RMANOVA), where applicable, were performed to compare the following variables that express ecosystem properties across land-use types: soil moisture and temperature, soil ammonium and nitrate quantities, C:N ratios and mass of C and N in each ten cm soil depth increment and in plant foliar tissue, mass of roots in each ten cm soil depth increment, and community diversity indices. ANOVA was then performed to compare the ecosystem process of seasonal litter inputs across land-use types. ANOVA and RMANOVA, where applicable, were performed to compare the ecosystem functions of net N mineralization and nitrification, primary productivity, soil respiration, decomposition, and N leaching.

For variables that demonstrated significant differences among groups and did not violate ANOVA assumptions, a Tukey test was performed for all pairwise comparisons. For variables that violated ANOVA assumptions, a Kruskal-Wallace test was performed, followed by a post-hoc Nemenyi test with a Tukey-Kramer distribution using the R package PMCMRplus (Pohlert, 2018). The R packages ggplot (Widkham, 2017) and wesanderson color palette (Ram and Widkham, 2018) were used for further data analysis and visualization.

4. Results

4.1 Ecosystem Properties

4.1.1 Soil Moisture & Temperature

Beginning mid-July (day 200) 2018, following the establishment of the agricultural field, soil moisture was significantly ($p < 0.05$) greater in the native prairie than in the agricultural field (32.20 ± 1.53% and 29.62 ± 1.00%, respectively). Soil moisture in the early successional field was intermediate (31.33 ± 1.91%) and did not differ significantly from the agricultural field or native prairie. Mean soil moisture in the native prairie was significantly ($p < 0.01$) greater than that of the early successional field throughout the growing season (from day 157 in early June to day 228 in August) and was 34.30 ± 1.52% and 31.76 ± 1.90%, respectively (Figure 4A).

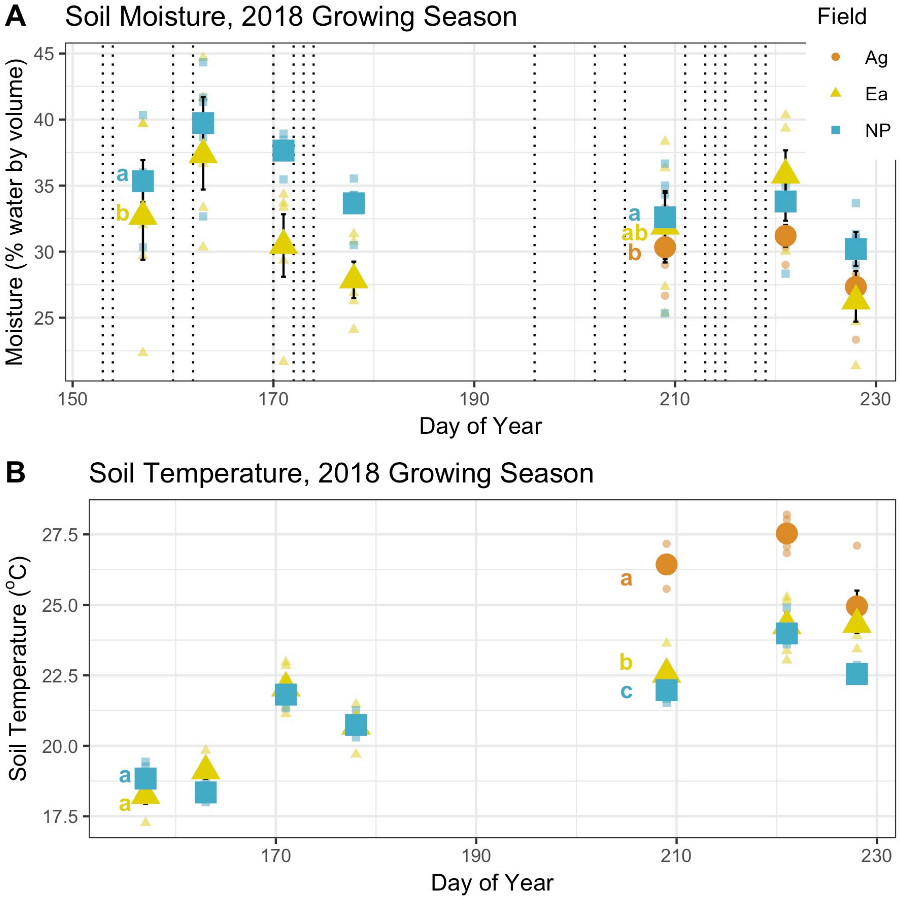

Figure 4. A) Soil moisture (% by volume) and **B)** temperature (°C) during the 2018 growing season. Plots in the agricultural field were not established until day 200, so it is excluded from measurements taken earlier in the growing season prior to day 200. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means across the five plots. Standard error bars are included for plot-level means. Letters to the left of day 200 denote statistically significant differences among all three fields in RMANOVA; letters at the start of the time series denote statistically significant differences among the early successional field and native prairie for the duration of the growing season.

Beginning mid-July (day 200) 2018, following the establishment of the agricultural field, the three fields differed significantly with regard to soil temperature. The mean temperature of the agricultural field was significantly ($p < 0.001$) greater (26.30 ± 0.10 °C) than that of the early successional field (23.70 ± 0.35 °C), which was significantly ($p < 0.05$) greater than that of the native prairie (22.84 ± 0.15 ^{o}C) from mid-July through August. The mean growing season soil temperature of the early successional field and native prairie, including all measurements recorded from June through August, did not differ significantly and was 21.60 ± 0.31 ^{o}C and 21.20 ± 0.15 ^{o}C, respectively (Figure 4B).

4.1.2 Soil Organic Matter

According to a repeated measures analysis of variance (RMANOVA) with depth as the repeated measure, soil C content in the top 10 cm did not differ significantly among any of the fields. Soil C was lowest in the agricultural field in the top 10 cm (1569.65 ± 62.36 g C m^{-2}), though it was not significantly lower than the native prairie and early successional field.

Soil C was significantly ($p < 0.05$) greater in the early successional field than in the native prairie in the 10 – 20 cm depth range (1310.65 ± 43.86 and 921.18 ± 113.52 g C m^{-2} respectively; Figure 5A). Soil C was intermediate in the agricultural field in the 10 – 20 cm depth range (1053.92 ± 51.39 g C m^{-2}) and did not differ significantly from the native prairie and early successional field. The three land-use types did not differ significantly in the 20 – 30 cm soil depth (Figure 5A).

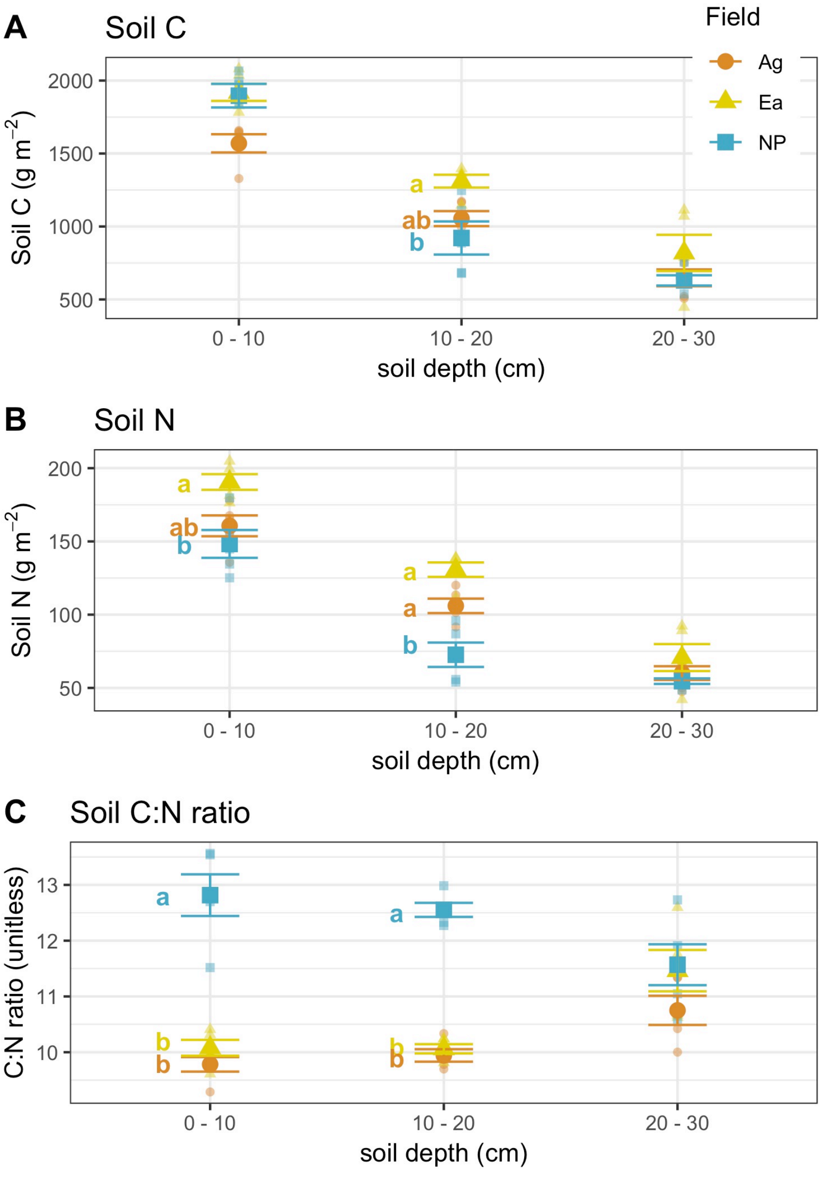

Figure 5. A) Soil C mass, **B)** Soil N mass, and **C)** Soil C:N ratios in each 10 cm soil depth increment, across land-use types. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each soil depth increment category represent statistically significant differences among fields found in RMANOVA with depth as the repeated measure (differences with regard to depth are not shown); non-significant

Soil N was significantly ($p < 0.05$) greater in the early successional field than in the native prairie in the top 10 cm. Soil N was intermediate in the agricultural field in the top 10 cm. Soil N was significantly ($p < 0.05$) greater in the early successional field and agricultural field than in the native prairie in the 10 – 20 cm depth. The three land-use types did not differ significantly in the 20 – 30 cm soil depth (Figure 5B).

The native prairie had significantly ($p < 0.05$) greater C:N ratios than the other two land-use types in the top 10 cm (12.82 ± 0.37). The agricultural field had the lowest C:N ratio in the top 10 cm (9.78 ± 0.13) but did not differ significantly from the early successional field. In the 10 – 20 cm depth, the native prairie had the greatest C:N ratio ($p < 0.05$; 12.55 ± 0.13). The agricultural field had the lowest C:N ratio (9.94 ± 0.11) but did not differ significantly from the early successional field. The three land-use types did not differ significantly from one another in the 20 – 30 cm depth (Figure 5C).

4.1.3 Root Biomass

The top 20 cm of soil contained about 95% of root mass in the native prairie and early successional field. The top 10 cm of soil contained 100% of roots in the agricultural field. In the top 10 cm of soil, the native prairie had significantly ($p < 0.05$) greater root mass than the agricultural field (927.59 ± 320.29 g m^{-2} and 16.45 ± 10.40 g m^{-2}, respectively). The early successional field had an intermediate root biomass (355.25 ± 129.98 g m^{-2}) that did not differ significantly from the other two fields. In the 10 – 20 cm soil depth, the native prairie had significantly ($p < 0.05$) greater root mass (180.91 ± 76.44 g m^{-2}) than the agricultural field, which was absent roots. The early successional field had an intermediate root biomass (23.03 ± 19.18 g m^{-2}) that did not differ significantly from the other two fields. Root mass in the 20 – 30 cm soil depth did not differ significantly among fields.

4.1.4 Plant Tissue Quality

Overall, foliar biomass in the early successional field consistently had lower C:N ratios than the native prairie, and the agricultural field had the lowest C:N ratios (Figure 6). The early successional field had significantly ($p < 0.01$) lower forb C:N ratios (35.20 ± 1.22) than the native prairie (51.03 ± 3.15). The agricultural field had significantly ($p < 0.05$) lower graminoid C:N ratios (22.52 ± 1.99) than the early successional field (30.67 ± 2.77) and native prairie (51.95 ± 8.96). The shrub foliar C:N differed the least ($p > 0.05$) between the two land-use types (26.73 ± 0.71 in the early successional field, 29.77 ± 7.03 in the native prairie).

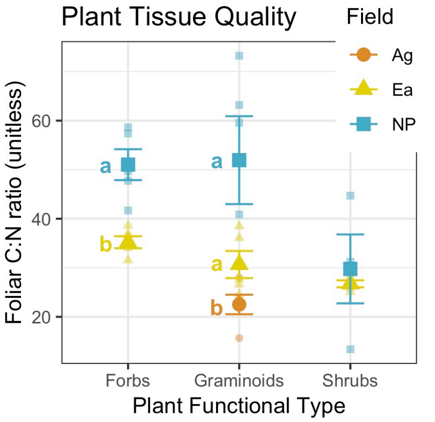

Figure 6. Foliar plant tissue quality, given as a unitless ratio of foliar C to N. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each plant functional type category represent statistically significant differences among fields found in an ANOVA; unmarked categories denote non-significant differences.

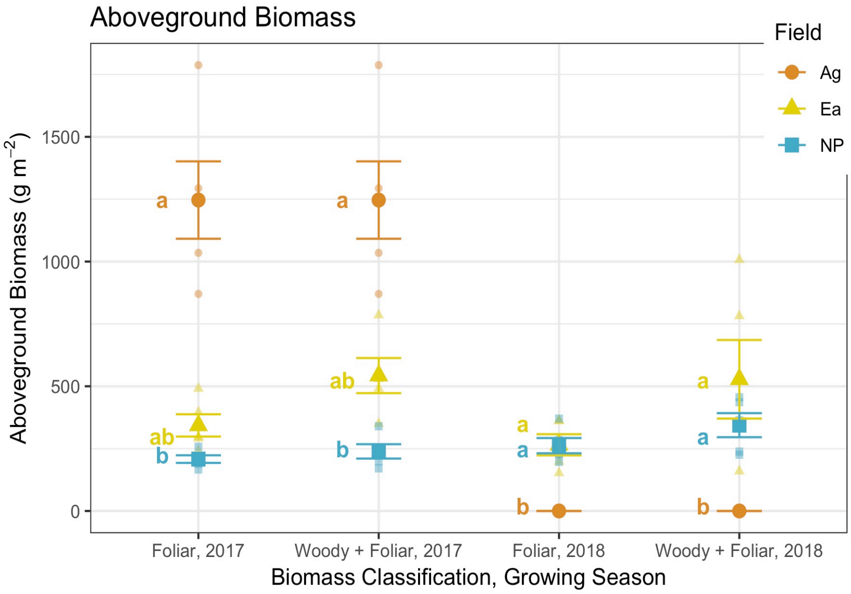

Figure 7. Mass of aboveground biomass collected at the end of the 2017 and 2018 growing seasons. Both foliar and total live (labeled as "woody + foliar") are presented for each growing season. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means across the five plots. Standard error bars are included for plot-level means. The agricultural field was not planted during summer 2018, so no biomass was available for collection at the end of the 2018 growing season. Letters to the left of each biomass classification and growing season category represent statistically significant differences among fields found in an ANOVA.

4.1.5 Aboveground Live Biomass

Total aboveground live biomass, which included both the foliar and woody components of vegetation differed significantly ($p < 0.01$) between the agricultural field (1250 ± 155 g m^{-2}) and native prairie (239 ± 29 g m^{-2}) in 2017, with intermediate total live biomass values in the early successional field (543 ± 70 g m^{-2}). During the 2018 growing season, the early successional field had slightly lower total aboveground biomass (528 ± 158 g m^{-2}) than in 2017, and the native prairie had slightly higher mean biomass (344 ± 48 g m^{-2}), but they did not differ significantly from one another in either year. The agricultural field was not planted during the 2018 growing season.

Foliar biomass differed significantly ($p < 0.01$) between the agricultural field (1250 ± 155 g m^{-2}) and native prairie (208 ± 15 g m^{-2}) in 2017, with intermediate foliar biomass values in the early successional field (343 ± 45). During the 2018 growing season, the early successional field had slightly lower foliar biomass (265 ± 42 g m^{-2}) than in 2017, and the native prairie had slightly higher foliar biomass (262 ± 31 g m^{-2}). The foliar biomass of the fields also did not differ significantly in 2018 (Figure 7).

4.1.6 Species Composition & Diversity

Though species richness, Simpson's and Shannon's species diversity indices, and Shannon's evenness index all do not differ significantly between the two non-monoculture fields (the early successional field and the native prairie; Table 2), their species composition differed substantially in several ways. First, dominant species within graminoid, forb, and shrub plant functional type differed between the two fields. *Andropogon gerardii* (big bluestem), a native warm-season grass, was the dominant graminoid in the native prairie, while *Festuca arundinaceae* (tall fescue) was dominant in the early successional field. *Galium verum* (yellow bedstraw), a non-native forb, was the dominant forb in both land-use types, *Caprifoliaceae symphoricarpos* (coralberry), a native shrub, dominated the early successional field, while *Rubus spp.* (both raspberry and blackberry species) dominated plots in the native prairie. Because of the lack of significant differences in species diversity indices, in conjunction with the relatively different species composition, further statistical analysis was performed using the additional categories

Table 2. Means of diversity indices: species richness, Simpson's diversity index, Shannon's diversity index, Shannon's evenness, and the proportion of evenness compared to total equality for comparison. No means differed significantly among fields.

Diversity Index	Early Successional Field	Native Prairie
Species Richness	19.60 ± 1.54	21.20 ± 1.16
Simpson's Diversity	0.71 ± 0.03	0.68 ± 0.03
Shannon's Diversity	1.81 ± 0.28	1.49 ± 0.11
Shannon's Evenness	0.53 ± 0.04	0.50 ± 0.03
Proportion of Shannon's Evenness to Totally Equality	0.19 ± 0.01	0.16 ± 0.02

of origin (native vs. non-native), plant type, and photosynthetic pathway.

The relative frequency of native and non-native species differed significantly ($p < 0.05$) between the fields; the early successional field had significantly less fractional cover of native plants (0.27 ± 0.06) than the native prairie (0.57 ± 0.07). The relative frequencies of observed forb and graminoid species also differed significantly ($p < 0.05$) between the fields, where the early successional field had significantly greater cover of forb species than the native prairie (0.48 ± 0.10 and 0.20 ± 0.05, respectively), and significantly less cover of graminoid species than the native prairie (0.25 ± 0.11 and 0.58 ± 0.07, respectively). Additionally, the relative frequency of observed species that utilize C_3- and C_4-photosynthetic pathways differed significantly between the fields; the early successional field had significantly greater cover of C_3 plants ($p < 0.001$) than the native prairie (0.93 ± 0.04 and 0.44 ± 0.06, respectively).

4.1.7 Litter Mass

Because the litter mass data violated homogeneity of variance, a more conservative Kruskall-Wallace test was performed. The agricultural field, with 0.00 ± 0.00 g m^{-2} of litter had a significantly smaller litter pool ($p < 0.01$) than the native prairie, but did not differ significantly from the early successional field. The early successional field had an intermediate litter pool (109.44 ± 22.06 g m^{-2}), and the native prairie had the largest litter pool (257.55 ± 29.04 g m^{-2}).

4.1.8 Soil NH_4^+ and NO_3^-

In the native prairie, soil nitrate quantities remained at or very close to 0.0 g NO_3^--N m^{-2} throughout summer 2017. According to a RMANOVA, soil nitrate in the early successional field did not differ significantly from that of the native prairie at the beginning of the first incubation in mid-June (day 170) of 2017. Because the agricultural field was not planted until after day 190 in 2017, ammonium and nitrate quantities were not sampled. The agricultural field had significantly ($p < 0.0001$) greater soil nitrate (0.13 ± 0.01 g NO_3^--N m^{-2}) than both the early successional field and native prairie at the beginning of the mid-summer incubation on day 198, which was expected due to fertilization and the prior absence of vegetation; the other unfertilized land-use types had soil nitrate quantities near zero (Figure 8A). In mid-July (day 207) in summer 2018 and in early October (day 296) in fall 2018, nitrate levels did not differ significantly among any of the fields, as the agricultural field was not fertilized during this growing season (Figure 8B).

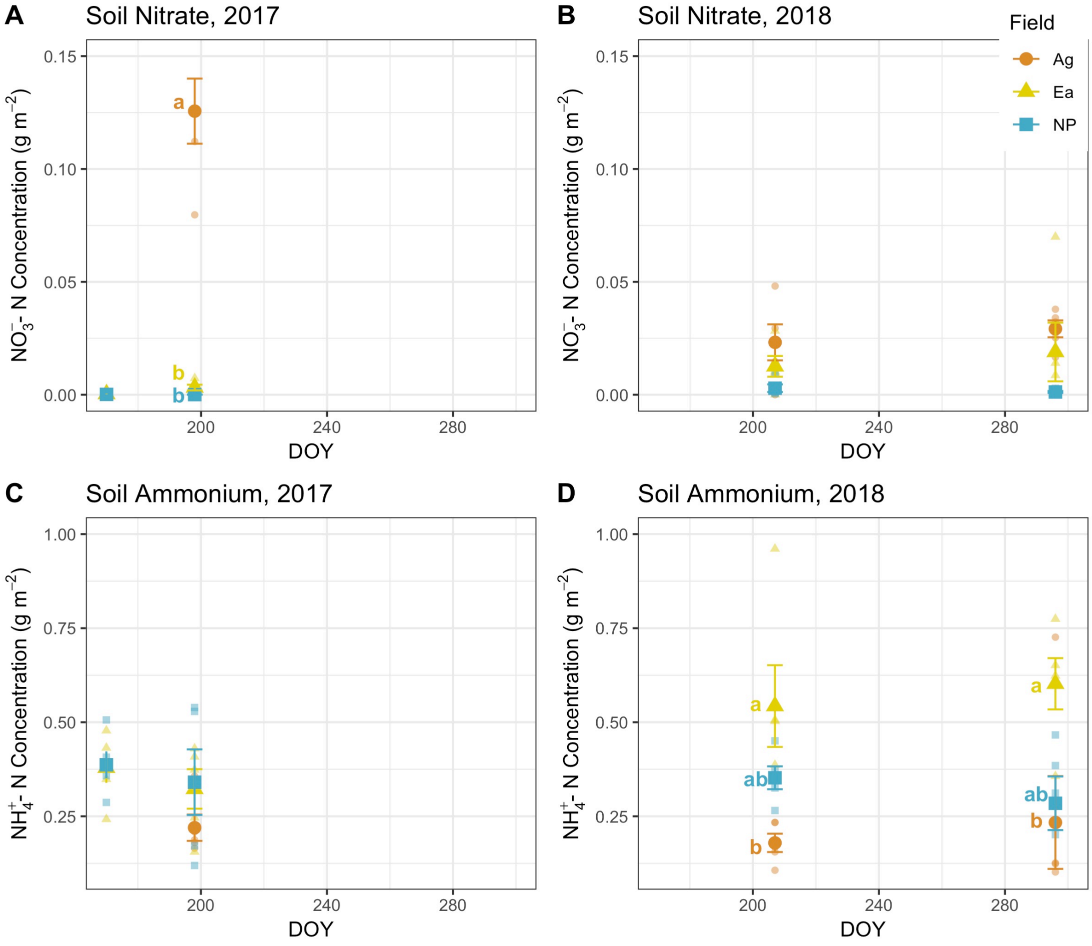

Figure 8. Time series of absolute levels (g m^{-2}) of **A)** soil NO_3^--N in 2017 and **B)** 2018 and **C)** soil NH_4^+-N in 2017 and **D)** 2018. Days 170 and 198 in 2017 and day 207 in 2018 represent quantities found the initial soil sample taken in each incubation. Day 296 in 2018 represents the quantities found in the top 10 cm collected to approximate leaching. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each DOY represent statistically significant differences among fields found in a RMANOVA (differences with regard to time are not shown).

According to a RMANOVA with time as a repeated measure, field and time did not account for significant differences in soil ammonium in 2017. The agricultural field had lower quantities on day 198 (0.22 ± 0.03 g NH_4^+-N m^{-2}), though it did not differ significantly from the other native prairie and early successional field (Figure 8C). On day 207 in summer 2018 and on day 296 in fall 2018, NH_4^+-N was significantly ($p < 0.05$) greater in the early successional field (0.54 ± 0.11 g NH_4^+-N m^{-2} on day 207; 0.60 ± 0.07 g NH_4^+-N m^{-2} on day 198) than in the agricultural field (0.18 ± 0.02 g NH_4^+-N m^{-2} on day 207; 0.23 ± 0.12 g NH_4^+-N m^{-2} on day 198). The native prairie had intermediate quantities of soil NH_4^+-N on both days (0.35 ± 0.03 g NH_4^+-N m^{-2} on day 207; 0.28 ± 0.07 g NH_4^+-N m^{-2} on day 196; Figure 8D).

4.2 Ecosystem Processes

4.2.1 Seasonal Litter Inputs

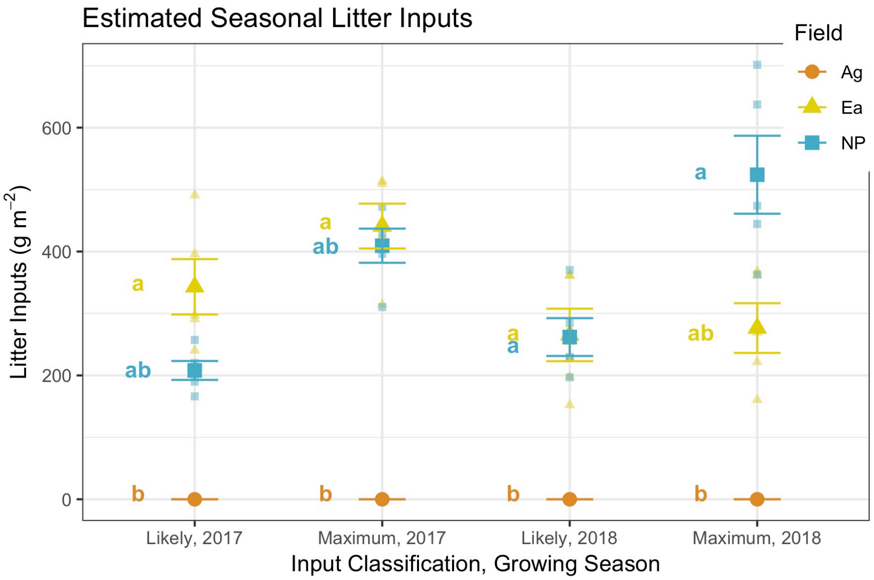

Figure 9. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means across the five plots. Standard error bars are included for plot-level means. Letters to the left of each classification and growing season represent statistically significant differences among fields found in an ANOVA.

Because estimated seasonal litter inputs violated homogeneity of variance, a more conservative Kruskall-Wallace test was performed. Likely seasonal litter inputs from the 2017 growing season, based on the foliar component of vegetation, differed significantly ($p < 0.01$) between the agricultural field (0.00 ± 0.00 g m^{-2}) and early successional field (343.07 ± 44.71 g m^{-2}), with an intermediate likely seasonal input in the native prairie (208.00 ± 15.27 g m^{-2}). Likewise, the maximum possible seasonal input, based on the live foliar component of vegetation in addition to the standing dead biomass, differed significantly ($p < 0.01$) between the agricultural field and early successional field, with an estimated seasonal input of 0.00 ± 0.00 g m^{-2} in the agricultural field, 441.09 ± 36.19 g m^{-2} in the early successional field, and 409.41 ± 27.60 g m^{-2} in the native prairie (Figure 9). Though the difference between the maximum litter inputs in 2017 in the native prairie and agricultural field was not statistically significant, the p-value obtained by the more conservative non-parametric Kruskall-Wallace test was noteworthy, at 0.051.

Likely seasonal litter input from the 2018 growing season was significantly ($p < 0.05$) lower in the agricultural field than the early successional field and native prairie (265.33 ± 42.38 and 261.92 ± 30.54 g m^{-2}, respectively). Maximum seasonal litter input of 2018 was significantly ($p < 0.01$) greater in the native prairie (523.99 ± 62.91 g m^{-2}) than in agricultural field (0.00 ± 0.00 g m^{-2}), with intermediate maximum seasonal input in the early successional field (276.43 ± 40.17 g m^{-2}; Figure 9).

4.3 Ecosystem Functions

4.3.1 Net N Mineralization and Nitrification

Net N mineralization in both 2017 incubations did not differ significantly among land-use types but was lower during the second incubation than the first. During the first summer 2017 incubation, beginning day 170, net N mineralization occurred in both fields (0.32 ± 0.09 g N m^{-2} $month^{-1}$ in the early successional field and 0.38 ± 0.11 g N m^{-2} $month^{-1}$ in the native prairie), while during the second incubation there was a mix of net N mineralization and net immobilization; overall, net N mineralization occurred in the agricultural field (0.37 ± 0.25 g N m^{-2}

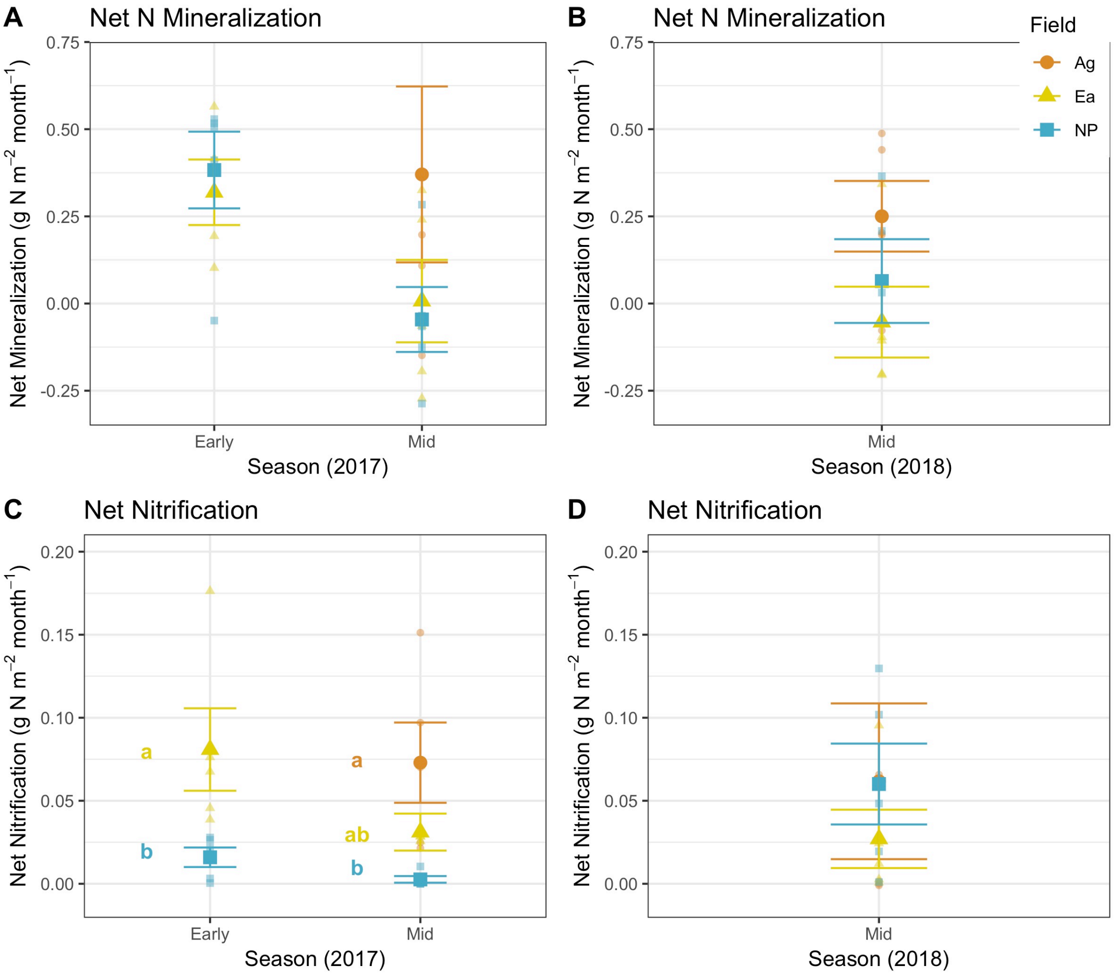

Figure 10. A) Net N mineralization during two incubations, in early summer ("Early") and mid-summer ("Mid") in the 2017 growing season and **B)** one incubation in mid-summer ("Mid") of the 2018 growing season and **C)** Net nitrification during two incubations, in early summer ("Early") and mid-summer ("Mid") in the 2017 growing season and **D)** one incubation in mid-summer ("Mid") of the 2018 growing season. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each time of season represent statistically significant differences among fields found in an ANOVA.

month^{-1}), a mix of net N mineralization and immobilization occurred across plots in the early successional field (0.01 ± 0.12 g N m^{-2} month^{-1} on average across early successional plots), and net N immobilization occurred in the native prairie (-0.05 ± 0.09 g N m^{-2} month^{-1} on average across native prairie plots; Figure 10A). Net N mineralization during the mid-summer incubation in summer 2018, beginning day 207, did not differ significantly among land-use types, although values exhibited similar patterns to those of mid-summer 2017, with greatest values in the agricultural field and least (net N immobilization on average) in the native prairie (Figure 10B).

Net nitrification during the first incubation of 2017 differed significantly ($p < 0.01$) between the early successional field (0.08 ± 0.02 g N m^{-2} month^{-1}) and native prairie (0.02 ± 0.01 g N m^{-2} month^{-1}). During the second incubation during summer 2017, the agricultural field had significantly ($p < 0.05$) greater net nitrification (0.07 ± 0.02 g N m^{-2} month^{-1}) than the native prairie (0.00 ± 0.00 g N m^{-2} month^{-1}), with intermediate rates of net nitrification occurring in the early successional field (0.03 ± 0.01 g N m^{-2} month^{-1}; Figure 10C). Net nitrification did not differ significantly among land-use types during the mid-summer incubation of 2018 (Figure 10D).

4.3.2 Soil Respiration

Beginning mid-July (day 200) 2018, following the establishment of the agricultural field, soil respiration was significantly ($p < 0.01$) greater in the early successional field (0.24 ± 0.03 g CO_2 m^{-2} h^{-1}) than in the agricultural field (0.18 ± 0.02 g CO_2 m^{-2} h^{-1}) and significantly greater in the agricultural field ($p < 0.0001$) than in the native prairie (0.15 ± 0.01 g CO_2 m^{-2} h^{-1}). Mean soil respiration

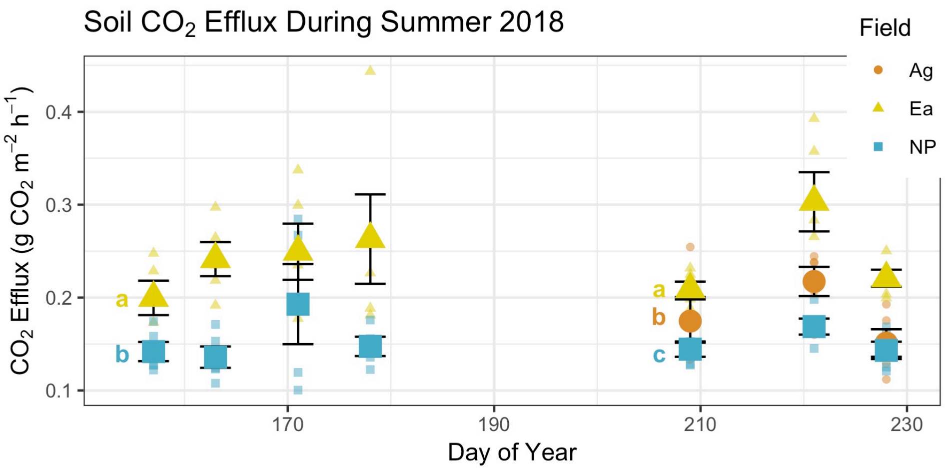

Figure 11. Soil CO_2 efflux (g CO_2 m^{-2} h^{-1}) during the 2018 growing season. Plots in the agricultural field were not established until day 200, so it is excluded from measurements taken earlier in the growing season prior to day 200. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of day 200 denote statistically significant differences among all three fields in RMANOVA; letters at the start of the time series denote statistically significant differences among the early successional field and native prairie for the duration of the growing season. Differences with regard to time are not shown.

of the early successional field (0.24 ± 0.03 g CO_2 m^{-2} h^{-1}) was significantly ($p < 0.0001$) greater than that of the native prairie (0.15 ± 0.02 g CO_2 m^{-2} h^{-1}) throughout the growing season (from day 157 to day 228), respectively (Figure 11).

4.3.3 Primary Productivity

Primary productivity estimated from foliar deciduous biomass at peak of growing season differed significantly ($p < 0.01$) between the agricultural field (1246.56 ± 155.13 g m^{-2}) and native prairie (208.00 ± 15.27 g m^{-2}) in the 2017 growing season. During the 2018 growing season, the early successional field had slightly lower primary productivity than in 2017 (265.33 ± 42.38 g m^{-2}), and the native prairie had slightly higher primary productivity than in 2017 (261.92 ± 30.54 g m^{-2}). The two land-use types did not differ significantly from each other in terms of their productivities. Woody biomass was greatest in the early successional field (199.76 ± 84.49 g m^{-2} during the 2017 growing season and 262.61 ± 117.92 g m^{-2} during the 2018 growing season), though not significantly greater than the native prairie (30.91 ± 14.79 g m^{-2} during the 2017 growing season and 82.08 ± 31.17 g m^{-2} during the 2018 growing season), so whereas productivity may be more greatly underestimated in the early successional field due to woody biomass increment, it may not be substantially more than the native prairie.

4.3.4 Litter Decomposition

The mean estimated litter decomposition rate was significantly ($p < 0.01$) greater in the early successional field than in the native prairie for the 2017 growing season. The slightly higher decomposition rates for each growing season are based on the maximum possible seasonal input, which includes standing dead material. During the 2018 growing season, the mean estimated litter decomposition rate did not differ significantly between the early successional

Table 3. Mean and standard error of estimated decomposition rates based on seasonal inputs for the 2017 and 2018 growing seasons. There are two estimates given for each year: one based on the foliar biomass present at the end of the growing season (labeled "Likely"), the other based on the standing dead biomass in addition to the foliar biomass present at the end of the growing season (labeled "Maximum").

		Decomposition rate estimate by field ($year^{-1}$)	
	Seasonal litter inputs used in estimate	Early Successional	Native Prairie
2017	Likely	3.73 ± 1.08 ^{a}	0.86 ± 0.13 ^{b}
	Maximum	4.76 ± 1.06 ^{a}	1.73 ± 0.34 ^{b}
2018	Likely	2.70 ± 0.48 ^{a}	1.05 ± 0.12 ^{a}
	Maximum	2.81 ± 0.48 ^{a}	2.13 ± 0.31 ^{a}

field and native prairie (Table 3). The agricultural field was excluded from this analysis, because of a lack of seasonal litter inputs and litter pool. The lack of litter decomposition in the agricultural field was included in the multifunctionality analyses, because the lack of seasonal litter inputs and litter pool are key characteristics associated with agriculture.

4.3.5 Nutrient Leaching

Total NH_4^+-N and NO_3^--N quantities in the top 10 cm of soil were significantly greater in the early successional field (0.69 ± 0.20 g N m^{-2} on day 207; 0.62 0.06 g N m^{-2} on day 296) than in the agricultural field (0.26 ± 0.07 g N m^{-2} on day 207; 0.26 0.12 on day 296) and native prairie (0.26 ± 0.07 g N m^{-2} on day 207; 0.29 ± 0.07 g m^{-2}; Figure 12A). Total NH_4^+-N and NO_3^--N quantities did not differ significantly in the 10 – 20 cm or 20 – 30 cm soil depths (Figures 12B and 12C).

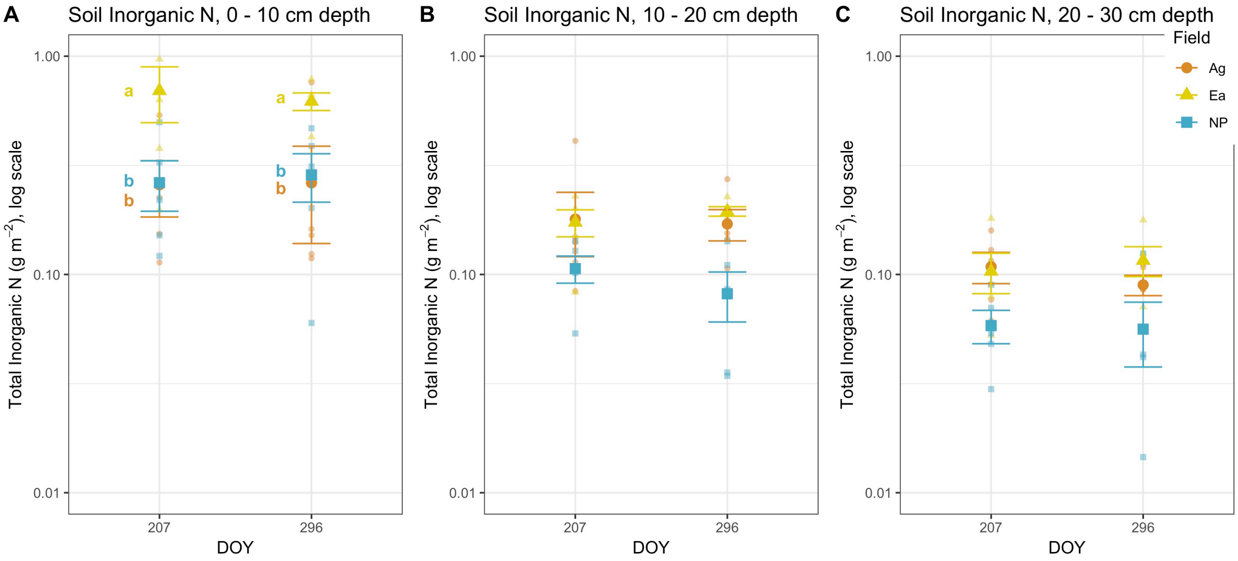

Figure 12. Soil inorganic N, in the form of ammonium and nitrate in **A)** 0 – 10 cm depth, **B)** 10 – 20 cm depth, and **C)** 20 – 30 cm depth, for mid-July (day 207) and mid-October (day 296) during the 2018 growing season. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each day denote statistically significant differences among all three fields in RMANOVA (differences with regard to depth are not shown).

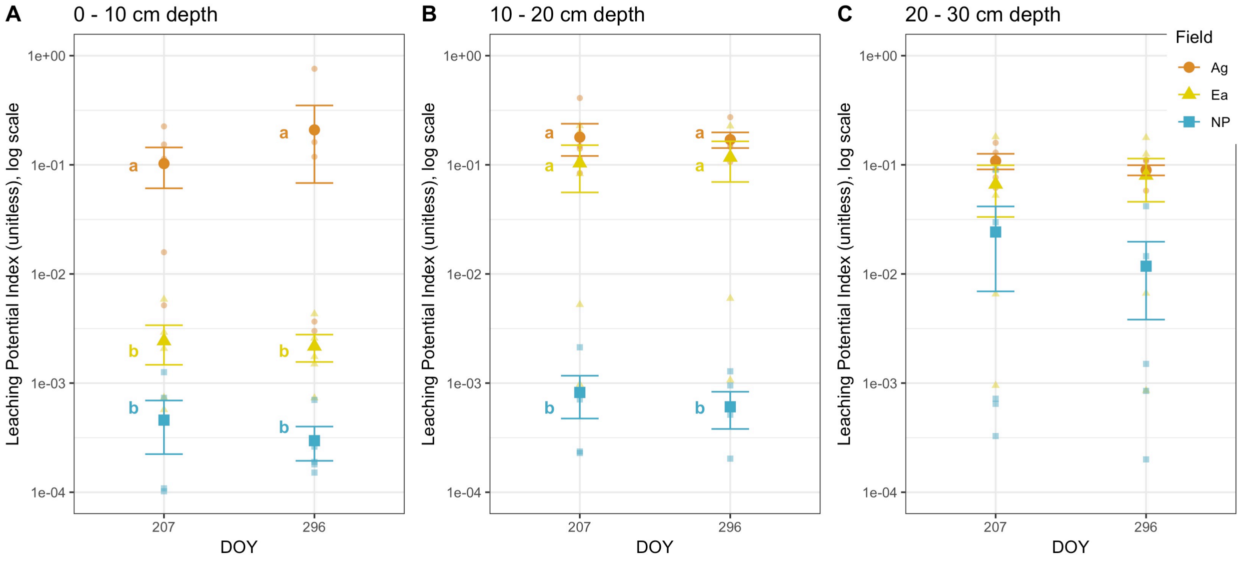

Figure 13. Leaching potential index for **A)** 0 – 10 cm depth, **B)** 10 – 20 cm depth, and **C)** 20 – 30 cm depth, for mid-July (day 207) and mid-October (day 296) during the 2018 growing season. Leaching potential index was calculated as the total inorganic N, in the form of NH_4^+ and NO_3^-, divided by the total mass of roots within and below the soil depth increment for each 10-cm incremental depth. Smaller points represent measurements of variables at the plot level. Larger points represent the field-level means from each of the five plots. Standard error bars are included for plot-level means. Letters to the left of each day denote statistically significant differences among the fields in a RMANOVA.

According to a RMANOVA with depth as the repeated measure, the agricultural field overall had significantly ($p < 0.01$) greater leaching potential index values than the early successional field and native prairie, which did not differ significantly ($p > 0.05$) from each other. In the 0 – 10 cm depth, the agricultural field had a significantly ($p < 0.05$) greater leaching potential index (0.10 ± 0.04 in mid-July and 0.21 ± 0.14 in mid-October) than both the early successional field and native prairie, which were near zero for both sampling dates (Figure 13A). In the 10 – 20 cm depth, the native prairie had a significantly lower leaching potential index (near zero) than both the early successional field (0.10 ± 0.05 in mid-July and 0.12 ± 0.05 in mid-October) and agricultural field (0.18 ± 0.06 in mid-July and 0.17 ± 0.03 in mid-October; Figure 13B). In the 20 – 30 cm depth, there were no significant differences in leaching potential among the fields, but the agricultural field had the highest leaching potential index (0.11 ± 0.02 mid-July and 0.09 ± 0.01 mid-October), closely followed by the early successional field (0.07 ± 0.03 in mid-July and 0.08 ± 0.03 in mid-October). There were similar patterns between the mid-July (day 207) and mid-October (day 296) leaching potential index values, with no significant difference between the two days (Figure 13C).

4.4 Multiple Functions and Multifunctionality Analysis

4.4.1 Multiple Functions

Net N mineralization rates did not differ significantly among land-use types during both 2017 and 2018, though on average, the agricultural field had higher rates of net N mineralization (and positive z-scores) than the early successional field and native prairie (with low or negative z-scores). Normalized soil respiration rates during August 2018 were significantly ($p < 0.0001$) greater in the early successional field than in the native prairie and agricultural field. During the 2017 growing season, productivity was significantly greater ($p < 0.001$) in the agricultural field than in the early successional field and native prairie.

Table 4. Mean and standard error of the ecosystem functions examined in this study (net N mineralization, soil respiration, productivity, decomposition, and leaching) for each field in each growing season. Significant differences are marked with different letters and are in bold.

Year	Function	Ag	Ea	NP
2017	Net N Mineralization	0.96 ± 1.20 ^{a}	-0.21 ± 0.41 ^{a}	0.00 ± 0.31 ^{a}
	Soil Respiration	---	---	---
	Productivity	**4.29 ± 0.77** ^{a}	**-0.22 ± 0.22** ^{b}	**-0.90 ± 0.0.08** ^{b}
	Decomposition	**-0.88 ± 0.00** ^{a}	**1.48 ± 0.68** ^{b}	**-0.33 ± 0.08** ^{a}
	Leaching	---	---	---
2018	Net Mineralization	0.39 ± 0.48 ^{a}	-1.05 ± 0.48 ^{a}	-0.49 ± 0.57 ^{a}
	Soil Respiration	**-0.24 ± 0.33** ^{a}	**1.09 ± 0.28** ^{b}	**-0.85 ± 0.16** ^{a}
	Productivity	**-1.94 ± 0.00** ^{a}	**-0.61 ± 0.21** ^{b}	**-0.63 ± 0.15** ^{b}
	Decomposition	**-0.88 ± 0.00** ^{a}	**0.83 ± 0.30** ^{b}	**-0.21 ± 0.07** ^{a}
	Leaching	**1.00 ± 0.35** ^{a}	**-0.11 ± 0.35** ^{b}	**-0.87 ± 0.06** ^{b}

During the 2018 growing season, when the agricultural field was not planted, its productivity was significantly lower than the early successional field and native prairie. Normalized decomposition rates from both 2017 and 2018 were significantly ($p < 0.05$ and $p < 0.01$, respectively) greater in the early successional field than in the agricultural field and native prairie. In 2018, the normalized leaching potential index was significantly greater ($p < 0.01$) in the agricultural field than in the early successional field and native prairie, each of which had z-scores lower than zero. Soil respiration and leaching were not measured in 2017, and no assumptions can be made about their values based on 2018 data, since the agricultural field was left bare for the summer of 2018. While the significantly different outcomes of the statistical tests of each normalized z-score can provide some idea of multiple benefits provided, they do not necessarily equate to multifunctionality, because multifunctionality recognizes the overall ability of a system to conduct multiple functions simultaneously.

4.4.2 Multifunctionality

Mean nitrogen (N) multifunctionality differed significantly among fields in 2017 ($p < 0.01$) and in 2018 ($p < 0.05$). In 2017, the agricultural field had significantly higher N multifunctionality (z = 4.37 ± 1.12) than the native prairie (z = -1.23 ± 0.35), with intermediate multifunctionality in the early successional field (z = and 1.06 ± 1.04). In 2018, the early successional field had significantly higher N multifunctionality (z = 0.37 ± 0.63) than the agricultural field (z = -3.67 ± 0.66), with the native prairie having intermediate N multifunctionality (z = -1.31 ± 0.69).

Mean carbon multifunctionality differed significantly among fields in 2017 ($p < 0.001$) and in 2018 ($p < 0.01$). In 2017, the agricultural field had significantly higher carbon multifunctionality (z = 3.41 ± 0.77) than the native prairie (z = -1.23 ± 0.13), with intermediate C multifunctionality in the early successional field (z = 1.26 ± 0.86). Mean C multifunctionality during the 2018 growing season was significantly higher in the native prairie and early successional field (z = 0.01 ± 0.32 and -0.87 ± 0.58, respectively) than in the agricultural field (z = -2.57 ± 0.33).

Total multifunctionality differed significantly among fields in both years. During the 2017 growing season, the agricultural field had significantly ($p < 0.05$) greater total multifunctionality (z = 7.78 ± 1.51) than the native prairie and early successional field (z = -2.46 ± 0.42 and 3.32 ± 1.86, respectively). During the 2018 growing season, the early successional field and native prairie had significantly ($p < 0.01$) greater total multifunctionality (z = -0.50 ± 1.08 and -1.30 ± 0.76, respectively) than the agricultural field (z = -6.24 ± 0.65).

Table 5. Mean z-score of nitrogen (N), carbon (C), and total multifunctionality for each growing season. Significant differences are marked with different letters and are in bold.

Year	Multifunctionality	Ag	Ea	NP
2017	Nitrogen	**4.37 ± 1.12 ^{a}**	**1.06 ± 1.04 ^{ab}**	**-1.23 ± 0.35 ^{b}**
	Carbon	**3.41 ± 0.77 ^{a}**	**1.26 ± 0.86 ^{ab}**	**-1.23 ± 0.13 ^{b}**
	Total	**7.78 ± 1.51 ^{a}**	**2.32 ± 1.86 ^{b}**	**-2.46 ± 0.42 ^{b}**
2018	Nitrogen	**-3.67 ± 0.66 ^{a}**	**0.37 ± 0.63 ^{b}**	**-1.31 ± 0.69 ^{ab}**
	Carbon	**-2.57 ± 0.33 ^{a}**	**-0.87 ± 0.58 ^{b}**	**0.01 ± 0.32 ^{b}**
	Total	**-6.24 ± 0.65 ^{a}**	**-0.50 ± 1.08 ^{b}**	**-1.30 ± 0.76 ^{b}**

5. Discussion

5.1 Ecosystem Properties and Processes

The significantly greater soil moisture in the native prairie can be explained by the abundance of warm season C_4 grasses in the native prairie, which are more water-use efficient than C_3 plants found in the early successional field. Though Foster et al. (2015) found that canopy cover rather than plant uptake was more important in predicting soil moisture in tallgrass prairie, the canopy cover provided by the small shrubs in the early successional field likely did not differ enough from the canopy of the native prairie to impact soil moisture, because of the non-significant differences between the two plots in relative shrub frequency. The agricultural field unsurprisingly had significantly lower soil moisture than the native prairie, as it remained bare through the sampling period and therefore was most susceptible to evaporation or leaching. As was expected, the soil temperature in the agricultural field was greatest; the significantly lower soil temperatures in the early successional field and native prairie were due to plant cover.

The agricultural field, as expected due to N inputs via fertilizer, had the lowest soil C:N ratios, though they were not significantly less than those in the early successional field. The early successional field was also expected to have lower soil C:N ratios than the native prairie, because soils underlying herbaceous C_4 photosynthetic plants, such as the native grasses that dominate the prairie, tend to have higher C:N ratios than soils underlying C_3 photosynthetic plants. In addition, much of the aboveground N in biomass is sent belowground during senescence in the prairie, leading to a lower N input via surface litter (Blair, 1997; Knapp and Seastedt, 1986). Though crop removal also accounts for substantial amounts of N lost from the soil system, the fertilization of the agricultural field restores N, with N inputs likely exceeding N loss.

As the early successional field and native prairie are both about 20 years old, soil C:N ratios were expected to be nearing steady-state conditions in which N is becoming more limited (Baer and Blair, 2008) of 3000-4000 g m^{-2} of soil C (Smith and Johnson, 2003) and 300-400 g m^{-2} of soil N (Lett et al., 2004). Soil C:N ratios in the early successional field were comparable to steady state conditions, while the native prairie had slightly higher C:N ratios. This could be a result of its species composition or of the controlled burns that take place in the field every three years.

Expectations for plant tissue quality were similar to those for soil quality: the agricultural field, as expected, had the lowest foliar C:N ratios, and the native prairie had the highest.

Because the native prairie has significantly more C_4 photosynthetic graminoids than the early successional field, the C:N ratios of graminoids was expected to differ more than they did between the two fields (Reynolds et al., 2003).

The native prairie was expected to have lower seasonal litter input due to the native prairie's tendency to maintain standing dead material as well as regular controlled burns in the native prairie decreasing litter inputs to soil organic matter (SOM) pools (Reynolds et al., 2003). Seasonal litter inputs, in conjunction with decomposition rates, can predict the litter pool. While estimated likely seasonal litter inputs were greater in the early successional field, they did not differ significantly from the native prairie. The maximum possible litter inputs for the 2018 growing season were significantly greater in the native prairie than in the early successional field. The higher C:N ratios of the plant tissue in the native prairie also suggest lower decomposition rates. Therefore, the native prairie, with its litter inputs relatively comparable to the early successional field, along with its high C:N ratios, provides context for the large litter pool of the native prairie at the start of the 2018 growing season.

5.2 Ecosystem Functions

Organic matter inputs with low C:N ratios, such as those in the early successional field, promote N mineralization during decomposition (Aerts, 1995; Hobbie, 1992; Pastor et al., 1984; Vitousek, 1982). Subsequently, foliar N is positively correlated with NNM rates (Uri et al., 2008), so the early successional field was expected to have higher NNM rates than the native prairie. Likewise, C_4 tallgrass litter, such as that of the native prairie, promotes lower N mineralization rates and N availability relative to some other plant types (Craine et al., 2002; Dijkstra et al., 2006; Tilman and Wedin, 1991; Wedin and Pastor, 1993; Wedin and Tilman, 1990). The lower rate of net mineralization in the native prairie was expected due to the dominance of C_4 tallgrasses, the litter of which promotes lower N mineralization rates and N availability relative to other plant types.

Though differences in NNM rates between the two fields were not significant, the early successional field showed a higher average rate than the native prairie. The native prairie also demonstrated NNM rates more in line with net immobilization later in the growing season both in 2017 and 2018, aligning with expectations. Also following expectations, net nitrification was significantly greater in the early successional field than in the native prairie in early summer

2017. Net nitrification was significantly greater in the agricultural field than in the native prairie in mid-summer 2017. Though the agricultural field lacks aboveground litter inputs, its low C:N ratios due to fertilization determine the relatively high NNM rates.

The results of litter decomposition rates related to those of soil respiration rates. The early successional field had a significantly greater litter decomposition rate than the other two fields in 2017, as was expected due to environmental conditions of intermediate soil moisture and temperature as well as low C:N ratio of litter. Likewise, greater rates of soil respiration were observed in the early successional field, as expected. In 2018, the early successional field had significantly greater litter decomposition rates than the other two fields. The agricultural field, with its lack of litter inputs but higher NNM rates, had intermediate rates of soil respiration. The low rate of soil respiration in the native prairie was expected due to its low litter decomposition rate.

Plant biomass growth is highly correlated with N pools and N cycling rates (Zak et al., 1990). Because the agricultural field had the lowest soil C:N ratio and received N inputs, its significantly greater aboveground productivity followed expectations. Similarly, the native prairie, with its high soil C:N ratios, had the lowest aboveground productivity. Though the native prairie was expected to have greater aboveground biomass than the early successional field, the early successional field produced slightly (non-significantly) more aboveground biomass. In 2018, foliar production in both fields was essentially similar.

Overall the agricultural field had significantly greater leaching potential index values, while the native prairie had the lowest. The greater quantities of inorganic N in the top 10 cm of the early successional field compared to the other fields aligns with its greater NNM rates. While total concentrations of inorganic ammonium and nitrate were generally greatest in the early successional field and lowest in the native prairie across all depths, leaching potential index was highest in the agricultural field due to the lack of roots in the field. The native prairie, with its higher belowground root mass, had the lowest leaching potential index in the top 20 cm of soil. In the 20 – 30 cm depth, the leaching potential index did not differ among fields.

5.3 Implications of Multifunctionality

5.3.1 Carbon (C) Multifunctionality

The significantly greater 2017 C multifunctionality in the agricultural field than in the native prairie aligns with its very high productivity, about 4 standard deviations above the mean of all fields across both growing seasons. While the early successional field did not differ from the native prairie in terms of productivity, its C multifunctionality was intermediate due to its high rates of decomposition, 1.89 ± 0.82 standard deviations above the mean. Utilizing a C multifunctionality approach for the 2017 growing season provides a more holistic perspective of the multiple functions carried out by each land-use type. For example, a focus on the lower productivity in the early successional field in 2017 misses the fact that its decomposition, a function critical to other functions, is occurring at a greater rate than other land-use types. Estimated C multifunctionality during the 2017 growing season does not, however, account for the disservice of soil respiration, as it was not measured in 2017.

C multifunctionality for the 2018 growing season was significantly greater in the early successional field and native prairie than in the agricultural field, which aligns with the high rates of primary productivity in both fields compared to the unplanted agricultural field. While decomposition was generally considered a positive component of C multifunctionality, the inclusion of soil respiration as a negative side effect balanced out the benefits of decomposition, particularly in the early successional field, which had significantly higher soil respiration rates than the native prairie and agricultural field. C multifunctionality is able to capture the balance of soil respiration, a negative function in terms of C (as it is associated with C loss from the system), and decomposition, a positive function in terms of C.

5.3.2 Nitrogen (N) Multifunctionality

N multifunctionality for the 2017 growing season, which included the normalized rates of primary productivity and decomposition (similar to C multifunctionality) also included normalized rates of NNM. N multifunctionality demonstrated the same patterns as C multifunctionality during 2017. This is likely due to the fact that NNM did not differ significantly among land-use types in 2017, so its inclusion did not largely impact multifunctionality. The absence of the function of leaching (a disservice not estimated in 2017) in the N multifunctionality analysis could have impacted the agricultural field, making it less desirable.

N multifunctionality for the 2018 growing season was significantly greater in the early successional field than in the agricultural field, with intermediate multifunctionality in the native prairie. The high multifunctionality in the early successional field can be attributed to its high rates of soil respiration and decomposition. Higher N multifunctionality of the early successional field and native prairie was also driven by the relatively low primary productivity of the bare agricultural field. The inclusion of the detrimental function of leaching, which was greater in the agricultural field than in the other two fields during the 2018 growing season, further diminished its N multifunctionality. A focus on each function individually misses the details of the suite of N-based functions that multifunctionality provides. Though soil respiration, primary productivity, decomposition, and leaching were all lowest in the agricultural field and highest in the early successional field, the rates of soil respiration and decomposition in the native prairie did not differ significantly from the agricultural field. N multifunctionality captures the balance of the suite of traits provided by each land-use type.

5.3.4 Total Multifunctionality

The significantly greater total multifunctionality of the agricultural field in 2017 follows its particularly high productivity, which was included in both the C multifunctionality and N multifunctionality analysis. Though the early successional field demonstrated intermediate C multifunctionality and N multifunctionality, the magnitude of difference from the native prairie was not enough to drive a significant difference in total multifunctionality. Therefore, the agricultural field appears to provide the greatest level of multifunctionality when planted.

During the 2018 growing season, when the agricultural field was not planted, its lack of productivity and high potential for leaching place its significantly lower multifunctionality in context. The native prairie, with its intermediate N multifunctionality and high C multifunctionality, and the early successional field, with its high N multifunctionality and C multifunctionality, do not differ significantly from each other in total multifunctionality for the 2018 growing season. The native prairie has lower rates of soil respiration and decomposition than the early successional field. Understanding the underlying ecosystem properties and processes related to functions provides some context; the more recalcitrant litter in the native prairie has subsequently low rates of decomposition, soil respiration, net N mineralization, and productivity. Therefore, its more conservative N use and longer time scales over which the native prairie cycles nutrients may be causing it to be slightly undervalued under the multifunctionality framework.

5.4 Technical Limitations of the Study & Future Work

The scope of this study is restricted to the underlying properties, processes, and functions of supporting services. While supporting services are foundational for all other subsequent services, regulating, cultural, and provisioning services were all excluded from this study. Only three land-use types of interest were included in this study, based on the differentiable human management of each. Because this study took place at BEF, where there were not multiple examples of each land-use type, replication took place at the field and plot levels only, so I acknowledge pseudo-replication. The study does, however, benefit from three study sites that have been under their respective management regimes for the same period of time.

Due to the scope of the study and the short duration of activity in the agricultural field, there is also a lack of complete datasets to make generalizations on a broader, annual scale. The assumptions made in this research are based predominantly on a single growing season, sometimes even as short as a month-long period nearing peak biomass. In order to overcome this limitation, multi-year, frequent monitoring would be beneficial for more robust results, which could more thoroughly account for underlying properties and processes as well as their long-term interactions with functions. Additionally, in order to broaden the scope of this study, I suggest utilizing similar methods and defined terms to examine the properties, processes, and functions of supporting services in multiple land-use types that are similar in age but in a variety of locations for more general applicability.

Though utilizing z-scores normalizes disparate metrics, aggregating and ranking across these assumes equal weighting of functions. Using z-scores to analyze multifunctionality brings about two questions: 1) is the importance of each function equal? 2) are the variations across functions equivalent? Equating the importance of productivity with the importance of soil respiration, for example, is not necessarily appropriate. Similarly, assuming that one standard deviation in decomposition, for example, carries the same weight as one standard deviation difference in leaching is likely not appropriate. Additionally, NNM, soil respiration, and decomposition are related ecosystem functions that tend to be correlated, so combining them in multifunctionality analyses potentially overestimates their overall importance in the suite of ecosystem functions.

Additionally, multifunctionality analyses may be misleading, as they appear to value functions carried out on shorter time scales. The results of this study demonstrate that the rates of functions in the native prairie are slower than those of the early successional field and agricultural field, making it appear less valuable. The slow decomposition in the native prairie, paired with lower soil respiration, and lower productivity, is a defining ecological feature of C_4 prairies, and should not necessarily be less valued than other systems. Nevertheless, the native prairie does not appear particularly undervalued in these analyses. Incorporating elemental stoichiometric ratios into a multifunctionality analysis could help account for the issue of slow rates of functions.

5.5 Benefits of the study

The lack of a thorough exploration of supporting services and their underlying functions in the ES conversation provided the motivation for this study. Supporting services, though integral to the provision of other services, are often overlooked in the literature. I utilized a suite of five ecosystem functions that can be associated with three agreed upon supporting ES of soil formation, nutrient cycling, and primary productivity. These functions were compared across three land-use types, both individually and through a multifunctionality framework. This study is the first of its kind to:

1. Approach supporting ES by attempting to quantify the underlying properties, processes, and functions
2. Combine multiple functions into a single metric based on means from multiple years, capturing interannual variability in conditions
3. Apply a multifunctionality framework to C-based and N-based functions.

While questions as to the appropriateness and usefulness of multifunctionality remain, this work is a step toward elucidating the difficult task of quantifying supporting services, which remain important to consider in sustainable land-cover decisions.

6. References

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Chapter 2: Incorporating land-use management into a multifunctionality framework to inform supporting ecosystem services

1. Abstract

Supporting ecosystem services, the underlying foundation for other ecosystem services (ES), are not consistently defined, nor do they have agreed upon metrics. They are less often considered in environmental decision-making due to their subtlety. In order to inform three supporting services (soil formation, nutrient cycling, and primary productivity), a suite of aboveground, belowground, and aboveground-belowground interface ecosystem properties, processes, and functions were measured in three land-use types under variable management in northwestern Virginia over the course of two growing seasons (2017 and 2018): an unmanaged early successional field, a semi-managed native prairie, and a heavily managed agricultural field. The ecosystem functions measured (net nitrogen (N) mineralization, soil respiration, primary productivity, decomposition, and N leaching) were then applied to an ecosystem function (EF)-multifunctionality framework based on their association with supporting services. Unwanted impacts of ecosystem functions, or disservices, and the ecological cost of management were considered. While EF-multifunctionality was greatest in the agricultural field in 2017 when it was planted, its mean multifunctionality over both years balanced out to near zero (near the overall mean multifunctionality across all fields), suggesting that the agricultural field provides a suite of supporting ES when planted, but when accounting for interannual variability associated with crop rotations, it does not appear to consistently provide those supporting ES. The EF-multifunctionality of the native prairie over both years was several standard deviations below the overall mean, suggesting that it provides supporting ES at a lower rate than the other land-use types. The early successional field demonstrated the highest mean EF-multifunctionality across both growing seasons, suggesting that it consistently provides a suite of supporting ES. Including the ecological cost of human management further alters the provision of supporting services; because the agricultural field and native prairie failed to demonstrate greater overall multifunctionality than the early successional field in order to compensate for their ecological cost of management, they do not appear as valuable in terms of the net supporting services they provide. This study is the first of its kind to implement an EF-multifunctionality approach to inform supporting ES, while also accounting for disservices and cost of management. Assessing multifunctionality with z-scores raises the question of whether the importance and variation can be equated across all functions. The question of how to integrate different variables under a common currency is also a remaining gap in the ES conversation.

2. Introduction

2.1 Supporting Services (nature of the definition of supporting services)

Though the concept of ecosystem services (ES) was first noted in the late 1960s by King (1966) and Helliwell (1969) as functions of nature that benefit human society, the ES framework was not truly established until the 2005 Millennium Ecosystem Assessment (Millennium Ecosystem Assessment (MEA), 2005). This foundational text established a detailed framework that has been influential for sustainable development from an ecological perspective. The ES framework links human societies to ecological systems and serves as a useful decision-making tool (Barot et al., 2017). The ES concept is fundamental to The Economics of Ecosystems and Biodiversity initiative, the Intergovernmental Panel on Biodiversity and Ecosystem Services and the Intergovernmental Panel on Climate Change (IPCC) of Biodiversity (Marris, 2010).

ES is a socio-ecological bridging concept, joining multiple frameworks and discourses (Braat and de Groot, 2012). Van den Belt and Stevens (2016) describe ES as a "boundary object," a social construct that can be flexibly interpreted to accommodate context and subsequently shape cooperative, sustainable action. Rather than functioning as an objective definition of nature's value, the ES framework is a useful and flexible tool in environmental management and decision-making (Costanza et al., 1997; Dunn, 2010; Spangenberg and Settele, 2010). This flexibility, though, is a double-edged sword; the number of conflicting definitions makes ES a "fuzzy," or ambiguous, analytical concept (Robinson et al., 2013).

The framework established by the MEA is widely embraced in the ES conversation. The MEA separates ES into four categories: supporting, regulating, provisioning, and cultural services. Provisioning services include food, water, and forest products; regulating services modulate changes in climate, weather, disease dynamics, and water quality; cultural services comprise recreational, aesthetic, and spiritual benefits. The MEA distinguishes supporting services from the other ES, because they are a necessary foundation for the production of the other three types of ES. Though they provide benefits, they do so indirectly and over particularly long time scales, so they are less noticeable and, as a result, more difficult to quantify. The MEA describes supporting services as fluxes and stocks of matter and energy more so than benefits.

Examples of supporting services are not consistently defined in the literature. The Economics of Ecosystems and Biodiversity (TEEB; Silvestri, 2010) and Common International Classification of Ecosystem Services (CICES; Haines-Young and Potschin-Young, 2018), for example, do not acknowledge supporting services as their own category, but rather, combine them with regulating services. Common examples of supporting services in the literature include soil formation and retention, nutrient cycling, primary production, net primary productivity, atmospheric oxygen formation, provision of habitat, and N mineralization (Ghaley et al., 2014; Jin et al., 2017; Millennium Ecosystem Assessment (MEA), 2005). A few studies consider carbon storage and sequestration supporting services (Dumbrell et al., 2016; González-Díaz et al., 2019), though these two services are more often grouped with regulating services. Abson and Termansen (2011) include environmental resistance, or the capacity of a system to withstand disturbance without a qualitative change in system state, in their list of supporting ecosystem services. Though the Ecosystem Goods and Services (EGS) framework acknowledges that supporting services support "higher-level" ES, their example of supporting ES is limited to "habitat services," which they argue are foundational (Greenland-Smith et al., 2016). Barot (2017) identifies supporting services by their function and restricts the use of the term "service" to end-product services.

Though ecosystem functions are often used synonymously with services, not all functions translate to services. Ecosystem disservices are unwanted or harmful effects that stem from processes and functions. They can impact humans by causing damage or cost to other services or other ecosystems (Sandbrook and Burgess, 2015; Shapiro and Báldi, 2014). Focusing on services alone and ignoring disservices can be misleading (Dunn, 2010; Heink et al., 2016). For example, nitrogen (N) mineralization is generally defined as a service; without N mineralization, primary production would be dramatically reduced. N mineralization, though essential for ecosystem productivity, can be a disservice under N-saturated conditions, when increasing mineralization increases the potential for nutrient losses via leaching (Dijkstra et al., 2006). Leaching of mineral nutrients from a system is not only a lack of regulation of nutrient cycling or water purification within that system, but it also has potential negative impacts outside of that system (Smith and Schindler, 2009). Additionally, the process of mineralization associated with decomposition decreases the stock of soil organic matter and releases CO_2 (Lal, 2004). The unintended side effects, or disservices, of N mineralization, leaching (N losses from the system) and soil respiration (C losses from the system), must be acknowledged when considering nutrient cycling as a supporting service.

2.2 Anthropogenic Impacts on Supporting Services

Though ES link human societies to ecological systems, they are not consistently or comprehensively applied to ecosystems that are heavily under human influence (Barot et al., 2017), despite the fact that most ecosystems are managed, and all ecosystems are impacted in some way by humans (Vitousek et al., 1997). Barot (2017) argues that the MEA definition of ES does not properly account for the degree to which artificialization and intervention take place in the process of humans deriving services from natural systems. It is difficult to separate human manipulation and natural ecological functions. First, human activity directly impacts ES by altering ecological processes (Jin et al., 2017). Second, many ES are provided by interactions between ecological systems and human work or non-natural capital (Heink et al., 2016). Though ecological functions can still provide services, even in completely human-made systems, the amount of human manipulation required to maintain the provision of these services in the long term is important to acknowledge. When services cannot be attributed to natural ecological processes alone, human inputs must be considered.

Land-use change can lead to the degradation of supporting ES. Though technical measures can be implemented to replace these services, they can be costly (Liekens et al., 2013). Some argue that services provided under land-use practices that deteriorate ecosystems should not be considered services (Barot et al., 2017). For example, human management that encourages rehabilitation of degraded systems, such as the addition of fertilizer, may have impacts on biodiversity both within the system and on other downstream systems (Bullock et al., 2011).

A common example of the complexities of anthropogenic inputs required for the provision of supporting services is modern agriculture. Cultivated land is valued for its production of food, fiber, and fuel. That production, however, not only requires natural ecological processes but also often depends on human inputs including fertilizers, pesticides, and non-renewable sources of energy. Therefore, human manipulation in these systems provides goods at an ecological cost: additional emissions of greenhouse gases, potential nutrient leaching and runoff and their downstream effects, soil erosion due to degraded soil structure, and losses of soil fertility. Further, loss of soil fertility requires chemical fertilizers to overcome (“Hidden Costs of Industrial Agriculture,” n.d.). Wide use of fertilizer contributes to the global enrichment of nitrogen and phosphorus in ecosystems (Cordell et al., 2009; Vitousek et al., 1997). If constant human inputs are required for the upkeep of a particular service, the valuation of that service must acknowledge the effects of management and reduce the overall value of that service.

2.3 Valuation of Supporting Services

Though the ES framework is commonly applied in economic valuation, several authors are against the utilitarian interpretation and subsequent commodification of nature that the ES framework promotes (van den Belt and Stevens, 2016). Robinson et al. (2013) argue against economic ecosystem valuation, because of its focus on human welfare; human action and intervention are valued, not the ecosystem or ecosystem services that they affect. In addition to the ethical implications of economic valuation, there is the issue of measuring a combination of ecosystem services - or natural capital - and human or physical capital (Fisher et al., 2008). Often a *change* in the service or natural capital is valued rather than the service itself (Toman, 1998).

Many argue that supporting services are intermediate services (Xue and Tisdell, 2001) and should be included as part of the other three ES categories; therefore they should be indirectly quantified to avoid double-counting (Barot et al., 2017; D'Amato et al., 2016; Francesconi et al., 2016). Nevertheless, this study acknowledges supporting ES due to their foundational nature to other services, despite the indirect uses society derives from them. Barot's (2017) decision to use ecosystem function rather than supporting services avoids double-counting. More importantly, it allows for the analysis of ecosystem functions, which interact with themselves and other system attributes in complex, non-linear, and hard-to-predict ways, some of which may be considered services or disservices.

While there are multiple examples of supporting services given in the literature, this study focuses on three broadly acknowledged and accepted examples of supporting services: soil formation, nutrient cycling, and primary production. I follow Barot's (2017) definition of supporting services as functions, utilizing an ecosystem function multifunctionality approach to inform these supporting services. The negative impacts of some functions were considered, along with the ecological cost associated with the management of each land-use type. Instead of focusing purely on services provided, I provide an example of a "net services" approach, which considers the negative impacts of disservices as well as the negative ecological impacts of human management.

3. Research Goals and Methods

3.1 Study System

Blandy Experimental Farm (BEF), a 700-acre research facility in northwestern Virginia (39.06^o N, 79.07^o W) contains multiple land-uses under variable human management and therefore presents a unique opportunity to examine ecosystem properties, processes, and functions. These data provide a foundation to comprehensively evaluate and compare a suite of functions that inform supporting services provided by an ecosystem while also considering the role and cost of human management in that ecosystem. Ecosystem functions of net nitrogen (N) mineralization, primary productivity, decomposition, soil respiration, and N leaching were estimated over the course of 1-2 growing seasons in three different land-use types: an agricultural field (cultivating millet for the duration of the first growing season of the study and left bare for the duration of the second growing season), a restored native C_4 tallgrass prairie, and an early successional field.

3.2 Research Objectives

The ES conversation needs to invest more effort in addressing the full ensemble of processes and feedbacks that allow ecosystems to contribute to human wellbeing (Sutherland 2017). Given that supporting services tend to be overlooked, these foundational services, which are critical for other ES, will be the focus of this research. It is imperative to determine productive and helpful ways to measure and evaluate ecosystem processes, functions, and their drivers, in order to improve assessments of ES (Costanza et al., 2017). The multiple land-use types at BEF under the same climate and geological conditions provide an opportunity to comprehensively do so. Three commonly agreed upon supporting services were chosen to be the focus of this study: soil formation, nutrient cycling, and primary production. A suite of eight biogeochemical properties, one process, and five functions that underlie these three supporting ES were then examined among three land-use types at BEF. Properties, processes, and functions (see Chapter 1 for definitions) were grouped by location: aboveground, belowground, and the aboveground-belowground interface. The properties and processes of each land-use type serve to place the functions in context. The resultant functions are used to inform the relative amount of supporting services provided by each land-use type.

The questions this research aims to answer are inspired by Barot's (2017) use of ecosystem function to quantify supporting services rather than attempting to quantify the services themselves, as well as van den Belt's (2016) argument against economic valuation:

1. Using ecosystem function multifunctionality as a framework for evaluation, which land-use types appear to best provide three supporting services of primary production, soil formation, and nutrient (N) cycling?
2. Does considering the human management required for the upkeep of each land-use type alter desirability of land-use types for their provision of supporting services?

The multifunctionality approach used here considers detrimental functions, such as nutrient leaching. Supporting services, being intermediate services, avoid the implication of stakeholders and subjective appreciation and valuation (van den Belt, 2016). The decision to focus on supporting services maintains an objective, purely ecological valuation. Furthermore, the subjectivity associated with attempts to measure supporting services themselves is avoided by utilizing a suite of ecosystem functions to inform supporting services.

3.3 Study Design

Research plots were located in three different land use types: restored native prairie, early successional field, and agricultural field. The area of focus within each field was approximately 1 hectare. Five representative 5 m x 5 m plots were randomly chosen in each of the three fields. Within each 5 m x 5 m plot, three 1 m^2 subplots were randomly chosen for replicate sampling.

In the fall of 1999, BEF began a native prairie restoration project on approximately 15 hectares of land. They conducted herbicide treatments and prescribed burns in order to favor native species over already-established exotic species. Prior to that, the area was an early successional field dominated by non-native plant species. BEF continues to conduct prescribed burns in the native prairie every three years to prevent encroachment of competitive non-native species. The continued existence of the native plant meadow is reliant upon human manipulation. Therefore, the benefits provided by this land-use type need to be adjusted to acknowledge the ecological cost of the controlled burns, in part as carbon loss from the system.

The early successional field was brought to seedbank in 2002. Prior to that, the area was used for research on habitat fragmentation in rodent populations. Unlike the native prairie, the early successional field has involved no human management since its establishment. As a result, non-native plants dominate the early successional field. The supporting services provided by this land-use type will differ as a result of the difference in species composition, among other things. Its benefits, however, are not diminished by human management, as there is no management-related ecological cost associated with the continued existence of this land-use type.

The agricultural field has been under the management of the same farmer for the past 17 years and was also used for agriculture prior to that. Over the past 17 years, the field has been planted with rotations of crops, including corn, winter wheat, and millet. Fertilizer is applied as a top dressing at the beginning of spring each year, and herbicide is applied the week prior to planting. The supporting services provided by this land-use type will differ from the less managed systems, and the level of human management required offsets its benefits. There is ecological cost associated with fertilizer and herbicide applications; bioavailable nitrogen is released to the environment through the fertilizer itself, and carbon dioxide and nitrous oxide are released to the atmosphere through the burning of fossil fuels associated with the use of motorized equipment for fertilizer and herbicide applications. There are additional carbon costs with the use of equipment during planting and harvesting.

3.4 Quantifying ecosystem properties, processes, and functions

3.4.1 Aboveground properties, processes, and functions

Species composition was determined for each plot in the native prairie and early successional fields using the point-intercept sampling method (Hails and Ormerod, 2013). A 5 m x 5 m grid was established with 0.5 m intervals, including plot edges, for a total of 121 points of intersection. At each point, a dowel was dropped, and overstory and understory species touching it were identified. The total observation frequency of each species in each plot was recorded and compared to the total number of hits (the inclusion of overstory and understory species resulted in a total number of hits greater than 121; the total ranged from 196 to 286) to calculate relative frequency of each species in each plot. Observed species were separated by native and non-native classification, photosynthetic pathway, and plant functional type in order to calculate the relative frequencies of each (Concilio et al., 2016).

Aboveground biomass was harvested from an undisturbed corner of each subplot at peak seasonal biomass in each field; the early successional field in mid-August of 2017 and 2018, the native plant meadow in late August 2017 and 2018, and individual millet plants from the agricultural field immediately before harvest in 2017. The agricultural field was left bare in 2018 due to weather issues. Samples were oven-dried and sorted into plant functional and tissue types (forbs, graminoids, shrubs - foliar and woody, and standing dead), and mass was determined for each group. Peak season aboveground foliar biomass components were used to estimate aboveground net primary productivity (ANPP), according to Briggs and Knapp (1991). The mass of all live graminoids, forbs, and the foliar component of deciduous shrubs (evergreen shrubs were absent from research plots) were used to estimate ANPP.

From the 2017 harvested biomass, representative subsamples of foliar components from each functional type were ground. Approximately 5 mg were taken from the ground subsamples for dry combustion in a CN analyzer (Flash 2000; Thermo Fisher Scientific, Inc., Milan, Italy). Using %C and %N, the C:N ratio of live foliage of each functional type was determined.

3.4.2 Aboveground-belowground interface properties, processes, and functions

The rate of decomposition was estimated as the ratio of the mass of seasonal litter inputs to the mass of the existing litter pool. Seasonal litter inputs were estimated from live deciduous foliar biomass at the end of the growing season, under the assumption that all foliar biomass would become litter. The mass of the litter pool was estimated through litter collection in an undisturbed corner of each subplot at the start of the 2018 growing season.

3.4.3 Belowground properties, processes, and functions

Soil moisture measurements, recorded as volumetric percent water within the top 12 cm of soil, were taken using a Hydrosense handheld time domain reflectometer probe (Campbell Scientific, Inc., Logan, UT). Soil temperature measurements were taken at a depth of 10 cm using a digital thermometer. Measurements of both were taken at multiple times throughout the summer 2018 growing season in the three replicate subplots within the five plots of the native prairie and early successional field. Because plot establishment in the agricultural field was delayed due to planting schedule, measurements were delayed at that location until late July. Local landscape-scale precipitation data were used to supplement the soil moisture dataset.

Soils were sampled to a depth of 30 cm at 10 cm increments to measure total carbon (C) and nitrogen (N), as well as soil bulk density. Soil samples were passed through a 2 mm sieve to homogenize, and rocks and roots were removed and weighed. The remaining homogenized soil sample was also weighed, and a representative subsample was taken and oven-dried until each sample reached a constant weight. The wet versus the dry weight of the subsample was used to estimate the dry weight of the full soil sample. Soil bulk density was calculated as the ratio of soil dry mass to volume by subtracting the density-based volume of rocks and roots from the volume of the entire soil core, and subtracting the mass of rocks and roots from the mass of the full soil sample. Approximately 20 mg of soil were taken from the representative oven-dried subsample for dry combustion in a CN analyzer. Using %C and %N, C:N ratios in each 10 cm incremental depth were determined, as well as absolute amounts of C and N using bulk density.

As described above, when soil samples were passed through a 2 mm sieve to homogenize, roots were removed, cleaned, and weighed. The total mass of roots within each 10 cm incremental depth to 30 cm was determined for each land use type.

Three one-month long net N mineralization (NNM) incubations were conducted according to the resin-core method of DiStefano and Gholz (1986). The incubations took place in mid-June and mid-July of 2017 and mid-July of 2018. Soil samples were collected and immediately frozen to minimize microbial activity. In the laboratory, samples were homogenized, and inorganic NH_4^+ and NO_3^- were extracted using 2M KCl solution (Baer and Blair, 2008), and samples were run through a Lachat autoanalyzer (Lachat QuikChem 8500; Hach Company, Loveland, CO) to detect NO_3^--N and NH_4^+-N. The concentrations of NO_3^--N and NH_4^+-N were compared to the response curves from the analyzer of standards of known concentration. Because sample concentrations often exceeded the threshold of the response curves for the analyzer, all samples were diluted with 2 M KCl to fall within the range of the response curves. The detection limit for this method was 0.005 mg/L for NO_3^--N and NH_4^+-N, so any concentration below that was assumed to be zero.

A leaching potential index was calculated using total soil NO_3^--N and NH_4^+-N quantities in 10 cm soil layers to 30 cm depth in relation to the mass of roots within and below each soil layer. Quantities of soil NO_3^--N and NH_4^+-N were divided by the mass of roots within and below that soil layer; the greater the root mass present within and below the layer, the lower the potential for leaching. Therefore, for example, a lower value represents a lower potential for leaching, through a combination of a lower quantity of available soil NO_3^--N and NH_4^+-N and a greater quantity of roots to inhibit leaching of reactive nitrogen.

In situ soil CO_2 efflux was measured with a portable infrared gas analyzer with attached soil respiration chamber (EGM-4; PP Systems, Amesbury, MA), to serve as an estimate of soil respiration. Measurements were taken mid-day between 10h and 14h once per week during May and June in the three replicate subplots within the five plots of both the native prairie and early successional field. Once plots were established in the agricultural field in late July, measurements were taken in the three replicate subplots within the five plots of each land-use type on a weekly basis through August. The time of day at which land-use types were sampled was rotated in order to eliminate any time of day sampling bias.

3.5 Informing supporting services through a suite of ecosystem functions

The MEA (2005) regards supporting services as production, fluxes, and stocks moreso than benefits. Though there are applicable metrics of fluxes and stocks, vague terms such as "soil formation" are less likely to have agreed-upon metrics. Additionally, many services do not have applicable metrics. Inspired by each of these realities, ecosystem functions that were measured and estimated were utilized to inform supporting services.

Functions associated with the final supporting service were identified and applied using a multifunctionality framework, according to Manning (2018). The multifunctionality framework uses z-scores based on means and standard deviations for individual fields under each land-use type across both years of data collection. Normalized means were then added (or subtracted in the case of detrimental functions like nutrient leaching) for each type of supporting ES: soil formation, nutrient cycling, and primary production.

3.5.1 Soil formation

Costanza et al. (1997) used the broad ecosystem function term of "soil formation" as an indicator of the ecosystem service of soil formation, which includes the weathering of rock and the accumulation of organic material. Ghaley et al. (2014) estimated *in situ* bio-physical quantities of soil formation via count and weight of earthworms, which the authors assumed was proportional to the amount of annual topsoil turnover. Others (Barrios, 2007; Bell et al., 2005; Gianinazzi et al., 2010; Guimarães et al., 2010; Smukler et al., 2010; van Eekeren et al., 2010) focus on belowground microbial communities to estimate soil formation. The supporting service of soil formation, an intermediate step toward more concrete ES, is often estimated through final products such as provisioning services (Antle and Stoorvogel, 2006; Dale and Polasky, 2007;

Power, 2010; Sandhu et al., 2010; Stallman, 2011; Swinton et al., 2007; Zhang et al., 2007). Others focus on soil properties, processes and functions, including soil organic carbon, roots (Powlson et al., 2011), and nutrient cycling (Andrews et al., 2004).

Rather than use provisioning services or soil biota as proxies for soil formation, a multifunctionality framework was used. The supporting service of soil formation was informed by the multiple functions that take place at the aboveground-belowground interface and belowground: decomposition, soil respiration, net N mineralization (NNM), and N leaching. Because soil respiration is associated with decomposition, it was considered beneficial in the context of soil formation, despite the fact that it is associated with C loss from a system. N leaching, a negative side effect of NNM, was considered detrimental in the context of soil formation. Therefore, soil formation multifunctionality is the sum of the normalized means of decomposition, soil respiration, and NNM, less N leaching (Equation 1).

$$SF = z_D + z_{SR} + z_{NNM} - z_L$$ **Equation 1**

Where SF is soil formation multifunctionality

z_D is the normalized mean of decomposition in each field

z_{SR} is the normalized mean of soil respiration in each field

z_{NNM} is the normalized mean of NNM in each field

z_L is the normalized mean of leaching in each field

Decomposition and NNM were estimated for both the 2017 and 2018 growing seasons, whereas soil respiration and leaching were only measured in 2018. In addition to the ecosystem functions used in the multifunctionality analysis, the belowground properties of soil temperature and moisture, plant tissue quality, and soil organic matter quality provide context for this supporting ecosystem service provided by each land-use type.

3.5.2 Nutrient cycling

Costanza et al. (1997) used the ecosystem functions of storage, internal cycling, processing, and acquisition of nutrients, which included nitrogen fixation, N cycling, and other elemental and nutrient cycles, as indicators of the ecosystem service of nutrient cycling. Ghaley et al. (2014) measured *in situ* bio-physical quantities of nitrogen mineralization using three variables: feeding activity of microbes on bait lamina probes in the top 10 cm of soil, elemental analysis of soil C and N, and soil temperature and moisture before and after the bait lamina feeding.

Similar to the work of both Costanza et al. (1997) and Ghaley et al. (2014), the N cycling function of NNM was applied in a multifunctionality framework along with other ecosystem functions related to nutrient cycling. The supporting service of nutrient (N) cycling is informed by the multiple functions that take place at the aboveground-belowground interface, belowground, and aboveground: primary productivity, decomposition, soil respiration, NNM, and N leaching. The functions of soil respiration and N leaching were treated in the same manner as they were for soil formation (added and subtracted, respectively). N cycling multifunctionality is therefore the sum of the normalized means of primary productivity, decomposition, soil respiration, NNM, less N leaching (Equation 2).

$$NC = z_{PP} + z_{D} + z_{SR} + z_{NNM} - z_{L} \qquad \textbf{Equation 2}$$

Where NC is N cycling multifunctionality

z_{PP} is the normalized mean of primary productivity in each field

z_{D} is the normalized mean of decomposition in each field

z_{SR} is the normalized mean of soil respiration in each field

z_{NNM} is the normalized mean of NNM in each field

z_{L} is the normalized mean of leaching in each field

The multifunctionality analysis of the 2017 growing season did not include soil respiration and N leaching. All five functions were, however, included in the multifunctionality analysis for 2018. In addition to the ecosystem functions used in the multifunctionality analysis, the above- and below-ground properties of species composition, plant tissue quality, soil temperature and moisture, and soil organic matter quality provide context for the supporting ecosystem service of N cycling provided by each land-use type.

3.5.3 Primary Production

Though primary productivity is often quantified, it is much less frequently quantified in the context of supporting ES. Liu et al. (2018) estimate annual C fixation within the context of ES by using the i-Tree Eco model that uses tree and shrub measurements. Gonzalez-Diaz (2019) also used a modeling approach, in which they determined key factors that impact C storage and sequestration, including forest structure, climate, and biodiversity. They also implemented an economic valuation of the yearly supporting service of carbon sequestration, considering risk, conservation, and management factors. Rather than utilize models to estimate primary production, the field-estimated ecosystem function of aboveground primary productivity was used to inform the service of primary production. In addition to the aboveground primary productivity, the aboveground properties of species composition and plant tissue quality provide context for the supporting ecosystem service of primary production provided by each land-use type.

$$PP = z_{PP} \qquad \textbf{Equation 3}$$

Where PP is primary production

z_{PP} is the normalized mean of aboveground primary productivity in each field

Figure 1. Properties and processes that were used directly in the estimate of functions are marked with **; the others are properties and processes that provide context for supporting services.

	PROPERTIES & PROCESSES	**FUNCTIONS**	**SUPPORTING SERVICE**
ABOVEGROUND	• Species composition • Aboveground live & standing dead biomass ** • Plant tissue quality	• Aboveground primary productivity	• Nutrient cycling • Primary production
INTERFACE	• Litter mass ** • Seasonal litter inputs **	• Decomposition	• Nutrient cycling • Soil formation
BELOWGROUND	• Soil temperature & moisture • Root biomass ** • Soil organic C & N • Soil inorganic NH_4^+ and NO_3^-	• Net N mineralization • Soil respiration • N leaching	• Nutrient cycling • Soil formation

3.6 Valuation

Following the multifunctionality analysis of ecosystem functions that informs the supporting ES of primary production, soil formation, and nutrient (N) cycling, the multifunctionality scores of each supporting ES provided by each land-use type were considered separately and as a sum. Additionally, the human management required for the maintenance of each land-use type was considered: the early successional field, which required no inputs, has no associated ecological cost; the native prairie, which requires controlled burns every three years, has a small associated ecological cost; the agricultural field, though less demanding of resources than industrial farming projects (for example, this individual farmer follows no-till practices), has a higher associated ecological cost, as its maintenance still requires the input of N-based fertilizer, herbicides, and the use of motorized farm equipment.

The native prairie is burned via a prescribed burn every three years. Prescribed burns are much less severe than naturally occurring wildfires and are therefore not associated with significant combustion of soil C (DeBano, 1991; Neary et al., 1999). Additionally, because the majority of C is stored underground in grasslands, the C released from the system through fire is small relative to the amount that remains stored belowground (Dass et al., 2018). Standing stock of aboveground biomass rarely exceeds a few US tons per hectare (Penman et al., 2003).

Winter wheat and oats each use approximately 10 GJ of fossil fuel energy per hectare (Samson et al., 2005), which is associated with about 0.7 T CO_2 and 0.9 T CO_2 from diesel fuel oil and coal electricity generation, respectively. Corn cultivation uses ~15 GJ of fossil fuel energy per hectare (Samson et al., 2005), which is the equivalent of approximately 1.1 T CO_2 and 1.4 T CO_2 from diesel fuel oil and coal electricity generation, respectively. Though it is difficult to directly quantify the ecological cost of management, these statistics from the literature provide some insight into the cost associated with the management of each land-use type.

If all of the aboveground biomass in the native prairie were burned during a controlled burn, a high estimate of carbon lost (based on the IPCC estimate) would be 3 tons per hectare. As burns are conducted every three years, a high estimate of the carbon lost would be 1 ton of C lost per hectare on an annual basis. By comparison, if a cereal crop and corn are cultivated in the same growing season, the carbon cost of management, based on Samson et al.'s (2005) estimated ecological cost, would total about two tons of CO_2. These estimates of ecological cost are inexact and are both likely greater than the actual ecological cost, but they provide some information about the ecological cost associated with the management of each land-use type; the average estimated ecological cost of the agricultural field is approximately twice as high as the native prairie, while there is no ecological cost of management in the early successional field. By this standard, valuation was based on the comparison of multifunctionality of supporting ES and the ecological cost of management; i.e. the aggregated suite of supporting ES provided by the agricultural field should be about twice that of the native prairie in order for it to compensate for the ecological cost associated with its management. Likewise, the agricultural field and native prairie should demonstrate greater multifunctionality than the early successional field in order to compensate for their ecological costs.

3.7 Statistical Analyses and Data Visualization

Analyses of variance (ANOVA) and repeated measures ANOVA (RMANOVA), where applicable, were performed to compare the following variables that express ecosystem properties across land-use types: soil moisture and temperature, C:N ratios in each ten cm soil depth increment and in foliar plant tissue, mass of roots in each ten cm soil depth increment, and community diversity indexes. ANOVA and RMANOVA, where applicable, were performed to compare the ecosystem functions of net N mineralization, primary productivity, soil respiration, decomposition, and N leaching.

For variables that demonstrated significant differences among groups and did not violate ANOVA assumptions, a Tukey test was performed for all pairwise comparisons. For variables that violated ANOVA assumptions, a Kruskal-Wallace test was performed, followed by a post-hoc Nemenyi test with a Tukey-Kramer distribution using the R Package PMCMRplus (Pohlert, 2018).

4. Results

4.1 Properties, Processes, and Functions

4.1.1 Aboveground

Aboveground properties of plant tissue quality and species composition provide context for aboveground functions as well as the supporting services of nutrient cycling and primary productivity. Forbs in the native prairie had significantly ($p < 0.05$) greater mean C:N ratios (51.0 ± 3.2) than in the early successional field (35.2 ± 1.2). The plant tissue quality of graminoids did not differ among the early successional field and native prairie, but both had greater C:N ratios (30.7 ± 2.8 and 52.0 ± 9.0, respectively) than the agricultural field (22.5 ± 2.0). The majority (93 ± 4%) of plants in the early successional field were C_3 plants, while less than half (44 ± 6%) in the native prairie were C_3. Greater than half (57 ± 7%) of the plants in the native prairie were native to Virginia, while only about a quarter (27 ± 6%) of the plants in the early successional field were native. The early successional field had significantly ($p < 0.05$) more forbs than the native prairie (48 ± 10% compared to 20 ± 5%), while the native prairie had significantly ($p < 0.05$) more graminoids than the early successional field (58 ± 7% compared to 25 ± 11%; Table 1).

Table 1. Aboveground properties, which provide context for aboveground functions and subsequently services of nutrient cycling and primary productivity. Mean values and standard error are given for each field and each growing season, where applicable. Significant differences are marked with different letters.

Property	**Year**	**Agricultural**	**Early Successional**	**Native Prairie**
Foliar tissue quality (unitless C:N ratio)				
graminoids	2017	22.5 ± 2.0 ^{a}	30.7 ± 2.8 ^{b}	52.0 ± 9.0 ^{b}
forbs		---	35.2 ± 1.2 ^{a}	51.0 ± 3.2 ^{b}
Species composition (%)				
C_3 photosynthetic		---	93 ± 4 ^{a}	44 ± 6 ^{b}
native	2017	---	27 ± 6 ^{a}	57 ± 7 ^{b}
forb functional type		---	48 ± 10 ^{a}	20 ± 5 ^{b}
graminoid functional type		---	25 ± 11 ^{a}	58 ± 7 ^{b}

Total foliar biomass, used as an approximation for aboveground net primary productivity (ANPP), was significantly ($p < 0.001$) greater in the agricultural field (1250 ± 155 g m^{-2}) than in the native prairie (208 ± 15 g m^{-2}) and early successional field (343 ± 45 g m^{-2}) during the 2017 growing season. ANPP was significantly ($p < 0.001$) greater in the native prairie (262 ± 31 g m^{-2}) and early successional field (265 ± 42 g m^{-2}) than in the agricultural field (0 ± 0 g m^{-2}) during the 2018 growing season, when the agricultural field was not planted (Table 2).

Table 2. Aboveground functions, which were subsequently used to inform services of nutrient cycling and primary productivity. Mean values and standard error are given for each field and each growing season. Significant differences are marked with different letters.

Function	**Year**	**Agricultural**	**Early Successional**	**Native Prairie**
Primary productivity (g m^{-2})	2017	1250 ± 155 ^{a}	343 ± 45 ^{b}	208 ± 15 ^{b}
	2018	0 ± 0 ^{a}	265 ± 42 ^{b}	262 ± 31 ^{b}

4.1.2 Aboveground-belowground interface

During 2017, the rate of decomposition, estimated as the ratio of the mass of seasonal litter inputs to the mass of the existing litter pool, was significantly greater in the early successional field (3.7 ± 1.1 y^{-1}) than in the agricultural field (0.0 ± 0.0 y^{-1}) and native prairie (0.9 ± 0.1 y^{-1}). During 2018, the rate of decomposition was significantly ($p < 0.05$) greater in the early successional field (2.7 ± 0.5 y^{-1}) and native prairie (1.1 ± 0.1) than in the agricultural field (0.0 ± 0.0 g m^{-2}). The high rate of decomposition in the early successional field is based on its high seasonal litter inputs (343 ± 45 g m^{-2} in 2017 and 265 ± 42 g m^{-2} in 2018) paired with an intermediate litter pool (109 ± 22 g m^{-2}) at the start of the 2018 growing season. The native prairie had intermediate to high litter inputs (208 ± 15 g m^{-2} in 2017 and 262 ± 31 g m^{-2} in 2018) paired with a relatively large litter pool (258 ± 29 g m^{-2}).

Table 3. Functions at the aboveground-belowground interface, which was subsequently used to inform the services of nutrient cycling and soil formation. Mean values and standard error are given for each field and each growing season. Significant differences are marked with different letters.

Function	**Year**	**Agricultural**	**Early Successional**	**Native Prairie**
Decomposition (y^{-1})	2017	0.0 ± 0.0 ^{a}	3.7 ± 1.1 ^{b}	0.9 ± 0.1 ^{a}
	2018	0.0 ± 0.0 ^{a}	2.7 ± 0.5 ^{b}	1.1 ± 0.1 ^{b}

4.1.3 Belowground

During the 2018 summer growing season, average soil temperature was significantly greater in the agricultural field (26.30 ± 0.10 ^{o}C) than in the early successional field (23.70 ± 0.35 ^{o}C), which was significantly greater than the native prairie (22.84 ± 0.15 ^{o}C). Average soil moisture was significantly greater in the native prairie (32.20 ± 1.53%) than in the agricultural field (29.62 ± 1.00%).

Ratios of C:N in the top 20 cm of soil differed among fields; there were no significant differences among fields in the 20 cm to 30 cm depth. In the top 10 cm of soil, the agricultural field and early successional field had lower C:N ratios (9.78 ± 0.13 and 10.08 ± 0.14) than the native prairie (12.82 ± 0.37). In the 10 – 20 cm soil depth range, the agricultural field and early successional field also had lower C:N ratios (9.94 ± 0.11 and 10.06 ± 0.08) than the native prairie (12.55 ± 0.13).

Table 4. Belowground properties, which provide context for belowground functions and subsequently services of nutrient cycling and soil formation. Mean values and standard error are given for each field and each growing season, where applicable. Significant differences are marked with different letters.

Property	**Year**	**Agricultural**	**Early Successional**	**Native Prairie**
Temperature (^{o}C)	2018	26.3 ± 0.1 ^{a}	23.7 ± 0.4 ^{b}	22.8 ± 0.2 ^{c}
Moisture (% water by vol)	2018	29.6 ± 1.0 ^{a}	31.3 ± 1.9 ^{ab}	32.2 ± 1.5 ^{b}
Soil C:N (unitless)				
0 – 10 cm	2017	9.8 ± 0.1 ^{a}	10.1 ± 0.1 ^{a}	12.8 ± 0.4 ^{b}
10 – 20 cm		9.9 ± 0.1 ^{a}	10.1 ± 0.1 ^{a}	12.6 ± 0.1 ^{b}
20 – 30 cm		10.8 ± 0.3 ^{a}	11.5 ± 0.4 ^{a}	11.6 ± 0.4 ^{a}

Average soil respiration rates were significantly greater in the early successional field (9.7 ± 1.6 g CO_2 m^{-2} h^{-1}) than in the agricultural field (7.2 ± 1.0 g CO_2 m^{-2} h^{-1}), which had significantly greater rates than the native prairie (6.1 ± 0.6 g CO_2 m^{-2} h^{-1}). Net N mineralization was greater in the agricultural field (0.4 ± 0.6 g N m^{-2} $month^{-1}$) than in the native prairie and early successional field, though there were no significant differences among fields due to high variability. Leaching potential index, which was based on the ratio of total soil NH_4^+ and NO_3^- in each soil depth profile and the total mass of roots within and below that depth, was significantly greater in the agricultural field than in the early successional field and native plant meadow, due to a combination of a lack of roots as well as greater soil NH_4^+ and NO_3^- quantities; the native prairie had significantly more roots than the agricultural field, while the early successional field had intermediate root biomass (Table 5).

Table 5. Belowground functions, which were subsequently used to inform services of nutrient cycling and soil formation. Mean values and standard error are given for each field and each growing season. Significant differences are marked with different letters.

Function	Year	Agricultural	Early Successional	Native Prairie
Soil respiration (g CO_2 m^{-2} h^{-1})	2018	7.2 ± 1.0 ^{a}	9.7 ± 1.6 ^{b}	6.1 ± 0.6 ^{c}
Net N mineralization (g CO_2 m^{-2} h^{-1})				
Early summer	2017	---	0.32 ± 0.21 ^{a}	0.38 ± 0.25 ^{a}
mid summer		0.4 ± 0.6 ^{a}	0.01 ± 0.26 ^{a}	-0.05 ± 0.021 ^{a}
Net N mineralization (g CO_2 m^{-2} h^{-1})				
mid summer	2018	0.25 ± 0.23 ^{a}	-0.05 ± 0.23 ^{a}	0.06 ± 0.27 ^{a}
Leaching potential index (unitless)	2018	0.14 ± 0.06 ^{a}	0.06 ± 0.04 ^{b}	0.01 ± 0.01 ^{b}

4.2 Ecosystem function multifunctionality and supporting services

4.2.1 Soil formation

The belowground and aboveground-belowground interface ecosystem functions of NNM, decomposition, respiration, and leaching were used in an ecosystem function-multifunctionality analysis to inform the supporting service of soil formation. During the 2017 growing season, soil formation did not differ significantly ($p > 0.05$) among fields. During the 2018 growing season, the early successional field had significantly ($p < 0.01$) greater soil formation (a sum of z-scores of 0.98 ± 0.52) than the agricultural field (a z-score sum of -1.73 ± 0.66), which lacked seasonal litter inputs and a surface litter pool. The native prairie demonstrated intermediate soil formation (a z-score sum of -0.68 ± 0.62).

4.2.2 N cycling

All five ecosystem functions observed in this study (NNM, primary productivity, decomposition, respiration, and leaching) were used to inform the service of nutrient cycling. During the 2017 growing season, the agricultural field had significantly ($p < 0.01$) greater nutrient cycling (4.37 ± 1.12) than the early successional field (1.06 ± 1.04) and native prairie (-1.23 ± 0.35). During the 2018 growing season, the early successional field had significantly ($p < 0.01$) greater nutrient cycling (0.37 ± 0.63) than the agricultural field (-3.67 ± 0.66), with intermediate nutrient cycling multifunctionality in the native prairie (-1.31 ± 0.69).

4.2.3 Primary production

The aboveground ecosystem function of primary productivity was used to inform the supporting service of primary production. During the 2017 growing season, the agricultural field had significantly ($p < 0.001$) greater productivity (a z-score of 4.29 ± 0.77) than the early successional field (a z-score of -0.22 ± 0.22) and native prairie (a z-score of -0.90 ± 0.08). During the 2018 growing season, the agricultural field had significantly ($p < 0.001$) lower productivity ($z = -1.94 \pm 0.00$) than the early successional field ($z = -0.61 \pm 0.21$) and native prairie ($z = -0.63 \pm 0.15$).

4.2.4 Suite of supporting ecosystem services

During the 2017 growing season, the agricultural field had significantly greater total multifunctionality (8.74 ± 2.24) than the native prairie (-2.46 ± 0.69). The early successional field had intermediate total multifunctionality (2.11 ± 2.09). During the 2018 growing season,

Table 6. Supporting services were informed by the z-scores of multiple functions. Mean values and standard error are given for each field and each growing season. Significant differences are marked with different letters.

Year	Supporting ES	Agricultural	Early Successional	Native Prairie
2017	Soil Formation	0.07 ± 1.20^{a}	1.28 ± 0.92^{a}	-0.33 ± 0.35^{a}
	Nutrient Cycling	4.37 ± 1.12^{a}	1.06 ± 1.04^{ab}	-1.23 ± 0.35^{b}
	Primary Production	4.29 ± 0.77^{a}	-0.22 ± 0.22^{b}	-0.90 ± 0.08^{b}
	Total	8.74 ± 2.24^{a}	2.11 ± 2.09^{ab}	-2.46 ± 0.69^{b}
2018	Soil Formation	-1.73 ± 0.66^{a}	0.98 ± 0.52^{b}	-0.68 ± 0.62^{ab}
	Nutrient Cycling	-3.67 ± 0.66^{a}	0.37 ± 0.63^{b}	-1.31 ± 0.69^{ab}
	Primary Production	-1.94 ± 0.00^{a}	-0.61 ± 0.21^{b}	-0.63 ± 0.15^{b}
	Total	-7.34 ± 1.32^{a}	0.75 ± 1.25^{b}	-2.62 ± 1.37^{ab}

when the agricultural field was not planted, the agricultural field had significantly lower total multifunctionality (-7.34 ± 1.32) than the early successional field (0.75 ± 1.25). The native prairie had intermediate total multifunctionality (-2.62 ± 1.37).

4.3 Valuation

Considering the multifunctionality provided by each land-use type across both years, the early successional field consistently provided a high multifunctionality; it demonstrated an intermediate multifunctionality during 2017 (2.11 ± 2.09) and the highest multifunctionality during 2018 (0.75 ± 1.25), averaging 1.43 standard deviations above the mean. The agricultural field was most variable; it demonstrated the greatest multifunctionality when planted in 2017 (8.74 ± 2.24) and the lowest multifunctionality when not planted in 2018 (-7.34 ± 1.32), averaging 0.70 standard deviations above the mean. The native prairie consistently demonstrated a lower multifunctionality; it had the lowest multifunctionality in 2017 (-2.46 ± 0.69) and an intermediate multifunctionality in 2018 (-2.62 ± 1.37), averaging 2.54 standard deviations below the mean.

Based on statistics from the literature, the annual carbon cost of management in the native prairie is about 1 ton per hectare, and the annual carbon cost of management (in terms of fossil fuel energy use) in the agricultural field is about 3 tons per hectare. There is no ecological cost of management in the early successional field. Therefore, the aggregated suite of supporting ES provided by the native prairie and agricultural field should be greater and much greater respectively than the early successional field. The mean multifunctionality of the agricultural field over the two years balanced out to near zero (near the mean multifunctionality), while the mean multifunctionality of the native prairie over the two years remained multiple standard deviations below the mean multifunctionality. The early successional field demonstrated the highest multifunctionality across both growing season, in addition to having the least ecological cost.

5. Discussion

5.1 Properties, processes, and functions as context for supporting services provided

The relatively high plant tissue quality (low C:N ratios) in the agricultural and early successional fields provided context for the aboveground supporting services of nutrient cycling and primary production. Organic matter inputs with low C:N ratios promote N mineralization (Aerts, 1995; Hobbie, 1992; Pastor et al., 1984; Uri et al., 2008; Vitousek, 1982). Likewise, C_4 tallgrass litter, such as that of the native prairie, promotes lower net N mineralization rates and N availability relative to some other plant types (Craine et al., 2002; Dijkstra et al., 2006; Tilman and Wedin, 1991; Wedin and Pastor, 1993; Wedin and Tilman, 1990). Plant biomass growth, and therefore the function of primary productivity, is also highly correlated with N pools and N cycling rates (Zak et al., 1990). Therefore, the early successional field and agricultural field should provide nutrient cycling and primary production to a greater extent than the native prairie. Based on multifunctionality analysis, the agricultural field provided the greatest supporting service of nutrient cycling in 2017 when planted. When the agricultural field was not planted in 2018, the early successional field provided the greatest supporting service of nutrient cycling. Likewise, when the agricultural field was planted in 2017, it provided the greatest supporting service of primary production. When it was not planted, the early successional field had the greatest primary productivity, though it did not differ significantly from the native prairie.

Decomposition rates provided some context for the supporting services of nutrient cycling and soil formation, though the multifunctionality analysis of each includes several functions in addition to decomposition. Estimated decomposition rates were highest in the early successional field in 2017 and were lowest in the agricultural field in both years. Based on decomposition alone, nutrient cycling and soil formation should both be high in the early successional field. In 2018, soil formation multifunctionality was greatest in the early successional field. In 2017, however, soil formation multifunctionality did not differ significantly among fields. This may be because soil respiration and N leaching were not measured in 2017 and therefore were excluded from the multifunctionality analysis. Nutrient cycling multifunctionality, which included the function of primary productivity, was greatest in the agricultural field in 2017, due to the fact that productivity in the agricultural field was about 4 standard deviations above mean productivity across both years in all fields, while the other functions included did not differ by much more than 2 standard deviations. Nutrient cycling was greatest in the early successional field in 2018, when the agricultural field did not have any productivity.

The relatively high soil quality (low C:N ratios) in the agricultural and early successional fields provided context for the belowground functions of soil respiration and NNM and, subsequently, the supporting services of nutrient cycling and soil formation. Soil respiration was greatest in the early successional field in the 2018 growing season, while NNM did not differ significantly among fields. The drier soils of the agricultural field likely impacted belowground functions such as soil respiration and net N mineralization, causing the agricultural field to have lower multifunctionality than the early successional field. Additionally, the higher leaching potential index in the agricultural field further diminished its multifunctionality. In 2018, when all functions were measured, the greatest multifunctionality indices informing both soil formation and nutrient cycling were in the early successional field. In 2017, when leaching and soil respiration were excluded from the multifunctionality analysis, soil formation did not differ among fields. The agricultural field demonstrated the greatest multifunctionality informing nutrient cycling.

5.2 Provision of supporting services

Using ecosystem multifunctionality as a framework for evaluation, the early successional field appears to provide the greatest soil formation and nutrient cycling in 2018. The agricultural field appears to provide the greatest nutrient cycling and primary production in 2017. In 2017 the total of multifunctionality scores was greatest in the agricultural field, intermediate in the early successional field, and least in the native prairie. In 2018 the total multifunctionality scores were greatest in the early successional field.

5.3 Valuation, the ecological cost of management, and implications

Considering human management required for the upkeep of each land-use type alters the desirability of land-use types for their provision of supporting services. The agricultural field, despite its cost of management, appears to be just as valuable, if not more valuable than the native prairie. While the agricultural field demonstrated the greatest multifunctionality in 2017, its average multifunctionality between the two years did not surpass that of the early successional field. Because the agricultural field and native prairie failed to demonstrate greater overall multifunctionality than the early successional field in order to compensate for their ecological cost of management, they do not appear as valuable in terms of the net supporting services they provide.

5.4 Technical limitations of the study and future work

The five functions used in this study are not an exhaustive list, but rather, a representative sub-sample of ecosystem functions that can be used to inform supporting services. Additionally, some were measured only during the 2018 growing season, leaving multifunctionality values of the 2017 growing season lacking robustness. Of the five functions, two were estimated rather than measured directly: decomposition and leaching. Decomposition was estimated by dividing seasonal litter inputs by the total litter pool, and leaching was approximated based on quantities of NH_4^+ and NO_3^- in each 10 cm increment of soil, compared to the roots within and below that depth. Future work could measure each of these functions more directly, over longer periods of time, and could include additional ecosystem functions in order to more holistically and exhaustively inform or even quantify supporting services.

Though utilizing z-scores normalizes disparate metrics, aggregating and ranking across these assumes equal weighting of functions. Using z-scores to analyze multifunctionality brings about two questions: First, is the importance of each function equal? Second, are the variations across functions equivalent? Equating the importance of productivity with the importance of soil respiration, for example, is not necessarily appropriate. Similarly, assuming that one standard deviation in decomposition, for example, carries the same weight as one standard deviation difference in leaching is likely not appropriate. Additionally, NNM, soil respiration, and decomposition are related ecosystem functions that tend to be correlated, so combining them in multifunctionality analyses potentially overestimates their overall importance in the suite of ecosystem functions.

The ecological cost of management in this study is loosely quantitative and based solely on fossil fuel use, and while it provides a general idea of cost of management, it is difficult to adjust multifunctionality values in order to interpret the results definitively. An examination of multiple land-use types under different land-use practices would be beneficial in future work to account for this. The question of how to integrate different variables under a common currency is a remaining gap in the ES conversation; comparing the ecological cost of gasoline for farming vehicles to the plant-available nitrogen produced through NNM is an example of such a scenario, though a carbon and nitrogen cost may be one useful approach for future work.

Multifunctionality analyses may be misleading, as they appear to value functions with faster rates. The results of this study demonstrate that the rates of functions in the native prairie are slower than those of the early successional field and agricultural field, making it appear less valuable in terms of the supporting ES it provides. The slow decomposition in the native prairie, paired with lower soil respiration, and lower productivity, is a defining ecological feature of C_4 prairies, and is not necessarily less valuable than other systems. Incorporating elemental stoichiometric ratios into a multifunctionality analysis could help account for the issue of rates of functions.

5.5 Benefits of the study

This study not only examines supporting services, which are often overlooked, but also attempts to examine them from an objective, ecological perspective. This is among the first of its kind to acknowledge two topics of interest in the scientific community examining ES: disservices and the ecological cost of management. I utilized a novel multifunctionality approach, grouping functions and their underlying properties and processes by aboveground, belowground, and the interface of the two areas. While supporting ES are not consistently defined in the literature and are measured even less consistently, this study attempts to fill this gap in the ES conversation, illuminating the path toward more objectively defined ecological research that can inform sustainable land-use decisions.

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Conclusion

The functions of NNM, decomposition, and soil respiration are all related. The early successional field, in which organic matter inputs had low C:N ratios, promoted N mineralization (though non-significantly greater) as well as significantly greater litter decomposition in both growing seasons and greater soil respiration than the other two land-use types in 2018. The native prairie demonstrated overall net N immobilization, particularly later in both growing seasons. The low rate of soil respiration in the native prairie can be contextualized by its low litter decomposition rate. Though the agricultural field lacked aboveground litter inputs, its low C:N ratios due to fertilization determine the relatively high NNM rates, and the balance of each resulted in intermediate rates of soil respiration. Plant biomass growth, also related to N pools and N cycling rates, was significantly greater in the agricultural field, because of its land-use management and N inputs via fertilizer. The native prairie, with its high soil C:N ratios, had the lowest aboveground productivity. In 2018, foliar production of the early successional field and native prairie were essentially similar. The agricultural field had the greatest leaching potential index values, while the native prairie had the lowest. While total concentrations of inorganic NH_4^+ and NO_3^- were generally greatest in the early successional field, aligning with its higher NNM rates, and lowest in the native prairie across all depths, leaching potential index was highest in the agricultural field due to the lack of roots in the field. The native prairie, which had higher belowground root mass, had the lowest overall leaching potential index.

The high C multifunctionality in 2017 in the agricultural field aligns with its very high productivity, and the intermediate C multifunctionality in the early successional field aligns with its high decomposition rates. Utilizing a C multifunctionality approach for the 2017 growing season provides a more holistic perspective of the multiple functions carried out by each land-use type. For example, a focus on the lower productivity in the early successional field in 2017 misses the fact that its decomposition, a function critical to others, is occurring at a greater rate than other land-use types. The inclusion of soil respiration in 2018 impacted C multifunctionality; while decomposition is a positive component of C multifunctionality, the inclusion of soil respiration as a negative side effect balanced out the benefits of decomposition. The particularly high soil respiration rates in the early successional field diminished its C multifunctionality in 2018 to a level similar to that of the native prairie.

N multifunctionality demonstrated the same patterns as C multifunctionality during 2017. This is likely due to the fact that NNM did not differ significantly among land-use types in 2017, so its inclusion did not largely impact multifunctionality. The absence of the function of leaching (a disservice not estimated in 2017) in the N multifunctionality analysis could have impacted the agricultural field, making it less desirable. In 2018, the early successional field and native prairie had the greatest and intermediate N multifunctionality, respectively. Though soil respiration, primary productivity, decomposition, and leaching were all lowest in the agricultural field and highest in the early successional field, the rates of soil respiration and decomposition in the native prairie did not differ significantly from the agricultural field. N multifunctionality captures the balance of the suite of traits provided by each land-use type.

Based on multifunctionality analysis in the context of supporting services, the agricultural field provided the greatest supporting services of nutrient cycling and primary production in 2017 when planted. Soil formation multifunctionality did not differ significantly among fields, possibly because soil respiration and N leaching were not measured in 2017 and therefore were excluded from the multifunctionality analysis. When the agricultural field was not planted in 2018, the early successional field provided the greatest supporting services of nutrient cycling, primary productivity (though it did not differ significantly from the native prairie), and soil formation.

When planted, the agricultural field appears to provide the greatest level of total multifunctionality, the sum of C and N multifunctionality, as well as the greatest level of the supporting ES of nutrient cycling and primary production. When the agricultural field was not planted, however, the early successional field surpassed it in total multifunctionality. The early successional field also demonstrated the greatest level of the supporting ES nutrient cycling, primary production, and soil formation. To compensate for the ecological cost of management, the aggregated suite of supporting ES provided by the agricultural field should far outweigh those of the native prairie and early successional field. The aggregated suite of supporting ES provided by the native prairie should also exceed that of the early successional field. The mean supporting ES multifunctionality of the agricultural field over the two years balanced out to near zero (near the mean multifunctionality), while the mean multifunctionality of the native prairie over the two years remained multiple standard deviations below the mean multifunctionality. The early successional field demonstrated the highest multifunctionality across both growing seasons with the least ecological cost. When the lack of ecological cost of management of the early successional field is considered, it appears even more desirable as a land-use type.

Though this project is a step toward elucidating the difficult task of quantifying supporting services in order to inform sustainable land-cover decisions, there are areas for improvement. The more conservative N use and longer nutrient cycling time scales in the native prairie may be causing it to be slightly undervalued under the multifunctionality framework. Incorporating elemental stoichiometric ratios into a multifunctionality analysis could help account for the issue of slow rates of functions. Additionally, the scope and duration of the study provides incomplete datasets to make generalizations on a broader, annual scale. Multi-year, frequent monitoring would be beneficial for more robust results, which could more thoroughly account for underlying properties and processes and their long-term interactions with functions.

Two questions remain about utilizing z-scores to analyze multifunctionality: First, is the importance of each function equal? Second, are the variations across functions equivalent? Equating the importance of productivity with the importance of soil respiration, for example, is not necessarily appropriate. Similarly, assuming that one standard deviation in decomposition, for example, carries the same weight as one standard deviation difference in leaching is likely not appropriate. Additionally, NNM, soil respiration, and decomposition are related ecosystem functions that tend to be correlated, so combining them in multifunctionality analyses potentially overestimates their overall importance in the suite of ecosystem functions.

The lack of a thorough exploration of supporting services and their underlying functions in the ES conversation provided the motivation for this study. Supporting services, though integral to the provision of other services, are often overlooked in the literature. This is the first study to approach supporting services in this way, attempting to quantify their underlying properties, processes, and functions, as well as combine multiple functions into a single metric based on means from multiple years, capturing interannual variability in conditions. This is also the first study to apply a multifunctionality framework to C-based and N-based functions. While questions as to the appropriateness and usefulness of multifunctionality remain, this work is a step toward elucidating the difficult task of quantifying supporting services, which remain important to consider in sustainable strategies.