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Carbon Dioxide Emissions from Switchgrass and Cottonwood Grown as Bioenergy Crops in the Lower Mississippi Alluvial Valley Carbon Dioxide Emissions from Switchgrass and Cottonwood Grown as Bioenergy Crops in the Lower Mississippi Alluvial Valley

> A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Crop, Soil, and Environmental Science

> > by

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### Abstract

Marginal land of the Lower Mississippi Alluvial Valley (LMAV) has the potential to be utilized for the production of bioenergy feedstocks. Soil respiration is the gaseous emission of carbon dioxide  $(CO_2)$  from microbes and plant roots in the soil, and these emissions play an important role in the global cycling of carbon. Soil respiration can act as a positive feedback affecting climate change, and has been shown to vary depending on soil moisture, temperature, and vegetation. The objectives of this study where to evaluate the effects of land use [switchgrass (Panicum virgatum), cottonwood (Populus deltoides), and a soybean (Glycine max)-grain sorghum (Sorghum bicolor) agroecosystem] on monthly soil respiration and estimated annual CO<sub>2</sub> emissions on a silt loam in east-central Arkansas throughout 2012 and 2013. Annual CO<sub>2</sub> emissions were calculated by linear interpolation between monthly measurements. Soil respiration from all three ecosystems followed the same general trend: increasing from January to May and decreasing from September to December, peak fluxes differed significantly (p < p0.05) among ecosystems for both years. Peak fluxes in 2012 were achieved for all three ecosystems in July. Soybean and switchgrass peak fluxes did not differ (8.1 and 7.6 µmol m<sup>-2</sup> s<sup>-1</sup> respectively) with cottonwood peak flux differing from other treatments (6.1  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>; p < 0.01). Peak fluxes for 2013 were achieved in May for both switchgrass and cottonwood (5.91 and 4.11  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, respectively), where the switchgrass peak flux was larger than that for cottonwood and the agroecosystem, which did not differ (p < 0.01). Annual CO<sub>2</sub> emissions differed among ecosystems (p < 0.001), but not between years (p = 0.45). Cottonwood had less CO<sub>2</sub> emitted for both years (7.3 and 7.4 Mg ha<sup>-1</sup> for 2012 and 2013, respectively) compared to the other two ecosystems, while emissions from the switchgrass did not differ from soybean in 2012 (10.3 and 9.5 Mg ha<sup>-1</sup>, respectively) or grain sorghum in 2013 (9.7 and 9.2 Mg ha<sup>-1</sup>,

respectively). Results showed established bioenergy feedstock cropping systems do not have greater soil respiration rates compared with a traditional soybean-grain sorghum crop rotation. Results also indicated that different bioenergy feedstocks can produce different quantities of  $CO_2$ emissions. Both factors are important to consider when enrolling marginal land in the LMAV in bioenergy feedstock cropping systems.

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Introduction

#### Introduction

Energy use has increased during the twentieth century. Not only are developed countries demanding more energy resources, developing countries are increasing their energy usage. Energy demands are expected to rise 50% by 2050 (Raguaskas et al., 2006). In order to meet the increasing demands for energy and keeping in mind concerns over national security, the United States (US) funded programs for the development of renewable energy that could be sourced from the country without imports. Corn (*Zea mays*)-based ethanol was a logical first stepping stone on the road to energy independence, but not a substitute for the majority of oil imports for the US. In general, cellulosic crops have greater biofuel yields and lower greenhouse gas (GHG) emissions per unit land area and per unit biofuel produced than conventional corn rotations (Adler et al., 2007).

Cell walls are the most abundant plant material on the planet (Vogel, 1996). The carbon (C)-rich combination of cellulose, hemicelluloses, and lignin has enormous energy potential. However, deriving energy from these compounds is much more difficult than traditional cornbased ethanol. In order to move forward in the commercialization of cellulosic biofuels, support from policy and industry is needed. In addition, increased funding for research and development of biomass crops and refining technologies is necessary to make biorefineries more efficient and sustainable (Raguaskas et al., 2006). Ethanol, gasoline, diesel, and electricity generation through gasification are also possible energy uses for cellulosic bioenergy. Currently, the most easily derived is electricity generation through gasification of biomass.

The biogeochemical cycling of carbon through the atmosphere, soil, and plant life was roughly balanced before human intervention, namely prior to the industrial era (Schlesinger and Andrews, 2000). However, the natural cycling of carbon through biogeochemical processes has

been disrupted by anthropogenic alterations to cycling through the combustion of fossil fuels. Conversion to sustainable cellulosic bioenergy crops could help alleviate the anthropogenic stresses to the biogeochemical cycling of carbon. One of the possible benefits researchers and policy makers are interested in is the potential for bioenergy feedstock cropping systems to be sinks for carbon and for the soil under these systems to act as a storage pool, sequestering carbon, essentially removing it from the terrestrial-atmospheric carbon cycle for the long term to help mitigate anthropogenic climate change. In order to make recommendations on best management practices and policy concerning carbon cycling in cellulosic bioenergy cropping systems, assessment of the carbon cycling in these ecosystems is necessary.

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Chapter 1

Literature Review

#### **Literature Review**

#### **Bioenergy Feedstocks, Soil, and Carbon Sequestration**

The carbon storage capacity of soil has been well-documented. The total soil carbon pool is approximately 2300 Pg, which is three times that of the atmospheric pool (~770 Pg; Lal, 2002). Soils play an important role in the global carbon cycle. The soil carbon pool is comprised of soil organic carbon (SOC) and soil inorganic carbon (SIC); there is approximately 1550 Pg and 750 Pg, respectively, stored in the top 1 meter of soil globally (Lal, 2002).

The Energy Independence and Security Act of 2007 mandates at least  $6.06 \times 10^6 L$  (16 billion gallons) of the  $1.36 \times 10^{11}$  billion liters of mandated renewable fuel be lignocellulosic biomass biofuels, which would require approximately 16.9 million ha (~10% of the US agricultural land). The majority of this land is expected to come from marginal land, too poor in quality to use for row crop production, and from Conservation Reserve Program (CRP) land (Boyer et al., 2013). Cellulosic bioenergy feedstocks are particularly suited for the southern US due to their longer growing season relative to corn. Also, corn yields in the region are lower compared to other regions in the US.

With cellulosic bioenergy, there is the opportunity to not only increase the United States' energy independency, but also create a closed system in which the  $CO_2$  production during combustion of fuel is at a net zero carbon input. This results from carbon being assimilated into the biomass during feedstock growth. A portion of the assimilated carbon is transferred to the pedosphere by processes such as root turnover, allowing the system used to grow the feedstock to act as a carbon sink. Factors such as, crop selection and management practices alter the quantity of C a particular soil is capable of storing. Cellulosic bioenergy feedstocks are crops well-suited to maximizing the soil's potential to store carbon (Dale et al., 2011). For example,

soil carbon accumulates under perennial grass cultivation, whereas it is often depleted under corn residue harvest (Anderson-Teixeira et al., 2009). It has also been reported that switchgrass (*Panicum virgatum*) and hybrid poplar (*Populus ssp.*) displaced the most fossil fuel compared to corn, soybean (*Glycine max*), alfalfa (*Medicago sativa*), and reed canarygrass (*Phalaris arundinacea*) derived bioenergies (Adler et al., 2007).

Soil carbon sequestration involves the capture of carbon from the atmosphere by plants and the long-term storage of the fixed carbon in the soil as soil organic matter (SOM) (Lal, 2004).The degree to which a biofuel crop acts as an agent for carbon sequestration is dependent on the soil environment. Factors such as soil quality, soil texture, soil moisture, soil temperature, and the C: nitrogen (N) ratio of the substrate all affect the magnitude to which a bioenergy cropping system can function as a carbon reservoir (Hartman et al., 2011). Cellulosic bioenergy crops continue to sequester carbon until equilibrium is met within the system, after which the system will act as a reservoir. (Hartman, et al., 2011). Biomass crops, as with conventional food crops, affect soil quality by changing organic matter, fluxes of nutrients, erosion (specifically during stand establishment), and soil compaction from planting and harvesting (Mann and Tolbert, 2000). These changes affect the activity of the soil microbial community and may alter its functioning.

#### Lower Mississippi Alluvial Valley (LMAV)

Vast areas of land that were once productive, but have become unproductive due to overuse or other degradative processes, are characterized by low SOC, soil quality, and biomass productivity (Lal et al., 1998). It has been proposed that this marginal land be utilized to grow bioenergy feedstocks. Oak Ridge National Laboratory defined marginal land as land that is

limited by erosiveness, excessive wetness, soil chemistry constraints, rooting constraints, or climate issues (Wright and Turhollow, 2010). This description is characteristic of many areas in the Lower Mississippi Alluvial Valley (LMAV). The LMAV has a history of intense agriculture. Alluvial floodplains, bottomland forests, and swamps were drained and converted into agricultural land dominated by cotton (*Gossypium spp.*) during the last century. Currently, the main crops grown in the LMAV are soybeans (*Glycine max*), rice (*Oryza sativa*), grain sorghum (*Sorghum bicolor*), and corn (*Zea mays*; NASS, 2013). Many areas of the LMAV are poorly suited for these agriculturally intensive crops and are susceptible to erosion, poor drainage during rainfall events, and drought during rain-limiting periods. A loss of SOM during land conversion and subsequent weathering events creates a soil carbon deficit (Lal et al., 1998).

The potential storage capacity for carbon in marginal soils is high due to their lack of SOM. Lal et al. (1998) estimated restorative efforts on degraded soils could increase C storage on those lands to 3 Pg C yr<sup>-1</sup>. When these soils are returned to more natural ecosystems with perennial vegetation and recommended management practices (RMPs) are used on agricultural soils, the storage capacity is greatly enhanced (Lal, 2004). Recommended management practices aim to simulate natural ecosystem functions on agricultural soils in order to retain or improve the conservation of resources and prolong agricultural sustainability. Many RMPs adopted by farmers, like reduced tillage and cover crops, are easily obtained by growing crops for cellulosic bioenergy.

The potential for marginal land to act as a carbon sink is enormous. Marginal lands are not at their maximal carbon storage capacity, so enrolling these lands in a production system that is environmentally viable serves to rehabilitate the land into a system that is economically desirable to producers. Nutrient cycling in bioenergy cropping systems can be extremely

efficient. Along with carbon sequestration, cellulosic bioenergy crops require reduced amounts of fertilizers and pesticides after establishment, which results in reduced nitrates, phosphorus, pesticides, and herbicide in surface run-off and groundwater (Mann and Tolbert, 2000). The combination of reduced input requirements and unique ecosystem attributes make bioenergy feedstocks ideal for the marginal lands of the LMAV.

#### Cottonwood

Cottonwood (*Populus deltoids*) was selected by the United States Department of Energy (US DOE) as a model energy crop for the US. Cottonwood is categorized as a short-rotation, woody crop (SRWC) (Kszos et al., 2001). Short rotation energy crops are fast growing woody crops capable of producing large amounts of biomass in a few short years. Along with cottonwood, poplar (*Populus* ssp), willow (*Salix* ssp.), sycamore (*Plantanus occidentalis*) and Eucalyptus have also been identified as potential species for bioenergy crops. The species selected as models under SRWC were chosen due to their wide range of adaptability to various environments and their disease tolerance (Lemus and Lal, 2005). Logistically, woody crops have several advantages over other bioenergy crops. Harvesting and transportation is similar to that of the pulp and paper industry. However, unlike seasonally-harvested bioenergy feedstocks, woody crops can be harvested as needed year round; making storage of harvested material, a big hurdle in the commercialization of other dedicated bioenergy feedstocks, a non-issue.

Eastern cottonwood is well-suited for the poor, marginal soils of the LMAV. Eastern cottonwood has been shown to have the fastest growth rate on LMAV soil of the SWRC identified by DOE, achieving growth rates of 1.5 to 2.0 m yr<sup>-1</sup> (Johnson et al., 2007). Data have demonstrated that converting traditional cropland to SRWC decreases surface runoff and

improves groundwater quality (Thornton et al., 1998). Eastern cottonwood has also shown to be tolerant to changes in available water. Below-ground carbon assimilation and osmotic adjustment due to water stress were greater in eastern cottonwood and its clones compared to black cottonwood and its clones (*Populus trichocarpa* L.) (Tschaplinski et al., 1993). It has been reported than cottonwood stand yields were consistent across various landscape positions, including flat, summit, depositional, West, South, Southwest, and North facing hillslopes at elevation from 311 to 318 meters at the University of Minnesota's Agricultural Ecology Farm on loamy, calcareous glacial till (Thelemann et al., 2010). These are crucial attributes when taking into account periods of saturated, as well as, periods of drought conditions common to the LMAV (Farmer, 1968).

Another significant aspect in the sequestration of carbon is the fine root dynamics of woody crops. The fine root biomass portion of these woody crops constitutes a potentially highly active carbon pool. While the fine root biomass may only be 1-15% of the total tree biomass, carbon is rapidly cycled through cottonwood fine roots (Kern et al., 2004). Fine root production can comprise 10 to 60 % of the total net primary production for the plant (Nadelhoffer and Raich, 1992). The life span of these fine roots can last from 20 to 200 days, indicating that the rapid turnover of this fine root pool can play a crucial role in the cycling of carbon (Essienstat and Yanai, 1997; Kern et al., 2004).

There are multiple factors that can affect the rate of fine root production (FRP). The largest factor affecting FRP is seasonality; FRP is associated with key phenological events during the growing season. Other factors like nutrient and water availability, temperature, and atmospheric  $CO_2$  concentrations also affect the rate of FRP (Kern et al., 2004). It has been shown that FRP decreased with increased soil water and nitrogen availability (Pregitzer et al., 1990;

Tingey et al., 2005). The lack of soil moisture may cause plants to invest an increased amount of carbon into FRP. In addition, it was also indicated that FRP increased with soil temperature (Fischer et al., 2007). Other studies have shown that overall root respiration may increase with increased water and nitrogen availability (Valentini et al., 2000; Burton et al., 2002).

Cottonwood was once a part of the bottomland forest in the LMAV before large conversions of forested area to cropland occurred in the 1960s and 1970s to meet the growing demand for soybean. In recent years, there has been a push to re-forest a portion of this historically converted cropland back to bottomland forest (Stanturf and Portwood, 1999). Planting of eastern cottonwood has been of intense interest as the first step in the reforestation process in the LMAV since cottonwood is native to the region (Gardiner et al., 2004).

Along with the benefits of afforestation, it has been speculated that growing cottonwood as a commercial product would also be economically advantageous to growers on the marginal land in the LMAV. Revenue from the timber, cost share programs, carbon credit programs, hunting leases, and cost savings from the re-propagation of cut stumps could provide economic incentive to convert marginal land to this agroforestry system (Standturf and Portwood, 1999).

#### Switchgrass

Switchgrass is a native North American prairie grass that is highly productive with vast amounts of potential above- and below ground biomass. Switchgrass is a clump-forming, warm season C4 grass that was a significant constituent of the North American Tallgrass Prairie, and has been used in recent years as a forage grass in the Midwest (Mclaughlin and Walsh, 1998).

Traditionally, when initially establishing switchgrass, germination with seeds is preferred, but switchgrass also can spread with rhizomes. Rhizomes vary in the extent to which

they spread. Some rhizomes are concentrated in groups or bunches, while very active rhizomes spread out and may be considered sod forming (Parrish and Fike, 2005). Stand establishment with rhizomes may not be economically feasible on large-scale production, however, when establishing by seed, fields are susceptible to weed infestation and emerged seedlings are unable to compete resulting in a possible crop failure. Thus, herbicides are necessary to reduce weed competition until a stand is established. Once a stand is established, routine applications of herbicides have been reported as unnecessary (Parrish and Fike, 2005).

Switchgrass was selected as the model herbaceous bioenergy crop by the US DOE in 1991 (Pimental and Patzek, 2005; Wright and Turnhollow, 2010). The US DOE determined that switchgrass was an ideal candidate due to its broad adaption; it can be grown in virtually all of North America. Switchgrass is native to the US east of the Rocky Mountains and south of latitude 51° (Parrish and Fike, 2005). Because of this broad adaptation, soil property effects on productivity are less than other grasses (Hartman et al., 2011). The designation of switchgrass as a model crop for bioenergy prompted a surge in research with the objectives of increasing yields, improving seed germination, increasing hardiness, analyzing fertilizer input requirements, and environmental ramifications of switchgrass crop production.

Switchgrass has several advantages over corn, for biofuel production. Switchgrass may have an economic edge over corn due to a longer growing season (Boyer et al., 2013). It has been reported that the belowground biomass of switchgrass is four to five times greater than that of corn (Hartman et al., 2011). In addition, compared to corn, switchgrass produces more root biomass (Frank et al., 2004). Carbon additions into the soil under switchgrass cultivation could be a great as 2.2 Mg C ha<sup>-1</sup> yr<sup>-1</sup> as a result, in part, of the magnitude of root biomass of switchgrass (Hartman et al., 2011). Grasslands, in general, contain high levels of SOC and have

the potential to sequester large amounts of carbon (Lee et al., 2007). These high levels result from low soil disturbance, more root biomass, and high quantities of residue return (Lal, 2002).

Switchgrass, as a dedicated bioenergy feedstock, requires little input. Little or no fertilizer, irrigation or pest management are needed to grow a successful switchgrass crop, and high productivity is expected across varied environments, including those which are water limited (Parrish and Fike, 2005; Pimental and Patzek, 2005; Sanderson and Adler, 2008). Switchgrass is well-known for its water use efficiency and heat tolerance, and is well-suited to arid environments (Parrish and Fike, 2005). Switchgrass has also been shown to be tolerant to somewhat saturated conditions (Parrish and Fike, 2005; Sanderson et al., 1996).

Switchgrass is perennial, while it may take several years to establish, many stands in the southeast have been productive for two decades. It would also be possible to have multiple cuttings in a growing season, similar to grasses used for livestock feed (Parrish and Fike, 2005). Although, in order to minimize fertilization needs, harvesting once in the fall after senescence ensures nutrient loss from harvest is minimal (Boyer et al., 2013). Switchgrass also provides ecosystem services including improving soil quality, preventing nutrient loss, and carbon sequestration (Pimental and Patzek, 2005). A significant contributing factor to some of the benefits listed above, specifically water-use efficiency and carbon storage, is the vast and deep root system of switchgrass (Frank et al., 2004). Water-use efficiency of switchgrass is 50% greater than cool season forage grasses as reported by Stout et al. (1998). Relative to traditional cropping systems, perennial grasses grown as bioenergy crops have been shown to reduce run-off and erosion and, therefore, reduce loss of nutrients and organic matter (Sanderson et al., 1996). It has been reported that levels of total soil nitrogen (TSN) and SOC had not declined in an unfertilized harvested grassland over a 50 year period in a Russian Chernozem (Mikhailova et

al., 2000; Mikhailova and Post, 2006). Similarly, TSN levels and biomass yields were maintained under unfertilized harvested perennial grass plots in the Continuous Hay Experiment at Rothamsted, United Kingdom over a 120 year period (Jenkinson et al., 2004; Jenkinson et al., 1994). A study on perennial grasses in Kansas reported that perennial grass fields maintained over 40 Mg ha<sup>-1</sup> more soil carbon and 4 Mg ha<sup>-1</sup> more nitrogen than traditional annual crops (Glover et al., 2010).

Another significant attribute studied with perennial grass bioenergy production is carbon sequestration and the associated accumulation of SOC. Switchgrass, as with any grass stand, can potentially be a large source of carbon. This is due to the extensive root systems and associated highly active microbial communities characteristic of grass stands (Hartman et al., 2011). Despite the high level of soil respiration in these systems, they are largely viewed as net carbon sinks (Hartman et al., 2011). A vast and deep root system allows switchgrass to accumulate greater SOC contents than cultivated cropland. Liebig et al. (2005) reported that total carbon contents for switchgrass were greater than cultivated cropland land in the 0-5 and 30-120 cm depths. Liebig et al. (2005) also reported that soil inorganic carbon (SIC) was greater in switchgrass than cultivated cropland in 0-120-cm depth, and SOC was greater in the switchgrass in 0-5 and 10-120 cm depths. The ability of switchgrass to sequester SOC at deeper depths (i.e. below 30 cm) is attributed to the vast root system of switchgrass that penetrates deep into the soil profile. The Soil Conservation Service reported that soil organic matter (SOM) accumulated at a rate of 1.1 Mg ha<sup>-1</sup> yr<sup>-1</sup> in the top 300 cm of midwestern soils during a 5 year study in which Conservation Resource Program (CRP) land was converted to perennial grass production. McLaughlin and Walsh (1998) stated that this conversion restored 23% of the soil carbon lost after decades of tillage prior to the study. It has been demonstrated that converting to fertilizedswitchgrass cultivation from a prairie system increased soil carbon storage and resulted in a negative net greenhouse gas (GHG) flux (Robertson et al., 2011). Fertilized and harvested switchgrass was shown to have increased SOC compared to non-fertilized, non-harvested switchgrass (Anderson-Teixeria et al., 2009).

However, switchgrass does have some disadvantages when grown for biofuels production. Stand establishment can be difficult; a stand could take two years before it is productive once it has been established (Sanderson and Adler, 2008). In addition, the cost of producing ethanol from switchgrass is estimated to be \$0.54, which is \$0.09 greater than for ethanol produced from corn (Pimental and Patzek, 2005). However, production costs continue to fall and biomass centers are close to beginning commercial production of switchgrass-derived ethanol. In addition, switchgrass, as with any crop grown in vast monocultures, can develop susceptibilities to disease and predation. It has been reported that switchgrass grown in monoculture shows some susceptibility to various strains of yellow barley dwarf virus. Switchgrass anthracnose is a result of the fungal species *Colletotrichum navitas* (Crouch et al., 2009). Anthracnose presents in switchgrass as elliptical foliar lesions with purple margins and white necrotic centers (Waxman and Bergstrom, 2011). One of the postulated benefits of largescale switchgrass production is increased wildlife habitat that is characteristic of tall grass prairie. However, switchgrass in monoculture would have no floral diversity, which could lead to a reduction in the faunal diversity (Lemaire et al., 2011). It is unclear whether avian populations would respond to a switchgrass monoculture similarly to a native tallgrass prairie. Lemaire et al. (2011) also postulated that soil erosion would increase and carbon storage decrease with switchgrass monoculture compared to a native prairie.

The two most popular commercially available varieties are 'Cave- in- Rock' and 'Alamo'. 'Cave-in-Rock' is a broadly adapted cultivar well-suited for the northeast, mid-Atlantic, and midwest US, while 'Alamo' is adapted for the southern portion of the country (Sanderson and Adler, 2008). While new varieties are being developed, switchgrass has received little attention from plant breeders, so developed cultivars are similar to native plants. This could mean that there is great opportunity for germplasm and yield improvements through more expansive breeding programs (Sanderson and Adler, 2008).

Yields for switchgrass in the United States are approximately 10 to 14 Mg ha<sup>-1</sup> of dry biomass (Wullschleger et al. 2010). Yields are affected by spatial variations in temperature and precipitation, and tend to decrease the further west and north in relation to the southern US (Berhman et al., 2013). Annual net aboveground biomass production in the southeast has been reported to be approximately 17-35 Mg ha<sup>-1</sup> (Liebig et al., 2005). The LMAV has been predicted to have the greatest yields (NRC, 2011). Lowland cultivars are the most commonly used for the southern US. Lowland cultivars include 'Alamo' and 'Kanlow' which have been reported to have yields greater than 28 Mg ha<sup>-1</sup> of dry biomass (Wullschleger et al., 2010).

Grasslands constitute 70% of agricultural land worldwide, but our understanding of their biogeochemistry is minimal (Lemaire et al. 2011). More research is needed to develop a clear understanding of the biogeochemistry of grasslands, and grasses grown in monoculture (i.e., switchgrass). The literature does not have a current understanding on the effects of monoculture switchgrass on faunal biodiversity, carbon sequestration, nutrient cycling, and pest and disease management. More research in these areas is needed before the benefits and limitations of large scale switchgrass for bioenergy production can be assessed.

### Soybean/Grain Sorghum Crop Rotation

Soybean is a common crop grown in the LMAV. In 2011, producers planted approximately 1.3 million hectares of soybean (NASS, 2013). Harvested soybeans are crushed for oil, protein-meal, or other valued-added products. The remaining uncrushed soy is shipped internationally. Planting depth is approximately 2.5 to 4 cm in silt loam soils. Plantings are generally preferred in April through mid-June, but soil moisture may be a limiting factor for nonirrigated soybeans planted in June. Soybean seeds will germinate between 3-43°C, but the optimum temperature for germination is approximately 35°C. Uniform stand establishment can be expected once the soil has reached 12.8°C. Non-irrigated yields are approximately 1.36 to 2.04 Mg ha<sup>-1</sup>. Seeding rates vary greatly between 24,000 and 97,000 seeds ha<sup>-1</sup>. Row spacing also varies from 18 to 97 cm, depending on variety and desired final plant population. Soybean yields approximately 0.136 Mg ha<sup>-1</sup> for every 2.5 cm of water during the growing season; therefore, 25 to 30 cm of water is needed to achieve adequate yields. Yield reductions due to soil acidity are expected when soil pH is below 5.8. Liming is a common practice for fields below the optimum soil pH range of 5.8 to 6.0. Molybdenum additions are recommended when soil pH is below 7.0. Phosphorus and potassium are other common additions. Soil tests on non-irrigated bottomland alluvial soils resulting in less than 25 kg P ha<sup>-1</sup> and 138 kg K ha<sup>-1</sup> indicate fertilization is needed. Nitrogen is not a common limiting nutrient as soybean and other legumes form symbiotic relationships with nitrogen fixing bacteria that are capable of supplying adequate amounts of nitrogen to the plant. However, if land has not been planted with legumes in the previous three to five years, then inoculation of seeds with Rhizobia bacteria before planting is necessary to ensure good nodulation of the roots (MP197, University of Arkansas Division of Agriculture, 2000).

Grain sorghum is a versatile crop grown in the US mainly for animal feed (UA-CES, 2012). Producers planted approximately 40,000 ha of grain sorghum in 2011 in Arkansas (NASS, 2013). Yields range from 0.8 to 0.9 Mg ha<sup>-1</sup>, but many farmers yielded 1.0 or more Mg ha<sup>-1</sup>. Yield data for sorghum in Arkansas go back to 1929. Grain sorghum is well-adapted to Arkansas soils, and grows best on well-drained, loamy soils. Planting should be as early in the spring as possible. Planting can occur after the soil reaches 18°C 5 cm below the soil surface. Under non-irrigated conditions, the recommended planting rate is approximately 120,000 seeds ha<sup>-1</sup> at a depth of approximately 4 cm. Row spacing varies widely from approximately 15 to 100 cm. Grain sorghum needs approximately 40 to 60 cm of water per growing season. The average non-irrigated sorghum yields are approximately 3.35 Mg ha<sup>-1</sup>. Grain sorghum grows best in a range of soil pH 6.0 to 7.5, and liming may be necessary below a soil pH of 5.7. Nitrogen is the most limiting nutrient for grain sorghum, and typical fertilization recommendations are approximately 37 kg N ha<sup>-1</sup> for non-irrigated double-cropped grain sorghum behind a small grain, such as wheat (*Triticum aestivum*; MP 297, UA-CES, 2012).

#### **Carbon Sequestration as an Ecosystem Service**

Carbon dioxide in the atmosphere is considered a greenhouse gas. Greenhouse gases provide insulation from the coldness of space; however, increased concentrations of GHG in the atmosphere trap re-radiated solar energy from the sun and prevent the solar energy from escaping the earth's atmosphere. The trapped energy increases, subsequently, atmospheric temperature and causes changes in climate. The emission of GHG from activities, such as, transportation and land use changes are responsible for anthropogenic climate change. Climatic forcing from increased levels of GHG such, as carbon dioxide, methane (CH<sub>4</sub>), and nitrous oxides (NO<sub>x</sub>), has increased the Earth's temperature and changed natural ecological cycles. These changes threaten our water, food, and energy security. Small changes in the Earth's temperature can have dramatic effects on our agricultural systems. Scientists and climatic modelers are intensely studying the effects that climate change will have on the earth.

The pedosphere interacts with the atmosphere, biosphere, hydrosphere, and lithosphere. These interactions influence the biogeochemical cycling of nutrients. The interactions of the pedosphere and atmosphere result in gaseous and energy exchanges between the atmosphere and soil, including the emissions of  $CO_2$  from soil into the atmosphere (Lal et al., 1998b). Current atmospheric levels of  $CO_2$  are approximately 398 parts per million (ppm), and have risen an average of approximately 2 ppm per year since 2000 (NOAA, 2013).

The burning of fossil fuels emits approximately 6 Pg C yr<sup>-1</sup> into the atmosphere (Lou and Zhou, 2006). Coal, crude oil, and natural gas are all relatively environmentally inert when encased in the Earth's crust. Once combusted, the carbon actively participates in the carbon cycle and can have increased environmental impact. One of the challenges of science is to develop technologies to not only reduce CO <sub>2</sub> emissions, but also remove what has already been emitted from the active carbon cycle. Bioenergy could potentially be a partial solution to both issues, not only supplying fuel that has a net zero carbon footprint, but also, through proper management, storing carbon in the soil removing it from the active carbon cycle.

The increase in soil carbon through soil carbon sequestration has two notable positive effects. First, there is an enhancement in soil quality, and second is the improvement of the soil's capacity to regulate the environment (Lal et al., 1998b). An increase in the organic carbon can lead to improvements in soil biodiversity, increased rooting depth of plants, improved soil structure, increased available water capacity, improvements in elemental and nutrient cycling,

and improved environmental regulation (Lal et al., 1998b). Progress in these areas also acts as feedbacks on SOC, increasing the amount or rate a soil can sequester. Soil is one of the five carbon pools described by Lal (2004). Enhancing the soil's ability to store carbon and increasing the length of time which that carbon resides in the soil through management of the soil resource is another important factor in the success of bioenergy cropping systems in mitigating climate change.

Quantities and rates of carbon sequestration, along with residence time in the soil have been studied in vast array in the last two decades. Soil carbon sequestration is dependent on individual ecosystem functioning. It has been suggested, based on models, approximately onethird of anthropogenic carbon emissions could be sequestered in plant and soil carbon pools (Schimel et al., 2001). Soil texture, climatic regime, vegetation, soil fauna and flora communities, time of year, and management impact the soil's ability to sequester carbon. For instance, soils high in clay generally have greater SOC than soils lower in clay due to the increased ability to form organo-mineral complexes, and therefore form more aggregates than coarser-textured soils (Lemus and Lal, 2005). On the other hand, coarser-textured soils promote more rapid decomposition of vegetation (Lemus and Lal, 2005). With regards to impacts from vegetation, it has been suggested by Lemus and Lal (2005) that perennial grasses are able to increase SOC due to an increase in SOM by stabilizing SOM and the biomass turnover of a dense root system. Short rotation woody crops (SWRC) like cottonwood maintain elevated levels of SOC by their characteristic high rates of litterfall constantly enriching SOM (Lemus and Lal, 2005).

Soil carbon sequestration involves three principle processes: humification, aggregation, and sedimentation (Lal et al., 1998). Understanding the soil processes underneath the bioenergy

crops is crucial to assessing the potential for bioenergy carbon sinks. Perennial cellulosic bioenergy crops promote aggregation and decrease erosion. Through a decrease or complete absence of tillage, cellulosic bioenergy crops decrease decomposition and volatilization stimulated by tillage (Lal et al., 1998b). The promotion of soil aggregation though the enrichment of the soil microbial community is one of the key processes underlying carbon sequestration in soils under cellulosic bioenergy crops. The avoidance of tillage not only decreases mineralization and degradation of soil aggregates by microbes, but also reduced or notillage systems help to prevent spikes in soil microbial respiration by keeping SOC protected from microbial decomposition through long-term stable aggregates (Lal et al., 1998b).

#### **Biogeochemical Cycling of Carbon in Soils**

There are five pools of global carbon that include the atmospheric pool (760 Pg), the oceanic pool (38,000 Pg), the geologic pool (5000 Pg), the soil pool (2500 Pg of SOC and SIC), and the biotic pool (560 Pg) (Lal, 2004). Carbon cycles within and throughout these pools, and the residence time in a given pool varies based on physical, chemical, and biological processes. Carbon sequestration varies depending on microbial biomass productivity, site history, management practices, and physical and biological properties (Lemus and Lal, 2005). Carbon cycles rapidly in tropical climates. Even with large, continuous carbon additions in the form of litterfall and root biomass cycling, SOC is low compared with the cool wet climates of the northern latitudes. While the additions of carbon are extremely low, the cold wet climate greatly slows the decomposition of detritus, which accumulates in the soil resulting in large quantities of carbon stored for the long term (Anderson-Teixeira et al., 2009). The soil pool is comprised of SOC and SIC. The soil pool differs greatly across regions, and there is a wide range of SOC

depending on ecosystem. For instance, estimated values for SOC range from 87 to 133 Mg C ha<sup>-1</sup> in temperate forests to 224 to 312 Mg C ha<sup>-1</sup> in boreal forests (Lal, 2004).

The fraction of soil that supplies nutrients for plant growth is soil organic matter (SOM). Soil organic matter is responsible for soil's cation exchange capacity, so it maintains soil fertility and soil structure. Soil organic matter can store carbon as SOC for hundreds or thousands of years before it is broken down and released during microbial respiration (Lou and Zhou, 2006). The SOM fraction is also where the SOC exists. Mineralization is the principle route of SOC loss (Lal, 2004). Soil organic carbon concentration generally decreases exponentially with depth, and its vertical distribution is affected by climate, soil texture, and vegetation type, with grasslands generally having greater SOC concentration than forested soils (Anderson-Teixeira et al., 2009).

Land-use changes can release large quantities of carbon as  $CO_2$  from the soil as mineralization of vegetation and humus increases (Lal, 2004). Terrestrial ecosystems were the largest sources of C until the 1970s when the combustion of fossil fuel became the number one source of C in the atmosphere (Lal et al., 1998). Land use changes, including land clearing, deforestation, and burning release approximately 1.2 Pg C yr<sup>-1</sup> into the atmosphere (Lou and Zhou, 2006). Approximately 55 to 78 Gt C have been released from the soil during the postindustrial period as a result of land-use changes (Anderson-Teixeira et al., 2009). The conversion of forested land to agricultural land is responsible for the greatest amount of  $CO_2$  emissions from the soil when considering land-use changes (Lal, 2004). The conversion of forest and grassland account for over one-third of the total land-use change, and a vast proportion of this conversion has happened since the early 1800s (Lal et al., 1998). The drainage of wetlands for agriculture or construction is also a large source of  $CO_2$  releases from the soil into the atmosphere. Perturbations caused from agriculture, such as plowing or burning of biomass, exacerbate soil

 $CO_2$  emissions (Lal, 2004). Land use history is an important factor in the carbon cycling of a particular land area (Lal, 2004). While land conversion from native to agricultural land can lead to large losses of carbon (~1.2 Pg C yr<sup>-1</sup>) the addition of SOC from the conversion from agricultural to perennial land use is comparatively slow (Post and Kwon, 2000). Post and Kwon (2000) reported that maximum sequestration of carbon from early conversion from agricultural land to perennial vegetation is less than 100 g C m<sup>-2</sup> yr<sup>-1</sup>.

#### **Soil Respiration**

The increased interest in climate change during the past few decades has necessitated increased research into soil respiration. The number of papers published on the topic of soil respiration has increased from 10 in 1985 to approximately 200 in 2004 (Luo and Zhou, 2006). The CO<sub>2</sub> emitted from all soil globally is recognized as one of the largest fluxes in the global carbon cycle. Estimates range from 68 to 77 x  $10^{15}$  g C yr<sup>-1</sup> (Schlesinger and Andrews, 2000). Soil respiration is significant to climate change research in two ways. The emission of CO<sub>2</sub> from the soil to the atmosphere is a greenhouse gas and acts as a part of the forcing behind climate change. Soil respiration is also a significant factor in soil carbon sequestration. The role of carbon sequestration is to reduce the amount of carbon in the atmosphere by storing it long term in the soil and soil respiration can have a negative impact on the process.

Soil respiration is the production of carbon dioxide by soil organisms and plant roots (Lou and Zhou, 2006). The carbon dioxide produced by the living biomass portion of the soil is a waste product from catabolizing organic matter in the production of energy. Net carbon dioxide efflux is strongly dependent upon environmental factors including solar radiation, temperature, and soil moisture availability (Lemaire et al. 2011). These factors can influence net carbon

dioxide efflux from the soil directly via altering physical gaseous diffusion from the soil into the atmosphere or indirectly by impacting respiration rates of plants and soil microbes. Not only do soil factors impact carbon dioxide emissions in the form of soil respiration, but carbon dioxide, as a gas or in solution, influences soil processes. It controls soil pH in the mildly-acidic to mildly-alkaline range, and it is also a key leaching agent as carbonic acid in solution (Lavelle and Spain, 2001).

#### **Mechanisms of Soil Respiration**

The production of carbon dioxide in the soil and its release into the atmosphere involves several processes, including the respiration of the living organisms and plant roots in the soil and the transport of the carbon dioxide to the atmosphere from the soil. Plant root respiration and microbial respiration are the main sources of carbon dioxide in the soil. Other soil fauna contribute a small portion of carbon dioxide to soil respiration, but their contribution has not been quantified in the literature. The breakdown of plant detritus and SOM by microbes results in the microbial fraction of soil respiration (Lou and Zhou, 2006).

The production of carbon dioxide by living tissues is the result of the common biochemical pathways including the tricarboxylic acid (TCA) cycle, glycolysis, pentose phosphate and electron transport pathways in aerobic respiration, and glucose fermentation in anaerobic conditions, which can occur through multiple pathways. At the biochemical level, respiration is regulated by energy needs of the cell and/or organism, substrate availability, temperature, and oxygen supply (Lou and Zhou, 2006).

Root respiration accounts for approximately 50% of total soil respiration, but can vary dramatically. Respiration from roots consumes 10-50% of the carbon assimilated through

photosynthesis each day, and therefore, measured soil respiration is strongly correlated with fineroot density (Hanson et al., 2000; Lambers et al., 1998; Lou and Zhou, 2006, Schlesinger and Andrews, 2000). The quantity of carbon dioxide respired by roots is determined by the root biomass and specific root respiration rates. Root biomass in an ecosystem depends on the individual ecosystem production and allocation patterns of specific plant species.

Plant physiology can have a dramatic effect on respiration rates. Plant and root longevity can impact respiration. In general, root respiration decreases with older roots (Lou and Zhou, 2006). Therefore, the fraction of soil respiration from roots (specific root respiration) is variable depending on plant species and overall ecosystem productivity. Specific root respiration is the respiration rate per unit of root biomass and reflects energy needs of processes occurring in the plant including production (biosynthesis) of new structural biomass, translocation of photosynthate, uptake of ions (nutrients) from soil, protein turnover, and cellular ion-gradient maintenance (Amthor, 2000). Environmental factors also influence the rate of root respiration. Flooding, salinity, water stress, nutrient supply, irradiance, and pH values affect root respiration rates (Lambers et al., 1998). Flooding decreases respiration, while increased salinity and water stress increases respiration as energy needs increase. An increase in temperature will result in an increase in root respiration due to the temperature sensitivity of the enzymes that catalyze the reactions required for respiration (Lou and Zhou, 2006).

There are two main categories of root respiration, maintenance respiration and growth respiration. Growth respiration results in energy and metabolic intermediates for the synthesis of structural compounds in the plant, while maintenance respiration produces the energy needed for the normal activities of the cells of the organism. In low nutrient environments, root respiration is lower than with plants grown with adequate nutrients. A portion of carbon that is fixed by

plants is transferred to root as exudates. This fraction can range from 10 to 70% of the total carbon fixed by a plant. Variations in this fraction are due to differences between species. In general, perennial plants transfer more carbon to roots than annual vegetation (Grayston et al., 1996).

#### **Carbon Dioxide Transport within the Soil**

Carbon dioxide concentration increases sharply with increasing soil depth (Lavelle and Spain, 2001). The concentration gradient along the vertical soil profile makes it possible for carbon dioxide to move through and out of the profile into the atmosphere via convection (mass flow) and diffusion. Mass flow occurs when a gradient exists in the air pressure between two points causing the air to move, while diffusion occurs when a concentration gradient exists with  $CO_2$  itself without air movement (Luo and Zhou, 2006). The concentration of carbon dioxide in the deep soil has been measured to be over 100 times that at the soil surface in a study from California (Lewicki et al., 2003). The majority of  $CO_2$  is produced in the upper portion of the soil profile, where the concentration of microbes and roots are the greatest, and where the gradient of  $CO_2$  in the soil is the steepest and, therefore, the place of the greatest movement of carbon dioxide through the soil and into the atmosphere. The greatest concentration of carbon dioxide in the soil is in the lower depths. (Lou and Zhou, 2006). Although the population of microbes and roots is considerably much lower after the first meter in the soil, due to the slow rate of diffusivity of the CO<sub>2</sub> whatever amount of CO<sub>2</sub> is produced by the small population of microbes and roots stays at that depth and the concentration builds up. The carbon dioxide gradient can vary depending on soil texture and porosity, rainfall and infiltration, and carbon dioxide production rate versus movement rate. Soil carbon dioxide concentration and the gradient within
the soil profile also exhibit seasonality, which causes changes in overall concentration and intensity of the gradient in the soil profile. These seasonal changes are largely controlled by changing rates of production from microbes and roots due to environmental controlling factors (Luo and Zhou, 2006). The soil's water content is an important factor to consider when discussing the diffusion of carbon dioxide in the soil. The diffusion coefficient of  $CO_2$  in water is about 10,000 times lower than that in the air phase,  $1.6 \times 10^{-9}$  and  $1.6 \times 10^{-5}$ , respectively (Luo and Zhou, 2006). Thus, the water-filled porosity can greatly affect soil carbon dioxide flux measurements.

## Soil- to-Atmosphere Carbon Dioxide Transport

Similar to water evaporation at the soil surface, carbon dioxide release from the soil to the atmosphere is strongly influenced by wind gusts and turbulence. Changes in barometric pressure can account for up to 60% of the variations in the diffusion rate of gases in deep soil layers (Kimball, 1983). A 25% increase in gas fluxes has been reported in silt-loam soils with low porosity (Kimball and Lemon, 1971).

Fluctuations of soil surface temperature and wind velocity may be strong controlling factors in the diurnal soil CO<sub>2</sub> efflux. At night cooler temperatures reduce wind turbulence and decrease CO<sub>2</sub> efflux from the soil. While during the day, increases in soil surface temperature and wind velocity not only increase soil CO<sub>2</sub> efflux, but increases in soil surface temperature increase respiratory activity in the soil (Luo and Zhou, 2006). A litter layer decreases soil surface CO<sub>2</sub> flux due to increased resistance of diffusion of the gas. Maier and Kress (2000) measured the CO<sub>2</sub> concentration at 15 cm of a mineral soil at 950  $\pm$  200 µmol mol<sup>-1</sup> in unfertilized plots with a thin litter layer and 1250  $\pm$  220 µmol mol<sup>-1</sup> in fertilized plots with a thick litter layer in a

loblolly pine forest in North Carolina. Soil texture also affects  $CO_2$  movement in soils, mainly due to soil texture's effects on porosity and water-holding capacity (Bouma and Bryla, 2000) Bouma and Bryla (2000) reported that finer textured (1:4 v/v Candler fine sandy soil and Hagerstown silty-clay, respectively) soil retained more water compared to a sandier soil (1:1 v/v Candler fine sandy soil and Hagerstown silty-clay, respectively) during an irrigation event, and subsequent soil respiration values were underestimated. The underestimation of  $CO_2$  values persisted for a longer time period for the finer textured soil. These differences result from the reduced diffusivity of  $CO_2$  in the finer textured soil caused by the increased soil water content (Bouma and Bryla, 2000).

### **Regulating Factors of Soil Respiration**

Soil carbon dioxide is a byproduct of the biochemical process of respiration, but is often studied on a spatial scale. Scientists are most concerned with the implications of this biological process on atmospheric CO<sub>2</sub> concentration and climate change. From the microscopic to global scales, soil respiration comprises different sets of biological, chemical, and physical processes that control CO<sub>2</sub> movement within the hierarchy of scales (Luo and Zhou, 2006). The processes involved in soil respiration are controlled by physical (i.e., soil moisture, temperature, porosity) and biological factors (i.e., root density, microbial community, rate of photosynthesis rate, and substrate availability; Vargas and Allen, 2008, and Berg et al., 1982). Substrate availability and CO<sub>2</sub> are linearly related, while the rates at which various substrates are converted to CO<sub>2</sub> differ with type of substrate (Berg et al., 1982). Simple sugars are the easiest for both microbes and roots to use. Humic acids are the most difficult to breakdown and have the longest residence time in the soil. Cellulose, hemicellulose, and lignin are intermediate with regards to

decomposition (Lou and Zhou, 2006). Therefore, vegetation type can have a dramatic effect on substrate availability. Overall, the heterogeneity of typical ecosystems makes deriving simple substrate and soil respiration relationships difficult. Studies have reported that removal of substrate supply from the photosynthesizing canopy can decrease soil respiration 50% within two months in a Scots pine (*Pinus sylvestris*) forest in Sweden (Högberg et al., 2001). It has also been reported that clipping and shading in a grassland, located in the U.S. Great Plains, decreased soil respiration by 70% in one week (Craine et al., 1999). There is a close relationship between seasonal fluctuations of aboveground photosynthesis and soil respiration, but it is difficult to measure directly. Often measurements like leaf area index serve as a proxy for above ground plant production (Lou and Zhou, 2006).

### Soil Temperature and Soil Moisture as Regulating Factors of Soil Respiration

All facets of soil respiration are affected by temperature to some level. The effects of temperature are most notable in the biochemical production of carbon dioxide by roots and soil microbes. Generally, respiration increases with increases in temperature until around a peak temperature, approximately 45 to 50°C for most soil organisms and approximately 35°C for roots, then decreases (Luo and Zhou, 2006). The limiting factor for soil respiration in the low temperature range is the maximum activity at a particular temperature for enzymes involved in respiration (Atkin and Tjoelker, 2003). While, in the high temperature range, adenylates and substrate supply play a greater role in controlling respiration (Douce and Neuburger, 1989; Atkin et al., 2002; Svensson et al., 2002; Atkin and Tjoelker, 2003). At high temperatures, just below 35°C, transport of substrates and products via diffusion becomes a limiting factor in root respiration (Kasper and Bland, 1992). The maximum rate of soil microbial respiration can vary

within a soil temperature range from 20 to 40°C depending on the physiological characteristics of soil microbes adapted to a particular area; microbes adapted to warmer conditions like in the LMAV would be expected to achieve maximum rates of respiration at higher soil temperatures (Fang and Moncrieff, 2001). Maximum root respiration varies depending on plant phenology. For instance, it was reported that the greatest increase in root respiration for both soybean and sorghum was during the transition from vegetative to flowering stages and then root respiration declines thereafter (Curiel Yuste et al., 2004).

### Soil Microbiology

The soil microbial community is complex. Microorganisms are incredibly diverse and enormously abundant in the soil, and include algae, bacteria, cyanobacteria, fungi, yeasts, myxomycetes, and actinomycetes (Kimble et al., 2003). A single gram of soil may contain 10,000 species of microorganisms (Lemaire et al., 2011). Understanding the role of soil microorganisms in the cycling of carbon and other nutrients in the soil is crucial to assessing the potential of cropland and agroforests to sequester carbon and mitigate climate change (Doran et al., 1994). Soil microorganisms serve to mediate the decomposition of organic material in soil, and therefore, fill a vital role in the carbon cycle. The microbial communities present in soil form the foundation of the soil food web, and function in nearly all biogeochemical transformations (Culman et al., 2010). It has been reported that microbes are responsible for maintaining native perennial grass soil fertility compared to adjacent high-input cropping systems in Kansas (Culman et al., 2010).

Microbes are responsible for the fraction of soil respiration not accounted for by root respiration. Microbial decomposition of litter and soil organic matter releases carbon dioxide,

while mineralizing or immobilizing nutrients. High proliferation/productivity of microbes increases soil respiration, decreasing SOC. Soil microbial respiration can account for up to 80% of total soil respiration in grasslands (Raich and Schlesinger, 1992). In forests, respiration from microbes can account for approximately 50% of total respiration (Edwards and Sollins, 1973). Soil microbes are capable of breaking down plant residues with a C:N ratio of 100 to a ratio around 10:1 (Lemaire et al., 2011). Soil microbes are in greatest concentrations in the rhizosphere, and the microbial communities located in the rhizosphere differ dramatically from the microbes in the bulk soil. Bacteria are responsible for the majority of the decomposition of root exudates in the rhizosphere. Generally, three genera of bacteria are the most common in bacteria in the rhizosphere; *Pseudomonas, Achromobacter*, and *Agrobacterium*. The release of carbon dioxide from microbes in the rhizosphere is stimulated by the addition of liable carbon from exudates, mucilage, and dead cells from plant roots. Most of the root exudates (64 to 86%) are consumed quickly by microorganisms (Hütsch et al., 2002).

Litter decomposition by microbes contributes significantly to the proportion of soil microbial respiration (Lou and Zhou, 2006). This respiration occurs primarily at the soil surface and removal of litter has been shown to reduce annual soil respiration by 15% in grasslands and 27% in a lemon (*Critus lemonia*) orchard (Wang et al., 1999). Rate of litter accumulation and composition can vary greatly depending on mean annual temperature and rate of litter fall. For instance, in tropical forests, the rate of litter fall is among the largest globally, but overall litter accumulation is low due to the high mean annual temperature, which promotes rapid decomposition. In contrast, boreal forests have large litter accumulation even though the rate of litter fall in boreal forests is low. Significantly lower mean annual temperatures inhibits rapid breakdown of litter, promoting litter accumulation of the forest floor (Schlesinger, 1997).

Litter decomposition and, in turn, soil respiration are regulated by climatic factors, litter quality, vegetation and litter types. Climatic factors include mean annual temperature and precipitation and annual evapotranspiration (Lou and Zhou, 2006). Temperature and moisture are the most important climatic factors. Differences in litter composition can also have an effect on decomposition. Litter with a large percentage of cellulose, hemicellulose, and lignin, such as grasses and woody vegetation, slow decomposition rates. Decomposition of woody detritus can take a relatively longer period of time compared to non-woody detritus decomposition. The soluble fraction of litter is the most readily decomposed by microbes. The soluble fraction consists of amino and organic acids and sugars. Bacteria can easily digest these compounds during litter decomposition (Lemaire et al., 2011). Litter decomposition consists of three processes: leaching, fragmentation, and chemical alteration of organic compounds in which the waste product, carbon dioxide, is respired. In addition, decomposition of detritus by microbes in a traditional cropping system can be enhanced by the physical act of plowing. Plowing can "wake-up" dormant microbes in the soil with an influx of oxygen and nutrients, which can significantly increase soil respiration for a period of time after plowing (Lemaire et al., 2011). In regards to soil structure, soil biodiversity impacts soil structure positively, with different organisms promoting soil aggregation through the production of organic polymers (Lal, 2004).

## **Similar Studies**

Measurements of soil respiration have been conducted for decades, but, until recently, very few studies have combined soil respiration from agricultural and ecological perspectives (Lou and Zhou, 2006). Studies conducted in the northern hemisphere in deciduous broadleaf forests (DBF) and grasslands indicate greater variability in peak CO<sub>2</sub> efflux in grasslands (GRS)

relative to DBF. Peak CO<sub>2</sub> efflux from the soil ranged from 3.8 to 5.6  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> for DBF, while CO<sub>2</sub> efflux for GRS ranged from 1.7 to 37.5  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>. Soybean-wheat rotations on a silt-loam in southeast Arkansas have been reported to have CO<sub>2</sub> effluxes between 10.0 and 13.0  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> (Bowden et al., 2000; Bremer et al., 1998; Brye et al., 2002; Davidson et al., 1998; Frank et al., 2004; Hibbard et al., 2005; Lee et al, 2007). Peak CO<sub>2</sub> efflux was reported during the months of May, June, July, and August with the majority of peak efflux being attained during July for those studies reporting peak flux timing.

Changes in microbial biomass are important indicators of ecosystem functioning and nutrient and energy cycling in the soil. Reductions in microbial biomass generally indicate negative impacts on ecosystem processes associated with management practices (DuPont et al., 2010). A microbial biomass study using the fumigation-extraction method has shown that additions of nitrogen fertilizer to a poplar (*Populus*) stand resulted in increased microbial activity; however, no changes in carbon sequestration were observed (Moscatelli et al., 2005). This study suggested that conservative N fertilization to SWRC would have little effect on the environmental goal of carbon sequestration (Moscatelli et al., 2005). In another study on microbial biomass in grass species grown for bioenergy, including switchgrass, showed enhanced carbon and nitrogen mineralization in monoculture perennial grass compared to monoculture corn. Switchgrass had a greater SOC content compared to corn in the upper 10 cm of the soil (Haney et al., 2010). Soil microbial biomass was shown to be greater in soil in the top 40 cm where perennial grass was plant compared to no-tillage rotations of soybean, grain sorghum, and wheat (DuPont et al., 2010).

### Justification

Although research of dedicated bioenergy feedstocks spans the last three decades, there is much that is still unknown about these unique cropping systems. The soil carbon pool is the least understood pool, much more is known about carbon cycling in the atmospheric and, arguably, the oceanic pools. Because of spatial variation, differences in vegetation, climatic regime, and soil physical properties results from studies on soil carbon cycling cannot easily be applied in blanket claims on how other, different systems will respond. For this reason, it is necessary to study how switchgrass and cottonwood, grown as dedicated bioenergy feedstocks, impact soil respiration and microbial biomass when grown in the LMAV. There have been no studies that have focused on the impacts these crops have on soil respiration and microbial biomass when grown in the LMAV. In order to move forward with commercial production of these feedstocks, there must be an increase in knowledge of how the climatic regime, unique soil physical properties, and vegetation impact carbon cycling in this area. It is likely that carbon credits will be given to producers who decide to move to bioenergy cropping systems; consequently, it is pertinent that soil respiration, as a major component of the soil-atmosphere carbon cycle is taken into account when assessing soil carbon storage capacity of soils in the LMAV under switchgrass and cottonwood as dedicated bioenergy cropping systems.

## **Objectives and Hypotheses**

The objectives of this study are to evaluate the effects of land use [switchgrass, cottonwood, and soybean/grain sorghum crop rotation] on monthly soil respiration and microbial biomass in a silt loam in the LMAV of east-central Arkansas. In addition to the evaluating monthly land-use effects, annual soil surface  $CO_2$  emissions from each treatment will be quantified and potential correlations between soil surface  $CO_2$  flux and soil temperature and

moisture will be identified. The goal of these objectives is to identify the effects that these two crops (i.e. switchgrass and eastern cottonwood) grown as dedicated bioenergy feedstocks have on  $CO_2$  emissions when grown on marginal land in the LMAV. A sub-objective of this study is to evaluate the effects of collar placement (in-row and between-row) on monthly soil respiration measurements in the agroecosystem.

It is hypothesized that annual CO<sub>2</sub> emissions from switchgrass and eastern cottonwood will be lower than from a soybean-grain sorghum rotation, while fluctuations in monthly soil respiration will be greatest in the soybean-grain sorghum rotation than in the other two ecosystems. Consequently, it is also hypothesized that soil microbial biomass carbon and nitrogen will be greatest in the soybean-grain sorghum rotation due to the influx of oxygen and nutrients resulting from tillage (Lemaire et al., 2011). With regards to monthly CO<sub>2</sub> flux, it is hypothesized that the greatest increase in flux in each treatment will be during the spring months (March, April, and May) with a plateau during the summer (June, July, and August) months and a decline in the fall (September, October, and November) followed by a plateau during the winter (December, January, and February), following the same general trends as reported in the literature. Similar to other studies on soil respiration correlations with soil temperature and moisture, it is hypothesized that soil temperature will be positively correlated and soil moisture will be negatively correlated to CO<sub>2</sub> flux after a maximum moisture requirement is reached, approximately 60% water-filled pore space, and these trends will be observed in all treatments (Luo and Zhou, 2006). It is also hypothesized that collar placement in the agroecosystem will effect monthly CO<sub>2</sub> flux measurements, with collars placed between rows having lower monthly CO<sub>2</sub> fluxes than collars placed in the rows.

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**Chapter II** 

Soil Carbon Dioxide Emissions from Switchgrass and Cottonwood Grown as Bioenergy in in the Lower Mississippi Alluvial Valley

### Abstract

Marginal land of the Lower Mississippi Alluvial Valley (LMAV) has the potential to be utilized for the production of bioenergy feedstocks. Soil respiration is the gaseous emission of carbon dioxide  $(CO_2)$  from microbes and plant roots in the soil, and these emissions play an important role in the global cycling of carbon. Soil respiration can act as a positive feedback affecting climate change, and has been shown to vary depending on soil moisture, temperature, and vegetation. The objectives of this study where to evaluate the effects of land use [switchgrass (Panicum virgatum), cottonwood (Populus deltoides), and a soybean (Glycine max)-grain sorghum (Sorghum bicolor) agroecosystem] on monthly soil respiration and estimated annual CO<sub>2</sub> emissions on a silt loam in east-central Arkansas throughout 2012 and 2013. Annual CO<sub>2</sub> emissions were calculated by linear interpolation between monthly measurements. Soil respiration from all three ecosystems followed the same general trend: increasing from January to May and decreasing from September to December, peak fluxes differed significantly (p < p0.05) among ecosystems for both years. Peak fluxes in 2012 were achieved for all three ecosystems in July. Soybean and switchgrass peak fluxes did not differ (8.08 and 7.59  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> respectively) and cottonwood peak flux differed from the other treatments (6.09  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>; p < 0.01). Peak fluxes for 2013 were achieved in May for both switchgrass and cottonwood (5.91) and 4.11  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, respectively), where the switchgrass peak flux was larger than that for cottonwood and the agroecosystem, which did not differ (p < 0.01). Annual CO<sub>2</sub> emissions differed among ecosystems (p < 0.001) but not between years (p = 0.45). Cottonwood had less CO<sub>2</sub> emitted for both years (7.3 and 7.4 Mg ha<sup>-1</sup> for 2012 and 2013, respectively) compared to the other two ecosystems, while emissions from the switchgrass did not differ from soybean in 2012 (10.3 and 9.5 Mg ha<sup>-1</sup>, respectively) or grain sorghum in 2013 (9.7 and 9.2 Mg ha<sup>-1</sup>,

respectively). Results showed established bioenergy feedstock cropping systems do not have greater soil respiration rates compared with a traditional soybean-grain sorghum crop rotation. Results also indicated that different bioenergy feedstocks can produce different quantities of  $CO_2$ emissions. Both factors are important to consider when enrolling marginal land in the LMAV in bioenergy feedstock cropping systems.

## Introduction

The Energy Independence and Security Act of 2007 mandates at least  $6.06 \times 10^{6} L$  (16) billion gallons) of the  $1.36 \times 10^{11}$  L biofuels produced be derived from lignocellulosic biomass by 2022, which would require approximately 16.9 million ha (~10% of the US agricultural land). The majority of this land is expected to come from marginal land, too poor in quality to use for row crop production, and from Conservation Reserve Program (CRP) land (Boyer et al., 2013). Cellulosic bioenergy feedstocks are particularly suited for the southern US due to their longer growing season relative to corn. Biofuel cropping systems have been touted as having the potential to sequester carbon from the atmosphere into the soil ecosystem and aid in the mitigation of climate change, while providing a fuel source that does not add additional carbon to the atmosphere, unlike conventional fossil fuel. The US Department of Energy (DOE) has selected numerous grasses, woody plant species, and more traditional crops as potential biofuel sources. For the purposes of this study, switchgrass and cottonwood were selected because of their broad adaption, tolerance to changes in available water, potential ecosystem services, and their abilities to have harvestable biomass while requiring relatively low amounts of input. Switchgrass, especially, has been shown to have high productivity across varied environments and is heat tolerant (Parrish and Fike, 2005). Cottonwood, particularly Eastern cottonwood, has been shown to have the fastest growth rate of all woody crops identified by the DOE as potential bioenergy crops on soil in the Lower Mississippi Alluvial Valley (LMAV), 1.5 to 2.0 meters per year (Johnson et al., 2007).

One ecosystem service which recent studies have focused on is quantity of  $CO_2$  flux as soil respiration from these biofuel cropping systems. Soil respiration is the production of carbon dioxide by soil organisms and plant roots (Lou and Zhou, 2006). Soil respiration is considered

one of the largest fluxes of carbon in the carbon cycle, representing 6.8 to 7.7 x  $10^{16}$  g C yr<sup>-1</sup> (Schlesinger and Andrews, 2000). Root respiration accounts for roughly 50% of total soil respiration; however this fraction can vary widely depending on vegetation type and age, soil type, and climatic regime (Lou and Zhou, 2006). Environmental factors such as soil temperature and soil moisture are strong regulating factors in the production of CO<sub>2</sub> by soil respiration processes and the diffusion of the gas out of the soil profile and into the atmosphere (Lou and Zhou, 2006). Other factors associated with vegetation type and management such as tillage and fertilization can have dramatic effects on CO<sub>2</sub> flux (Lemaire et al., 2011).

Although research of dedicated bioenergy feedstocks spans the last three decades, there is much that is still unknown about these unique cropping systems. The soil carbon pool is the least understood pool, much more is known about carbon cycling in the atmospheric and, arguably, the oceanic pools. Because of spatial variation, differences in vegetation, climatic regime, and soil physical properties, results from studies on soil carbon cycling cannot easily be applied in blanket claims on how other, different systems will respond. For this reason, it is necessary to study how switchgrass and cottonwood, grown as dedicated bioenergy feedstocks, impact soil respiration and microbial biomass when grown in the LMAV. There have been no studies that have focused on the impacts these crops have on soil respiration and microbial biomass when grown in the LMAV. In order to move forward with commercial production of these feedstocks, there must be an increase in knowledge of how the climatic regime, unique soil physical properties, and vegetation impact carbon cycling in this area.

The objectives of this study are to evaluate the effects of land use [switchgrass, cottonwood, and soybean/grain sorghum crop rotation] on monthly soil respiration and microbial biomass in a silt loam in the LMAV of east-central Arkansas. In addition to the evaluating

monthly land-use effects, annual soil surface  $CO_2$  emissions from each treatment will be quantified and potential correlations between soil surface CO<sub>2</sub> flux and soil temperature and moisture will be identified. The goal of these objectives is to identify the effects that these two crops (i.e. switchgrass and eastern cottonwood) grown as dedicated bioenergy feedstocks have on CO<sub>2</sub> emissions when grown on marginal land in the LMAV. It is hypothesized that annual CO<sub>2</sub> emissions from switchgrass and eastern cottonwood will be lower than from a soybeangrain sorghum rotation, while fluctuations in monthly soil respiration will be greater in the soybean-grain sorghum rotation than in the other two ecosystems. With regards to monthly  $CO_2$ flux, it is hypothesized that the greatest increase in flux in each treatment will be during the spring months (March, April, and May) with a plateau during the summer (June, July, and August) months and a decline in the fall (September, October, and November) followed by a plateau during the winter (December, January, and February), following the same general trends as reported in the literature. Similar to other studies on soil respiration correlations with soil temperature and moisture, it is hypothesized that soil temperature will be positively correlated and soil moisture will be negatively correlated to CO<sub>2</sub> flux after a maximum moisture requirement is reached, approximately 60% water-filled pore space, and these trends will be observed in all treatments (Luo and Zhou, 2006).

#### **Materials and Methods**

#### **Site Description**

This study was conducted at the University of Arkansas Division of Agriculture's Pine Tree Research Station (PTRS) (35°8'33.12"N, 90°44'24'66"W), near Colt, AR. The PTRS is located in the LMAV in St. Francis County in east-central Arkansas. The study site is poorly drained land that was previously used for row-crop production at the PTRS. The approximately 12-ha study area is surrounded on the north, south, and west by forest and by other cropping systems to the east (Fig.1). St. Francis County is located in the Southern Mississippi Valley Loess which is Major Land Resource Area (MLRA) 134 (NRCS, 2006). The study area is comprised of Calloway silt loam (~ 45%), Henry silt loam (~30%), and Loring silt loam (~17%) (NRCS, 2012). The Calloway silt loam is classified as fine-silty, mixed, active, thermic Glossaquic Fragiudalf that is somewhat poorly drained with 30 to 60 cm to the water table and contains a fragipan at 40 to 60 cm (NRCS, 2012). The Henry silt loam is classified as coarse-silty, mixed, active, thermic Typic Fragiaqualf that is poorly drained with the water table at 0 to 30 cm and a fragipan at 35 to 56 cm (NRCS, 2012). The Loring silt loam is classified as fine-silty, mixed, active, thermic Typic Fragiudalf that is moderately well-drained with the water table at 46 to 76 cm and a fragipan at 60 to 81 cm (NRCS, 2012).

The climate of the region is warm and wet with a 30-yr mean annual temperature minimum of -11.9°C in January and a 30-yr mean annual maximum of 37.6°C in August (NOAA, 2013). The 30-yr mean annual precipitation is 127 cm (NOAA, 2013).

#### **Field Treatments and Establishment**

Eastern cottonwood and switchgrass were selected as bioenergy feedstock for this study. A soybean/grain sorghum rotation was selected as a control treatment to represent the common upland row-crop rotation in the region. A set of three plots was used for each treatment. Two plots measured 30 m x 90 m, and the third measured 90 m x 90 m. A 17 m x 45 m subplot area was the measurement area within each plot (Fig. 1). The 90 m x 90 m plots served to provide

more accurate yield data, as the primary goal of this study was a complete economic and environmental assessment of switchgrass and cottonwood as bioenergy crops in the LMAV.

Cottonwood establishment occurred in February 2009 using 40-cm cuttings of three clones (ST-66, S7C20, and a mix of clones from a Louisiana Department of Agriculture and Forestry nursery). Cuttings were planted at a density of 4,495 cuttings ha<sup>-1</sup> after mid-winter site preparation consisting of sub-soiling and application of a pre-emergent herbicide. Cuttings were planted in spring 2009.

Switchgrass establishment occurred between April and May 2009. Switchgrass was drilled into the soil at a rate of 11.2 kg ha<sup>-1</sup>. Switchgrass has been harvested, baled, and removed annually since October 2010. A commercial forage-grass cutter was used to harvest the switchgrass to cure to less than 20% moisture content (wet basis) for baling.

Soybeans were planted and harvested in 2009. Grain sorghum was planted and harvested in 2010. Soybeans were then planted the following two years (2011 and 2012). Grain sorghum was cultivated during the 2013 growing season. Typical agricultural equipment is utilized during harvest of both crops. No irrigation was used in any treatment and fertilizer was only used where needed in the switchgrass treatment during establishment of a stand.

### Soil Surface CO<sub>2</sub> Flux

The procedures used in this study to measure soil surface  $CO_2$  flux was similar to that of Brye et al. (2002), Brye et al. (2006), and Brye and Riley (2009). Five, 10.2-cm diameter x 7.6cm tall polyvinyl chloride (PVC) collars were installed in each plot of all three ecosystem treatments. Collar placement in the soybean-grain sorghum crop rotation was within the 17 m x 45 m subplot located in the center of each plot alternating with between-row and in-row

placements as collars were moved periodically. In the switchgrass and cottonwood treatments, collar placement was at least three meters inside the outermost perimeter to eliminate edge effects. The PVC collars were inserted ~2 cm into the soil. Measurements were made between 1000 and 1600 hours on a single day once a month from January 2012 through December 2013. Collars were moved after two consecutive monthly measurements were recorded. Collar placement was random within each switchgrass and cottonwood plot or subplot in the soybean-grain sorghum crop rotation.

Soil surface CO<sub>2</sub> flux was measured using a LI-6400 CO<sub>2</sub> analyzer with a LI-6400-09 soil respiration chamber (Li-Cor, Inc., Lincoln, NE). The ambient CO<sub>2</sub> concentration in the atmosphere was measured and set as the target  $CO_2$  concentration. The instrument was set to measure the  $CO_2$  flux from the soil after being placed on top of the collar. It was necessary to remeasure and re-set the ambient  $CO_2$  concentration as the day progressed in order to account for any changes in the ambient CO<sub>2</sub> concentration. Before making each measurement, any green photosynthetically active plant material in the collar was removed. The soil respiration chamber was placed on to the collar and the measurement sequence was initiated, where the first step is the circulation of the chamber's headspace gas through soda lime. Soda lime is a mixture of compounds primarily consisting of calcium hydroxide  $[Ca(OH)_2]$ , water (H<sub>2</sub>O), sodium hydroxide (NaOH), and potassium hydroxide (KOH). Soda lime removes carbon dioxide from the air or water due to the reaction of calcium hydroxide with carbon dioxide to form calcium carbonate ( $CaCO_3$ ). This process continues until the concentration of carbon dioxide in the chamber's headspace falls below the target ambient concentration by approximately 40 mg L<sup>-1</sup>. After the chamber has been scrubbed down, the CO<sub>2</sub> concentration in the enclosed headspace is allowed to naturally build back up. At 10 mg  $L^{-1}$  below the target concentration, the flux

measurement will begin until the CO<sub>2</sub> concentration has increased to 10 mg L<sup>-1</sup> above the target concentration. The measured CO<sub>2</sub> flux was recorded in micromoles of CO<sub>2</sub> per square meter per second ( $\mu$ mol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>). Estimates of annual CO<sub>2</sub> emissions were determined by taking point data from the device for each measurement date and extrapolating CO<sub>2</sub> flux quantities for each day of the year. The extrapolated numbers were then summed and reported on a Mg-C ha<sup>-1</sup> per year for each plot.

Along with soil respiration measurements, 2- and 10-cm soil temperatures were measured next to each collar using probe thermometers. A Theta Probe (model TH2O, Dynamax, Inc., Houston, TX) was also used to measure the volumetric soil water content in the top 6 cm next to each collar.

### Soil Sampling, Processing, and Analysis

Soil core samples were collected in the spring of 2012 and 2013. To assess soil physical and chemical properties three, 0-10 cm and 10-20 cm soil samples were taken in each plot using a 4.7-cm diameter soil core chamber attached to a slide hammer. Samples were dried in an oven at 70°C for 48 hours and weighed to obtain a dry-weight in order to calculate bulk density (g cm<sup>-3</sup>) by dividing the mass of the dry soil by the volume of the sample. To determine particle-size distribution, a modified method of Gee and Or (2002) was used. Samples were ground using a mechanical grinder and a 50 g sub-sample, from the 2012 samples, was weighed and mixed with 50 mL of 100 g L<sup>-1</sup> sodium hexametaphosphate ((NaPO<sub>3</sub>)<sub>6</sub>). The mixture was quantitatively transferred into a 1-L sedimentation cylinder and brought to volume with tap water. The cylinder was then allowed to come to room temperature (20-25°C). The contents were mixed uniformly by hand plunging the cylinder. A hydrometer with a Bouyoucos scale (g L<sup>-1</sup>) measured the

density of each sample at 40 sec, 6 hr, and 11 hr after plunging. The 40-sec reading was conducted three times. A blank hydrometer reading was recorded in a 1-L cylinder with 50 mL of sodium hexametaphosphate brought to volume with tap water. The solution temperature was also measured in the blank.

Chemical analyses were performed on samples from 2012 and 2013 for both depths. Soil OM was determined by weight-loss-on-ignition. Electrical conductivity (EC) and soil pH were determined using an electrode and a 1:2 soil-to-water solution. Soil was extracted with a Mehlich-3 extractant solution in a 1:10 soil-to-extractant ratio (Tucker, 1992) and analyzed for extractable nutrients (i.e. P, K, Ca, Mg, S, Na, Mn, Cu, and Zn). Total C and N were determined using by high-temperature combustion with an Elementar VarioMAX Total C and N Analyzer (Elementar Americas Inc. Mt. Laurel, NJ).

Soil samples were collected using a standard push probe immediately after soil respiration measurements to assess soil microbial carbon and nitrogen. Five cores were collected from the top 10 cm around each flux collar of the first three replications in all nine plots for a total of 27 soil samples collected per month. Samples were stored at approximately 4°C until analyzed.

Samples were sieved moist through a 2-mm sieve that had been washed with soap and sterilized with ethanol in a Bunsen burner flame. To obtain the moisture content for each sample, a 10-g subsample was dried at 105°C for 24 hours. Biological organic carbon (BOC) and biological total nitrogen (BTN) concentration was determined using a modified chloroform-fumigation extraction method (Jenkinson and Powlson, 1976). Two, 8 g moist subsamples from each sample were weighed out, one set for a non-fumigated organic carbon extraction and the other set for a fumigated organic carbon extraction. For fumigation, samples were placed in a

desiccator for 24 hours with a temperature range of 21 to 24°C with chloroform. After 24 hours, the desiccator was attached to a vacuum pump and the chloroform vapor was evacuated out of the container. Forty milliliters of 0.5 *M* potassium sulfate (K<sub>2</sub>SO<sub>4</sub>) was added to each sample in both groups. The samples were shaken for 30 minutes on a slow speed in an oscillating shaker (approximately 200 oscillations min<sup>-1</sup>). The samples were then filtered through Whatman no. 42 filter paper (Whatman Int., Maidstone, UK) to separate the supernatant. The supernatant was analyzed for organic carbon using a Total Organic Carbon Analyzer with a total nitrogen unit (TOC-V Model, Shimadzu Scientific Instruments, Columbia, MD). Microbial biomass carbon and nitrogen was determined as the biological portion of organic carbon and total nitrogen, respectively, calculated by the difference in carbon from fumigated and unfumigated samples. The biological carbon was then corrected with the measured moisture content and reported on a dry-weight basis.

#### **Statistical Analyses**

Based on a completely randomized design with three replications (Figure 1), the effect of ecosystem (i.e., switchgrass, cottonwood, and agro-ecosystem) on soil particle-size distribution (measured in 2012), 0-10 cm (measured in 2012 and 2013) and 10-20 cm (measured in 2012) bulk density, and 0-10 cm and 10-20 cm soil chemical properties (P, K, Ca, Mg, S, Na, Fe, Mn, Zn, Cu, SOM, TN, and TC contents; measured in 2012 and 2013) were evaluated by analysis of variance (ANOVA) using SAS (version 9.1, SAS Institute Inc., Cary, NC). In addition, the effect of time (2012 and 2013) on 0-10 cm bulk density and 0- 10 cm and 10-20 cm soil chemical properties was assessed by ANOVA. An ANOVA was also conducted to evaluate the effects of ecosystem (i.e., switchgrass, cottonwood, and agro-ecosystem), month, year, and their

interactions on soil respiration, 0-10 cm biological soil carbon and nitrogen, 0-6 cm volumetric water content, and 2- and 10-cm soil temperature. An ANOVA was conducted to evaluate the effects of collar placement (i.e. in-row and between-row), month, year, and their interactions on soil respiration from the agro-ecosystem only. Linear correlation and multiple regression analyses were preformed to evaluate the relationships among 0-6 cm volumetric water content and 2- and 10-cm soil temperature, their quadratic terms, and the product terms of the 2- and 10-cm soil temperature with soil respiration.

An ANOVA was conducted to evaluate the effects of ecosystem (i.e., switchgrass, cottonwood, and agro-ecosystem), year (i.e., 2012 and 2013), and their interaction on annual CO<sub>2</sub> emissions. In addition, an ANOVA was conducted to evaluate the effects of ecosystem on the 2-year cumulative CO<sub>2</sub> emissions. An ANOVA was conducted separately by year to evaluate the effects of collar placement (i.e., in-row and between-row) on annual CO<sub>2</sub>emissions in the agroecosystem only. When appropriate, means were separated by the most conservative least significant difference (LSD) at  $\alpha = 0.05$ .

#### **Results and Discussion**

### **Initial Soil Physical and Chemical Properties**

With the exception of clay and extractable soil Mn, all initial soil properties measured in May 2012 differed among ecosystems, between soil depths, or among ecosystem-depth combinations (Table 1). Averaged across ecosystems, bulk density and sand and silt fractions differed between the 0-10 and 10-20 cm depths (p < 0.05; Table 1). As expected, bulk density was 8% greater in the 10-20 than in the 0-10 cm depth (Table 2). Sand was 2% greater and silt was 3% less in the 10-20 than in the 0-10 cm depth (Table 2). The clay fraction was unaffected

by any treatment (Table 1) and averaged 0.16 g  $g^{-1}$  in the top 20 cm across all treatment combinations.

In contrast to physical properties, numerous initial soil chemical properties differed among ecosystems, between soil depths, and among ecosystem-depth combinations (Table 1) in 2012 after three years of consistent management since treatment establishment in 2009. Soil EC and extractable soil Zn differed among ecosystems (p < 0.05; Table 1). Averaged across soil depths, soil EC was lower in the agroecosystem (0.05 dS m<sup>-1</sup>) than in the switchgrass (0.06 dS m<sup>-1</sup>) and cottonwood (0.07 dS m<sup>-1</sup>) ecosystems, which did not differ. Averaged across soil depths, extractable soil Zn was greater in the cottonwood (1.6 kg ha<sup>-1</sup>) than in the agroecosystem (1.0 kg ha<sup>-1</sup>) and switchgrass (1.0 kg ha<sup>-1</sup>), which did not differ.

Soil EC, extractable soil S, Fe, Zn, and Cu, SOM, TC, and TN contents, and C:N ratio differed between soil depths (p < 0.03; Table 1). Averaged across ecosystems, soil EC, extractable soil Fe, Zn, Cu, SOM, TC, and TN contents, and C:N ratio were greater in the top 10 cm than in the 10-20 cm depth (Table 2). In contrast, extractable soil S was greater in the 10-20 cm depth than in the top 10 cm (Table 2).

Soil pH and extractable P, K, Ca, Mg, and Na contents differed between soil depths among ecosystems (p < 0.05; Table 1). Soil pH was lower in the 10-20 cm depth in the agroecosystem than in all other ecosystem-depth combinations, which did not differ (Figure 2). Extractable soil P and K contents were greatest in the top 10 cm of the agroecosystem than in all other ecosystem-depth combinations (Figure 2) likely due to previous additions of P and K fertilizers in 2011 to optimize crop growth and production. Extractable soil P and K contents were at least numerically smallest in the 10-20 cm depth and did not differ among ecosystems (Figure 2). Extractable soil Ca was greater in the top 10 than in the 10-20 cm depth in the agroecosystem, but was similar between soil depths in the switchgrass and cottonwood ecosystems (Figure 2). In contrast to P and K, extractable soil Mg was greater in both depths of the cottonwood ecosystem, which did not differ, than in both depths of the agroecosystem, which did not differ (Figure 2). Also in contrast to P and K, extractable soil Na was greatest in the 10-20 cm depth in the cottonwood ecosystem than in any other ecosystem-depth combination. Extractable soil Na was lowest in the top 10 cm and did not differ among ecosystems (Figure 2).

### Soil CO<sub>2</sub> Flux, Temperature, and Moisture

## 2012 – A Dry Year

In 2012, soil CO<sub>2</sub> fluxes from all ecosystems followed the same general trend. Soil CO<sub>2</sub> flux started low in the first few months, slowly increased from January until May, peaked during July, and slowly decreased from August to December (Figure 3). Soil temperatures at the 2- and 10-cm depths followed a similar pattern as did CO<sub>2</sub> flux, starting low early in the year, increasing to a maximum during June, and decreasing thereafter to December (Figure 3). In contrast, the 0- 6 cm volumetric soil water content started large early in the year, decreased to a minimum in June and August with a sharp increase in July, and increased after August to December (Figure 3). Overall, 2012 could be characterized as a drought year with 31% lower than average rainfall.

Soil CO<sub>2</sub> flux differed among ecosystems (p = 0.011) and differed among months (p < 0.001) throughout 2012 (Table 3). Averaged over time, soil CO<sub>2</sub> flux was greater in the agroecosystem and the switchgrass ecosystem (2.7 and 2.6 µmol m<sup>-2</sup> s<sup>-1</sup>, respectively) than in the cottonwood, which averaged 1.94 µmol m<sup>-2</sup> s<sup>-1</sup>, for 2012.

Averaged over ecosystems, soil CO<sub>2</sub> fluxes were lowest (i.e.,  $< 1.8 \ \mu mol \ m^{-2} \ s^{-1}$ ) during January, November, and December when soil water contents were greater than 30 % (v/v) in the

top 6 cm (Figure 3). Franks and Dugas (2001) reported a similar trend during the winter months in a tallgrass prairie in North Dakota on a variety of loam, silt loam, and silty clay loam soils. Soil CO<sub>2</sub> fluxes were greatest for all three ecosystems in July (i.e., a mean of 8.1  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> across ecosystems) during which time the soil water contents in the top 6 cm significantly increased to ~ 25 % (v/v) following a period when soil water contents were low [~ 10% (v/v); Figure 3] after a prolonged period of minimal rainfall (Figure 4). The months of low and peak CO<sub>2</sub> fluxes also corresponded to months when soil temperatures were low and relatively high respectively (Figure 3). These results generally support previous reports that soil temperature is a direct controlling factor, while soil moisture is an inverse controlling factor on soil respiration (Smith et al., 2014; Brye and Riley, 2009; Lee et al., 2007; Frank and Dugas, 2001). Some studies have shown the inverse relationship between soil temperature and respiration to be weak and generally vary more than the relationship between soil temperature and respiration, except during moisture limiting periods (Brye and Riley, 2009; Lee et al., 2007)

There are few studies on soil respiration in switchgrass. Brye and Riley (2009) reported  $CO_2$  flux did not systematically differ among 3-, 4-, 5-, 26-year old prairie restorations and a native, mixed tallgrass prairie ecosystem on fine-textured soils in the Ozark Highlands of Arkansas. In addition, the peak flux reported for the study occurred in the 26-year old prairie restoration (~12 µmol m<sup>-2</sup> s<sup>-1</sup>; Brye and Riley, 2009). Another study from the Ozark Highlands on a Captina silt loam reported peak fluxes around 31 µmol m<sup>-2</sup> s<sup>-1</sup> 5 days after broiler litter application (McMullen, 2014).

In similar studies across the United States, peak  $CO_2$  fluxes from grassland soils ranged from 1.7 to 37.5 µmol m<sup>-2</sup> s<sup>-1</sup> (Bremer et al., 2002; Al-Kaisi and Grote, 2007; Lee et al., 2007). Lee et al. (2007) reported peak flux occurred in an established switchgrass plot fertilized with manure harvested every other year (12.5  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) on a silty clay loam in South Dakota compared to treatment combinations of non-fertilized or ammonium nitrate fertilized soil and annual or bi-annual harvesting. Also noted by Lee et al. (2007), harvesting aboveground switchgrass biomass significantly increased soil temperature. Annual burning of switchgrass in Iowa also produced a greater soil CO<sub>2</sub> flux compared to switchgrass burned every five years (21.7 and 16.9  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, respectively ), while overall, switchgrass, regardless of burning regime, had the greatest increase in soil carbon compared to a soybean-corn rotation when grown on exposed loam subsoil (9.7  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>; Al-Kaisi and Grote, 2007). Bremer and Ham (2002) reported a peak CO<sub>2</sub> flux of 13.0  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> from an ungrazed tallgrass prairie on a silty clay loam in Kansas. Also reported was the considerable spatial variability range of CO<sub>2</sub> flux measurements, 3 to 9  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, due to spatial heterogeneity in soil moisture content and physical properties (Bremer and Ham, 2002).

There are no previous studies on soil respiration in monoculture cottonwood trees in the Arkansas. However, similar studies have been conducted in the United States; however, most of these studies have focus on mixed deciduous vegetation and northern geographic locations such as Utah, Pennsylvania, and Massachusetts. Bowden et al. (2000) reported a peak  $CO_2$  flux of approximately 3.8 µmol m<sup>-2</sup> s<sup>-1</sup> from a temperate deciduous forest on a silty loam in Pennsylvania after a dramatic increase in soil temperature from approximately 9 to 18°C. A study from a mixed forest in Massachusetts on a silt loam reported greater peak fluxes from a swamp location compared to a poorly drained site (4.6 and 6.9 µmol m<sup>-2</sup> s<sup>-1</sup>, respectively; Davidson et al., 1998). A study conducted along a riparian zone of the Weber River, in northern Utah, reported  $CO_2$  fluxes of approximately 0.5 to 1.8 µmol m<sup>-2</sup> s<sup>-1</sup> along a hybridization gradient of *Populus augustifila* and *Populus fermontii*, respectively (Fischer et al., 2007). In
addition, Fischer et al. (2007) concluded that cottonwood species was a significant factor when quantifying  $CO_2$  emissions, where  $CO_2$  emissions were 1.5 times greater from *P. femontii* than from *P. angustifolia*, suggesting that species might be a significant factor in soil respiration when monoculture stands are present.

Soybean-wheat rotations on a silt-loam in eastern Arkansas have been reported to have peak CO<sub>2</sub> fluxes between 10 and 41 µmol m<sup>-2</sup> s<sup>-1</sup>, while corn-based agroecosystems have reported CO<sub>2</sub> fluxes ranging from 9.6 to over 20 µmol m<sup>-2</sup> s<sup>-1</sup> (Amos et al., 2005; Brye et al., 2006; Al-Kaisi and Grote, 2007; Motschenbacher, 2012; Smith et al. 2014). Furthermore, Brye et al. (2006) reported greater peak CO<sub>2</sub> fluxes from double-cropped soybean following wheat under conventional tillage than from no-tillage in a silt-loam soil in eastern Arkansas. Thus, it is clear that field and agronomic management practices can have a large impact on soil respiration. Smith et al. (2014) reported wheat residue burning increasing soil respiration by 39% compared to a non-burned, no-tillage soybean-wheat rotation system, while a non-burned, conventional tillage treatment combination increased soil respiration by 84% compared to a non-burned notillage system on a silt-loam soil in eastern Arkansas. In a similar study of rice-based crop rotations on a silt loam in eastern Arkansas, soil respiration from soybean-wheat rotations ranged from 2.3 to 2.8  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>, while rotations with corn (i.e., either corn-rice or rice-corn-soybean) had greater soil respiration rates ranging from 4.7 to 5.8 µmol m<sup>-2</sup> s<sup>-1</sup> (Motschenbacher, 2012). In addition, Motschenbacher (2012) reported great variability in CO<sub>2</sub> fluxes with respect to tillage and crop rotation and relationships among CO<sub>2</sub> flux and management practices may fluctuate during the growing season as crops mature. These results highlight the complex relationship between management practices and soil respiration.

Though  $CO_2$  flux did not differ among ecosystems over time in 2012 (Table 1),

differences in 2- and 10-cm soil temperatures were measured among ecosystems on seven and five of 12 measurement dates, respectively (p < 0.001; Figure 3). With the exception of in March when both the 2- and 10-cm soil temperatures were greater in the switchgrass than in the agroecosystem, the 2- and 10-cm soil temperatures greater in the agroecosystem than in the switchgrass on all other measurement dates when soil temperatures differed among ecosystems (Figure 3). Irradiance of exposed soil before canopy closure and lack of ground cover in the agroecosystem is a likely explanation for the increased soil temperatures (Bremer et al. 2002).

Similar to CO<sub>2</sub> flux, the volumetric soil water content in the top 6 cm differed over time (p < 0.001; Table 1; Figure 3), but not among ecosystems (p > 0.05; Table 1). Soil water contents in the top 6 cm were greatest, > 40 % (v/v) on average, during January, February, March and December and lowest, ~ 10 % (v/v) on average, during June and August (Figure 3). The low soil water contents in June were expected considering this month had below normal rainfall, 90% lower than 30-year averages for the month (Figure 4). August, however, was 0.5% greater than the 30-year average for the month.

#### 2013 – A Wet Year

Compared to 2012, which had 31% below-average precipitation, 2013 was a much wetter year with 18% above-average precipitation (Figure 4). Davidson et al. (1998) reported that poorly drained soil conditions negatively impacted soil  $CO_2$  flux in a mixed hardwood forest in Massachusetts relative to well-drained soil conditions within the forest. Despite the differences in annual precipitation, soil  $CO_2$  flux in 2013 (Figure 5) followed the same general trend as in 2012 (Figure 3). Fluxes started low in the beginning of the year, during the winter months when the soil was wet and the soil temperatures were low (Figure 5), increased throughout the spring months, peaked during the summer months, and decreased during the fall months until fluxes in December were similar to those at the beginning of the year (Figure 5). In addition, soil temperature at both depths (Figure 5) followed a similar general trend as in 2012 (Figure 3). Soil water content patterns in 2013 were similar to those in 2012 early and late in the year. However, in contrast to 2012, soil water contents in the top 6 cm were generally much greater during the summer months of 2013 (i.e., May through August) when rainfall was 11cm above average (Figure 4).

In contrast to 2012, soil CO<sub>2</sub> fluxes throughout 2013 differed among ecosystems over time (p < 0.001; Table 1). On four of 12 measurement dates, soil CO<sub>2</sub> flux differed among ecosystems, while there were no differences among ecosystems on the remaining eight measurements dates (Figure 5). Soil  $CO_2$  flux was greater from the agroecosystem than from the other two ecosystems, which did not differ, in April and July 2013, at which time both soil temperatures were also greatest in the agroecosystem, while soil moisture in the top 6 cm did not differ among ecosystems (Figure 5). Similar to that in 2012, the peak soil  $CO_2$  flux from the agroecosystem (8.0  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) also occurred in July in 2013, in which the magnitudes of the peak fluxes were strikingly similar in both years. However, in contrast to 2012 when the peak soil  $CO_2$  flux from all three ecosystems occurred in July, the peak soil  $CO_2$  flux from both the switchgrass (5.9 µmol m<sup>-2</sup> s<sup>-1</sup>) and cottonwood (4.1 µmol m<sup>-2</sup> s<sup>-1</sup>) ecosystems occurred in May in 2013 when the flux from the switchgrass was greater than that from the agroecosystem (Figure 5). During May 2013, the soil water content in the top 6 cm and the 2-cm soil temperature were also greater in the switchgrass than in the other two ecosystems, while the 10-cm soil temperature did not differ among ecosystems (Figure 5). Despite occurring in different months

in 2013, rather than in the same month in 2012, peak  $CO_2$  fluxes for each ecosystem occurred when soil water contents were within an optimum range, ~25 to 40% (v/v), and when the 2- and 10-cm soil temperatures were above 25°C (Figure 5).

However, the peak soil CO<sub>2</sub> fluxes measured in 2013 in the switchgrass and cottonwood ecosystems represented only 26 and 49%, respectively, of the peak flux measured in 2012. Similar to that in May, soil CO<sub>2</sub> flux was greater from the switchgrass than from the cottonwood ecosystem in August 2013 when soil water content was also greater in the switchgrass than in the other two ecosystems, which did not differ, despite any differences in 2- or 10-cm soil temperature among ecosystems (Figure 5). Though soil CO<sub>2</sub> flux differences were not measured, soil water contents and/or temperatures also differed among ecosystems in March, June, September, and October 2013 (Figure 5). All ecosystems experienced an increased CO<sub>2</sub> flux in October after rainfall following a period of drought in September even though the 2- and 10-cm soil temperatures decreased to approximately 20°C (Figures 4 & 5). In addition, the 0-6cm volumetric water content also increased dramatically from September to October after rainfall (Figure 5). Franzluebbers et al. (2002) reported that neither soil temperature nor water filled pore space had a positive effect on soil respiration when the other factor fell below a base level, approximately 10°C and 0.4 m<sup>3</sup> m<sup>-3</sup>, respectively in a tall grass prairie on a loam in Kansas.

## **Annual Soil CO<sub>2</sub> Emissions**

Despite 2012 and 2013 precipitation differing widely from the 30-yr mean, averaged across ecosystems, annual estimated CO<sub>2</sub>-C emissions did not differ between years. However, annual estimated CO<sub>2</sub>-C emissions similarly differed (p = 0.001) among ecosystems in both years. Annual estimated CO<sub>2</sub>-C emissions were similar from the agroecosystem and the

switchgrass ecosystem and both were greater than that from the cottonwood ecosystem in both years (Figure 6). Lower emissions from the cottonwood could be due to the ecosystem becoming established and a microbial community dominated by fungi, which better adapted to the breakdown of complex cellulose and lignin associated with litterfall from trees. Also, the effects of shading from the branches and leaves could decrease irradiance to the soil surface, decreasing soil temperature and microbial activity. Also, overall decreased root respiration due to physiological differences of the cottonwood compared to switchgrass and the agroecosystem surely contributed to the differences in annual emissions.

Annual estimated CO<sub>2</sub>-C emissions in 2012 from the agroecosystem (i.e., soybean), switchgrass, and cottonwood ecosystems were 10.3, 9.5, and 7.3 Mg CO<sub>2</sub>-C ha<sup>-1</sup>, respectively, while in 2013 annual estimated CO<sub>2</sub>-C emissions were 9.7, 9.2, and 7.4 Mg CO<sub>2</sub>-C  $ha^{-1}$  from the agroecosystem (i.e., grain sorghum), switchgrass, and cottonwood ecosystems, respectively. In addition, annual emissions from the crop rotation did not differ for 2012 and 2013 (p > 0.05, figure 6). Despite the climatic extremes for both 2012 and 2013, particularly for precipitation, annual estimated CO<sub>2</sub>-C emissions from all three ecosystems in the Lower Mississippi River Valley were consistent over the two years of this study. These results suggest that, while soil moisture and temperature are known regulating factors for CO<sub>2</sub> flux; overall CO<sub>2</sub>-C emissions during a given annual cycle are much less sensitive to inter-annual climatic variations. It must be noted that lack of irrigation for both the soybean and grain sorghum in the agroecosystem crop rotation could have affected the CO<sub>2</sub> emissions from the treatment for both years, and irrigation of the rotation, which is common in the LMAV, could have greatly increased CO<sub>2</sub> emission values altering the statically differences between the years for the agroecosystem treatment themselves and with the other treatments.

There is great variation in annual emissions data, which suggests that environmental factors, such as temperature and moisture, play an important, though differing, role in controlling soil respiration depending on location. Furthermore, it is clear that different ecosystems, with differing vegetation characteristics, emit varying quantities of CO<sub>2</sub>-C. Annual emission estimates from studies conducted on cottonwood and other hardwood species show the greatest similarity to the results of this study. Results from Davidson et al. (1998) and (2002) in mixed hardwood forests on a Tennessee Typic Paleudult and a silty loam in Massachusetts showed that annual emissions ranged from 5.3 Mg CO<sub>2</sub>-C ha<sup>-1</sup> in poorly drained soil conditions in valleys to 9.2 Mg  $CO_2$ -C ha<sup>-1</sup> in well-drained soil conditions on ridges, suggesting that soil drainage (i.e., moisture) likely plays an important role in moderating both CO<sub>2</sub> fluxes and emissions.

In contrast, results of previous studies showed that annual CO<sub>2</sub> emissions from switchgrass and similar tallgrass prairies ranged from a low of 5 to 6.2 Mg CO<sub>2</sub>-C ha<sup>-1</sup> in Iowa from a loam sub-soil, after topsoil removal during road construction, when burning regimes of once annually and once every 5 years, respectively, were implemented (Al-Kaisi and Grote, 2007) to a high of 10.7 to 13.4 Mg CO<sub>2</sub>-C ha<sup>-1</sup> from a tallgrass prairie in Kansas (Bremer and Ham, 2002). Emissions of approximately 3.5 Mg CO<sub>2</sub>-C ha<sup>-1</sup> were also reported from soybean in a soybean-corn rotation (Al-Kaisi and Grote, 2007), where the difference compared to the more natural switchgrass and tallgrass prairie ecosystems in Iowa can probably be attributed to the infertility of the sub-soil and general lack of productivity from the agroecosystem.

Similar studies in Arkansas have focused on traditional crop production, such as soybean corn, but only quantifying emissions within the growing season. Smith et al. (2014) reported growing season emissions from a soybean-wheat rotation in eastern Arkansas on a silt loam ranging from 4.5 to 6.0 Mg  $CO_2$ -C ha<sup>-1</sup>. Another study in eastern Arkansas on a silt loam

reported growing season emissions ranging from 3.8 to 6.8 Mg  $CO_2$ -C ha<sup>-1</sup> from wheat, corn, soybean rotations with rice (Motschenbacher, 2012). Amos et al. (2005) reported 11.5 Mg  $CO_2$ -C ha<sup>-1</sup> in annual emissions from soil in an irrigated maize-based agroecosystem on a silt loam in Nebraska.

## **Relationships among Soil Temperature, Moisture and CO<sub>2</sub> Flux**

As others have reported (Davidson et al., 1998; Dilustro et al., 2005; Lee et al., 2007, Brye et al., 2009), soil moisture and temperature are often significantly correlated with soil CO<sub>2</sub> flux. In this study, soil water content in the top 6 cm (r = -0.135; p < 0.01), 2-cm soil temperature (r = -0.092; p < 0.01), and 2-cm-soil temperature squared (r = 0.592; p < 0.01) were significantly correlated with soil CO<sub>2</sub> flux when data were combined across time and all ecosystems. Soil temperatures at the 2- and 10-cm depths were highly correlated to one another; therefore, only the 2-cm soil temperature was used in the subsequent regression analyses. The product term for 2-cm soil temperature and soil moisture was not significantly correlated to soil respiration.

When combined across time and all ecosystems, 0-6 cm soil water content, 2-cm soil temperature, and the quadratic term for 2-cm temperature explained 37.8% of variation in CO<sub>2</sub> flux (p < 0.05, Table 4). In this all-ecosystems model, the strongest predictive variable was the linear term for 2-cm soil temperature, which explained 92% of the total sum of squares. The water content and quadratic term for 2-cm temperature explained only 4.8 and 3.1% of the total sum of squares, respectively. Lee et al. (2006) reported soil temperature was highly correlated to CO<sub>2</sub> flux, while soil moisture and CO<sub>2</sub> flux were not highly correlated due to a small seasonal range from a switchgrass stand managed for bioenergy production on a silt clay loam in South

Dakota. Lee et al. (2006) also suggested that soil moisture would be more highly correlated when soil moisture is a limiting factor. These results were similar to those from other studies conducted on silt-loam soils in the Lower Mississippi River Valley region of eastern Arkansas (Brye et al., 2006; Motschenbacher, 2012; Smith et al., 2014) where significant relationships were reported among soil temperature, moisture, and soil respiration, with soil moisture generally having a weaker relationship to soil respiration than soil temperature.

When the all-ecosystems multiple regression model was fit to the data from each ecosystem separately, the quadratic term for 2-cm temperature was non-significant in the model for the agroecosystem (p > 0.05). However, model coefficients from fitting the all-ecosystems multiple regression model to the data from each ecosystem separately did not differ from the coefficients from the all-ecosystems model (Table 4). This result suggests that, while the three ecosystems evaluated in this study (i.e., switchgrass, cottonwood, and an agroecosystem) had different vegetation characteristics, the predictive relationship among soil temperature, moisture and  $CO_2$  flux was consistent across the three ecosystems evaluated in this study. In other words, under the similar climatic regime and soil characteristics and conditions of this study, it appears that a single, multiple regression model using the linear and quadratic terms for 2-cm soil temperature and the linear term for 0-6 cm soil water content may be adequate to predict soil CO<sub>2</sub> fluxes from a grass or woody biomass bioenergy crop or a traditional agroecosystem. This is consistent with what others have reported in Arkansas. Smith et al. (2014) also reported that a single regression model, including soil moisture and temperature, was adequate for prediction of  $CO_2$  flux in a wheat-soybean double cropping system on a silt loam in Arkansas.

#### **Microbial Biomass**

Microbial biomass C and N measured in the top 10 cm during the 2013 growing-season months (i.e., April through October) differed over time (p < 0.001, Table 5). The largest microbial biomass C concentrations were observed in May and October (91.9 and 87.3  $\mu$ g g<sup>-1</sup>, respectively), while April had the lowest microbial biomass C concentration among any month, which was ~ 33% of the concentration measured in May (Figure 7). Similar to C, microbial biomass N concentration was largest in May (15.38  $\mu$ g g<sup>-1</sup>; Figure 7). September had the lowest microbial biomass N concentration, which was less than one third of that measured in May (Figure 7). This could be related to dry conditions in September, inhibiting bacteria allowing fungi to dominate and a general decrease in soil respiration for the measurement date (Figure 5). Lauber et al. (2013) reported that bacterial taxa and lineage diversity where more variable temporally than across land uses (i.e. conventional corn, reduced input corn with cover crop, and early successional grass), while bacterial community composition differed more across land uses rather than temporally on a loam soil in Michigan. Lauber et al. (2013) also reported that the conventional corn agricultural system had significantly greater temporal variability relative to other land uses, possibly the result of plant community composition and phenology

In contrast to microbial biomass C and N concentrations, the microbial biomass C:N ratio differed among ecosystems over time (p = 0.018; Table 5). The largest microbial biomass C:N ratio (21.1) occurred in the grain-sorghum agroecosystem in September (Figure 7), which could indicate a shift in the microbial community during an extremely dry period to which the fungi are more adapted, when the ecosystem had been previously dominated by bacteria. Relatively low microbial biomass C:N ratios (i.e., < 8) were measured for the majority of the ecosystem-month combinations (Figure 7). Microbial biomass C:N ratios can indicate shifts in microbial community dominated by bacteria in microbial biomass C:N ratios (i.e., 12:1 to 20:1) are indicative of

a bacteria-dominated community, while lower ratios indicate more fungal biomass could be present. Culman et al. (2010) reported significantly greater soil microbial biomass in switchgrass stands than in cultivated agroecosystems on silt-loam, clay-loam, and silty-clay soils in Kansas at soil depths to 80 cm. Haney et al. (2010) also reported switchgrass having greater microbial biomass than a corn agroecosystem on a black clay in Texas. Greater microbial biomass is often associated with greater SOM mineralization rates and an increase in general soil fertility (Culman et al., 2010; Haney et al., 2010).

#### Growing-season Relationships among Soil Microbial Biomass, Moisture, and CO<sub>2</sub> Flux

Similar to the regression analyses conducted with all data across all ecosystems and measurement dates, a multiple regression procedure was conducted using soil CO<sub>2</sub> flux, 2-cm soil temperature, 0-6 cm soil water content, and soil microbial biomass C (MBC) and N (MBN) concentration data from only the growing-season months of April through October 2013 to evaluate the predictive relationship among these variables. As others have reported, soil moisture and temperature are often significantly correlated with soil CO<sub>2</sub> flux (Culman et al., 2010; Haney et al., 2010). In this study, soil water content in the top 6 cm (r = 0.277; *p* < 0.01), its quadratic term (r = 0.189; *p* < 0.05), and the MBN quadratic term (r = 0.352; *p* < 0.01, and their quadratic terms) were significantly correlated to CO<sub>2</sub> flux when data were combined across ecosystems during the 2013 growing season. Combined across ecosystems, 26% of the variation in soil respiration was explained by soil water content, its quadratic term, and the quadratic term for MBN for the growing-season months of April through October 2013 (*p* < 0.05; Table 6). The strongest predictive variables were VWC, and quadratic terms for VWC and MBN, which explained 29.5, 44.6, and 25.8% of the total sum of squares, respectively.

There was great variation in the significance of variables when the all-ecosystem model was applied separately to data from each ecosystem (Table 6). For the agroecosystem and the switchgrass ecosystem, VWC and its quadratic term were non-significant in the model with MBN the only significant variable from the all ecosystem model, while for the cottonwood ecosystem the only terms significant were VWC and its quadratic term (Table 6). Lee et al. (2006) reported MBC and CO<sub>2</sub> flux were not highly correlated and suggested seasonal changes in MBC were not responsible for changes in  $CO_2$  flux. Also, it was reported by Lauber et al. (2013) that bacterial taxa and lineage was poorly correlated to CO<sub>2</sub> flux, and temporal variation in bacterial community diversity was unrelated to changes in biogeochemical changes across the time scale. Despite numerous models with non-significant terms compared to the terms significant in the all-ecosystems model, the multiple regression models fit to the data separately by ecosystem, the agroecosystem and the cottonwood ecosystem had larger coefficients of determination ( $r^2 > 0.304$  and 0.321, respectively) than that for the all-ecosystems model ( $r^2 =$ 0.260; Table 6). This result indicates slightly greater predictive power when relationships among soil properties and CO<sub>2</sub> flux were modeled separately by ecosystem, which was somewhat different from the conclusion drawn when the whole 2-yr data set was evaluated.

Unlike the multiple regression analysis conducted using the entire 2-yr data set, which was dominated by soil temperature and moisture, soil temperature was not significant in the allecosystem model when MBC and MBN were included from only the 2013 growing-season months. However, this is to be expected since the microbial biomass only accounted for the growing season when temperature would be at optimum levels for soil respiration and other factors, such as soil moisture and the microbial community, play larger roles in regulating soil respiration. These results also indicate that MBN might be more useful for describing soil

microorganism respiration than MBC alone, as MBN is more sensitive to changes within the community.

## Collar Placement Effects on CO<sub>2</sub> Flux in the Agroecosystem

A small sub-objective of this study was to determine if collar placement within or between crop rows would affect soil CO<sub>2</sub> flux measurements. In both years, there was no effect of collar placement on soil CO<sub>2</sub> flux (p = 0.252 and 0.575, respectively; Table 7). Despite theoretically more concentrated plant roots within a crop row than between crop rows, the 19-cm row spacing in the drill-seeded soybean in 2012 and the 19-cm row spacing for the grain sorghum in 2013 were not large enough to result in CO<sub>2</sub> flux differences within and between rows. However, similar to previous analyses, soil CO<sub>2</sub> fluxes differed over time in both years (p< 0.001; Table 7). These results indicate that collar placement need not be taken into account in a soybean-grain sorghum rotation when rows are spaced relatively close. However, Amos et al. (2005) reported a 64% greater CO<sub>2</sub> flux from measurements made in the row compared to those made between rows in an irrigated maize-based agroecosystem in Nebraska with 76-cm row spacing. Differences in the results of this study and those reported by Amos et al (2005) are likely due to the row-spacing differences for the different crops.

#### **Summary and Conclusions**

This study demonstrated that switchgrass and cottonwood grown as bioenergy feedstocks in the LMAV did not increase soil respiration relative to a traditional soybean-grain sorghum agroecosystem. Although 2012 and 2013 differed greatly in precipitation, similar trends for soil temperature, moisture and  $CO_2$  flux were observed for both years. As predicted, all treatments showed general trends in  $CO_2$  flux throughout the year. Carbon dioxide fluxes increased from winter lows throughout the spring, peaking during summer months, then falling during autumn back to winter lows. Additionally, the greatest  $CO_2$  flux measured was in the agroecosystem, as predicted. However, collar placement did not affect measurements in the agroecosystem for either year.

In general, the agroecosystem and the switchgrass ecosystem were similar in CO<sub>2</sub> fluxes throughout both years, while the cottonwood had generally lower fluxes throughout both years. This trend was also evident when annual emissions from the ecosystems were quantified. Contrary to what was hypothesized, annual emissions from the agroecosystem and switchgrass ecosystem were similar, while the cottonwood had significantly lower annual emissions. Soil MBC and MBN did not differ significantly by treatment for the 2013 growing season, unlike what was predicted, however, the agroecosystem did have the greatest C:N ratio

As predicted, soil temperature and moisture play large roles in controlling soil respiration, and can be used to account for a large portion of the variation in soil respiration. However, this study showed that, when only accounting for growing-season  $CO_2$  flux, when soil temperature are more consistent, other parameters, such as MBN, should be taken into account. This suggests that, during optimum soil temperature conditions, other variables form a more complex set of controlling factors. This study suggests that switchgrass and cottonwood, grown as bioenergy feedstocks in the LMAV, do not increase soil respiration compared to a regionally common agroecosystem. In addition, cottonwood grown as bioenergy feedstock may decrease soil respiration, which may eventually help to increase soil C stocks in the LMAV.

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	Source of Variation			
Soil Property	Ecosystem	Depth	Ecosystem*depth	
<u> </u>		p		
Bulk Density	0.657	< 0.001	0.319	
Sand	0.557	0.045	0.930	
Silt	0.739	< 0.001	0.133	
Clay	0.972	0.337	0.608	
рН	0.346	0.125	0.043	
Electrical Conductivity	0.045	0.002	0.452	
Extractable P	0.184	< 0.001	0.027	
Extractable K	0.123	< 0.001	< 0.001	
Extractable Ca	0.356	< 0.001	0.001	
Extractable Mg	0.071	< 0.001	0.003	
Extractable S	0.382	0.027	0.227	
Extractable Na	0.094	< 0.001	0.010	
Extractable Fe	0.936	< 0.001	0.921	
Extractable Mn	0.048	0.583	0.608	
Extractable Zn	0.048	< 0.001	0.335	
Extractable Cu	0.316	< 0.001	0.570	
Organic Matter	0.998	< 0.001	0.760	
Total Carbon	0.256	< 0.001	0.250	
Total Nitrogen	0.403	< 0.001	0.145	
C:N	0.404	< 0.001	0.506	

**Table 1.** Analysis of variance summary of the effects of ecosystem, soil depth, and their interaction on 2012 soil properties in the top 20 cm.

	Depth	
Soil Property	0-10 cm	10-20 cm
Bulk Density (g cm <sup>-3</sup> )	1.32b	1.43a
Sand $(g g^{-1})$	0.106b	0.126a
Silt $(g g^{-1})$	0.739a	0.71b
Electrical Conductivity (dS m <sup>-1</sup> )	0.067a	0.057b
Extractable S (kg ha <sup>-1</sup> )	11.1b	17.0a
Extractable Fe (kg ha <sup>-1</sup> )	216.3a	159.5b
Extractable Zn (kg ha <sup>-1</sup> )	1.59a	0.815b
Extractable Cu (kg ha <sup>-1</sup> )	1.67a	1.47b
Organic Matter (Mg ha <sup>-1</sup> )	31.0a	23.5b
Total Carbon (Mg ha <sup>-1</sup> )	14.0a	7.84b
Total Nitrogen (kg ha <sup>-1</sup> )	1313.3a	834.3b
C:N ratio	10.7a	9.48b

**Table 2.** Soil depth effects on 2012 initial soil properties. Different letters following means in a row are statistically different at the 0.05 level.

Measured property/Source of variation	2012	2013
CO <sub>2</sub> flux		
Ecosystem	0.011	0.003
Month	< 0.001	< 0.001
Ecosystem* month	0.098	< 0.001
2-cm soil temperature		
Ecosystem	0.006	0.002
Month	< 0.001	< 0.001
Ecosystem* month	< 0.001	< 0.001
10-cm soil temperature		
Ecosystem	0.086	0.001
Month	< 0.001	< 0.001
Ecosystem* month	< 0.001	< 0.001
0-6 cm volumetric soil water content		
Ecosystem	0.603	0.394
Month	< 0.001	< 0.001
Ecosystem* month	0.504	< 0.001

**Table 3.** Analysis of variance summary of the effects of ecosystem, month, and their interaction on soil surface carbon dioxide ( $CO_2$ ) flux, 2- and 10-cm soil temperatures, and 0-6 cm volumetric soil water content for 2012 and 2013.

**Table 4.** Multiple regression summary of the effects of volumetric water content (VWC), 2-cm soil temperature (Temp), and its quadratic term on  $CO_2$  flux. Effects considered significant (p < 0.05) are indicated by bolded text. Numbers in parentheses are the upper and lower confidence intervals for each coefficient. Asterisks (\*) indicate where confidence interval half widths of coefficients from each treatment model overlap with the all-ecosystems model.

Model	Intercept	VWC	2-cm Temp	(2-cm Temp) <sup>2</sup>	r <sup>2</sup>
All Ecosystems	-0.729 (± 0.106)	-0.325 (± 0.029)	0.228 (± 0.011)	$-0.002 (\pm 0.0003)$	0.378
Agroecosystem	0.0614 (± 0.213)	-0.629 (± 0.140)	0.17 (± 0.021)	-0.001 (± 0.0004)	0.297
Switchgrass	-1.56 (± 0.190)	-0.258 (± 0.043)*	0.313 (± 0.021)	-0.003 (± 0.001)	0.485
Cottonwood	-0.792 (± 0.127)*	-0.343 (± 0.027)*	0.229 (± 0.014)	-0.003 (± 0.0003)	0.469

**Table 5.** Analysis of variance summary of the effects of ecosystem, month, and their interactions on microbial biomass carbon (MBC), microbial biomass nitrogen (MBN), and the microbial biomass C:N ratio for the 2013 growing season (April through October).

Soil Property	Ecosystem	Month	Ecosystem*month
MBC	0.683	< 0.001	0.240
MBN	0.184	< 0.001	0.281
C:N	0.070	< 0.001	0.018

**Table 6**. Multiple regression summary of the effects of volumetric water content (VWC), its quadratic term, and the quadratic term for microbial biomass nitrogen (MBN<sup>2</sup>) on soil surface carbon dioxide (CO<sub>2</sub>) flux. Effects considered significant (p < 0.05) are indicated by bolded text. Numbers in parentheses are the upper and lower confidence intervals for each coefficient. Asterisks (\*) indicate where confidence interval half widths of coefficients from each treatment model overlap with the all-ecosystems model.

Model	Intercept	VWC	MBN <sup>2</sup>	VWC <sup>2</sup>	r <sup>2</sup>
All Ecosystems	1.06 (± 0.202)	<b>19.98</b> (± <b>1.97</b> )	$0.003 (\pm 2.5 \ge 10^{-4})$	-36.5 (± 4.312)	0.260
Agroecosystem	$1.91 (\pm 0.313)$	12.5 (± 3.11)	<b>0.003</b> (± <b>3.2</b> x 10 <sup>-4</sup> )*	-21.1 (± 6.85)	0.304
Switchgrass	2.30 (± 0.446)	11.19 (± 4.65)	<b>0.006</b> (± <b>0.001</b> )	$-21.4(\pm 9.90)$	0.121
Cottonwood	-0.193 (± 0.309)	27.99 (± 3.23)	0.003 (± 8.7 x 10 <sup>-4</sup> )*	-52.1 (± 6.88)	0.321

**Table 7.** Analysis of variance summary of the effects of collar placement in a soybean-grain sorghum rotation on soil surface carbon dioxide  $(CO_2)$  flux. Soybean was grown in 2012, and grain sorghum was grown in 2013. Collars were placed either in the crop row or between crop rows.

Source of Variation	2012	2013
		<i>p</i>
Collar Placement	0.252	0.575
Month	< 0.001	< 0.001
Collar Placement * month	0.939	0.852



**Figure 1**. Aerial image of study site at the Pine Tree Research Station near Colt, Arkansas. Switchgrass (S1, S2, S3), cottonwood (W1, W2, W3), and soybean-grain sorghum crop rotation (C1, C2, C3) treatments, individual plot locations, and dimensions are noted.



**Figure 2**. Summary of the effects of ecosystem and soil depth on 2012 initial soil chemical properties including: Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), and Sodium (Na), from the Pine Tree Research Station near Colt, Arkansas. Bars with different lower case letters within a soil property indicate differences at the 0.05 level.



**Figure 3.** Monthly mean volumetric soil water contents (VWC), 2- and 10-cm soil temperatures, and soil surface carbon dioxide (CO<sub>2</sub>) fluxes by ecosystem for 2012 measured at the Pine Tree Research Station near Colt, Arkansas. Different lower case letters associated with the VWC and CO<sub>2</sub> flux panels indicate differences in monthly means averaged across ecosystems at the 0.05 level. Asterisks (\*) associated with the 2- and 10-cm soil temperature panels indicate measurement dates with significant differences among ecosystem at the 0.05 level.



**Figure 4.** Annual precipitation events and air temperature for Pine Tree Research Station near Colt, Arkansas for 2012 and 2013. Precipitation events are represented by the bars, and average daily air temperatures represented by the line graph.



**Figure 5.** Monthly volumetric soil water contents (VWC), 2- and 10-cm soil temperatures, and soil surface carbon dioxide (CO<sub>2</sub>) fluxes for 2013 measured at the Pine Tree Research Station near Colt, Arkansas. Asterisks (\*) associated with each panel indicate measurement dates with significant differences among ecosystem at the 0.05 level.



**Figure 6.** Annual carbon dioxide-carbon (CO<sub>2</sub>-C) emissions for ecosystems in 2012 and 2013 at the Pine Tree Research Station near Colt, AR. The 2012 crop in the agroecosystem was soybean, while the 2013 crop was grain sorghum. Lower case letters indicate were means were separated by least significant difference (LSD) at  $\alpha$ =0.05 level.



**Figure 7.** Summary of the monthly differences in microbial biomass carbon (MBC) and nitrogen (MBN) averaged across ecosystems and the effects of ecosystem and month on microbial biomass C:N ratios at the Pine Tree Research Station near Colt, Arkansas during the 2013 growing season. Lower case letters indicate were means were separated by the most conservative least significant difference (LSD) at  $\alpha$ =0.05

# **Appendix I**

SAS Input file for Bulk Density (BD), Sand, Silt, Clay, pH, Electrical Conductivity (EC), Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), Sulfur (Su), Sodium (Na), Iron (Fe), Manganese (Mn), Zinc (Zn), Copper (Cu), Organic Matter (OM), Total Nitrogen (TotN), Total Carbon (TotC, and C: N ratio (C:N).

Title 'Pine Tree Soil Respiration Study - 2012'; options ls = 110 ps = 68;

data soil; infile 'PSA2012.csv' firstobs = 2 delimiter = "," truncover LRECL = 600; input ecosystem \$ plot rep depth \$ BD sand silt clay pH EC P K Ca Mg Su Na Fe Mn Zn Cu OM TotN TotC CN; run;

## proc print data = soil; run;

ods rtf file='psa.rft' bodytitle style=journal; **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model BD= ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model sand = ecosystem depth ecosystem \* depth / ddfm = kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model silt = ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model clay = ecosystem depth ecosystem \* depth / ddfm = kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run; **proc mixed** data = soil method=type3; class ecosystem depth rep plot;

model pH = ecosystem depth ecosystem\*depth /ddfm=kr;

```
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model EC = ecosystem depth ecosystem*depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model P = ecosystem depth ecosystem*depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model K = ecosystem depth ecosystem*depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model Ca = ecosystem depth ecosystem*depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model Mg = ecosystem depth ecosystem ^{e} depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model Su = ecosystem depth ecosystem*depth /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
class ecosystem depth rep plot;
model Na = ecosystem depth ecosystem ^{\text{depth}} /ddfm=kr;
random plot(ecosystem)rep(plot*ecosystem);
lsmeans ecosystem depth ecosystem*depth /diff;
run:
proc mixed data = soil method=type3;
```

```
94
```

class ecosystem depth rep plot; model Fe = ecosystem depth ecosystem \* depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model Mn = ecosystem depth ecosystem\*depth /ddfm=kr;random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model Zn = ecosystem depth ecosystem\*depth /ddfm=kr;random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model Cu = ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model OM = ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model TotN = ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model TotC = ecosystem depth ecosystem\*depth /ddfm=kr; random plot(ecosystem)rep(plot\*ecosystem); lsmeans ecosystem depth ecosystem\*depth /diff; run: **proc mixed** data = soil method=type3; class ecosystem depth rep plot; model CN = ecosystem depth ecosystem\*depth /ddfm=kr;

random plot(ecosystem)rep(plot\*ecosystem) ;

lsmeans ecosystem depth ecosystem\*depth /diff;

run;

ods rtf close; quit;

# SAS Input File for CO<sub>2</sub> Flux, soil temperature, soil moisture for each year

```
Title 'Pine Tree Soil Respiration Study - 2013'; options ls = 110 ps = 68;
```

data soil; infile '2013flux.csv' firstobs = 2 delimiter = "," truncover LRECL = 600; input month \$ treatment \$ rep vwc temp2cm temp10cm flux; run; **proc print** data = soil; **run**; **proc sort** data=soil; by treatment rep month; run: **proc means** data=soil noprint; by treatment rep month; var vwc temp2cm temp10cm flux; output out=new mean=mvwc mtemp2cm mtemp10cm mflux; run; **proc print** data=new; run; ods rtf file='2013fluxes2.rtf' bodytitle style=journal; **proc mixed** data = new method=type3; class treatment month rep; model mvwc = treatment month treatment\*month /ddfm=kr; random rep(treatment); lsmeans treatment month treatment\*month/ diff; run: **proc mixed** data = new method=type3; class treatment month rep; model mtemp2cm = treatment month treatment\*month /ddfm=kr; random rep(treatment); lsmeans treatment month treatment\*month/ diff; run: **proc mixed** data = new method=type3; class treatment month rep; model mtemp10cm = treatment month treatment\*month /ddfm=kr; random rep(treatment); lsmeans treatment month treatment\*month/ diff: run; **proc mixed** data = new method=type3; class treatment month rep; model mflux = treatment month treatment\*month /ddfm=kr;

random rep(treatment); lsmeans treatment month treatment\*month/ diff; **run**; ods rtf close; **quit**;

## SAS Input File for Annual CO<sub>2</sub> Emissions for 2012 and 2013

Title 'Pine Tree Soil Respiration Study - 2012'; options ls = 110 ps = 68;

data soil; infile '1213CO2emissions.csv' firstobs = 2 delimiter = "," truncover LRECL = 600; input ecosystem \$ rep year emissions; run;

proc print data = soil; run;

```
proc mixed data = soil method=type3;
class ecosystem rep year ;
model emissions= ecosystem year ecosystem*year /ddfm=kr;
random rep(ecosystem) ;
lsmeans ecosystem year ecosystem*year /diff;
run;
```

# SAS Input File for Growing Season Microbial Biomass. Microbial Biomass Carbon (TOC), Microbial Biomass Nitrogen (TN), and Microbial Biomass C:N ration (CN).

```
Title 'Pine Tree Soil Respiration Study - 2012'; options ls = 110 ps = 68;
```

data soil; infile 'MBsas.csv' firstobs = 2 delimiter = "," truncover LRECL = 600; input month \$ treatment \$ rep TOC TN CN; run;

proc print data = soil; run; proc sort data=soil; by treatment rep month; run; proc means data=soil noprint; by treatment rep month; var TOC TN CN; output out=new mean=mTOC mTN mCN; run; proc print data=new; run; ods rtf file='2013micro.rtf' bodytitle style=journal;

```
proc mixed data = new method=type3;
class treatment month rep;
model mTOC = treatment month treatment*month /ddfm=kr;
random rep(treatment);
lsmeans treatment month treatment*month/ diff;
run;
proc mixed data = new method=type3;
class treatment month rep;
model mTN = treatment month treatment*month /ddfm=kr;
random rep(treatment);
lsmeans treatment month treatment*month/ diff;
run:
proc mixed data = new method=type3;
class treatment month rep;
model mCN = treatment month treatment*month /ddfm=kr;
random rep(treatment);
lsmeans treatment month treatment*month/ diff;
run;
ods rtf close;
quit;
```

## SAS Input File for Row Spacing in the Agroecoystem

```
Title 'Pine Tree Soil Respiration Study - 2013'; options ls = 110 ps = 68;
```

```
data soil;
infile 'spacing.csv' firstobs = 2 delimiter = "," truncover LRECL = 600;
input month $ space $ rep vwc temp2cm temp10cm flux;
run;
```

```
proc print data = soil; run;
proc sort data=soil; by space rep month;
run;
proc means data=soil noprint; by space rep month;
var vwc temp2cm temp10cm flux;
output out=new mean=mvwc mtemp2cm mtemp10cm mflux;
run;
proc print data=new;
run;
ods rtf file='2012spacing.rtf' bodytitle style=journal;
proc mixed data = new method=type3;
class space month rep;
model mvwc = space month space*month /ddfm=kr;
random rep(month);
lsmeans space month space*month/ diff;
```
## run;

```
proc mixed data = new method=type3;
class space month rep;
model mtemp2cm = space month space*month /ddfm=kr;
random rep(month);
lsmeans space month space*month/ diff;
run;
proc mixed data = new method=type3;
class space month rep;
model mtemp10cm = space month space*month /ddfm=kr;
random rep(month);
lsmeans space month space*month/ diff;
run:
proc mixed data = new method=type3;
class space month rep;
model mflux = space month space*month /ddfm=kr;
random rep(month);
lsmeans space month space*month/ diff;
run;
ods rtf close;
quit;
```

**Appendix II** 

This appendix contains the input files for the SAS programs in appendix I.

Ecosystem(Eco), Plot (Plot), Repetition within plot (Rep), Depth: 0-10cm (10) and 10-20cm (20), for Bulk Density (BD), Sand, Silt, Clay, pH, Electrical Conductivity (EC), Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), Sulflur (Su), Sodium (Na), Iron (Fe), Manganese (Mn), Zinc (Zn), Copper (Cu), Organic Matter (OM), Total Nitrogen (TotN), Total Carbon (TotC, and C: N ratio (C:N).

Eco	plot	rep	depth	BD	sand	silt	clay	pН	EC	Р	Κ	Ca	Mg
С	1	1	10	1.3	0.1	0.75	0.15	6.97	0.06	41	103	1900	201
С	1	1	20	1.4	0.08	0.73	0.19	6.1	0.05	7.4	41.1	1212	188
С	1	2	10	1.2	0.07	0.77	0.16	6.94	0.05	31	88.4	1861	235
С	1	2	20	1.4	0.15	0.74	0.11	7.32	0.06	15	49.4	2004	232
С	1	3	10	1.3	0.16	0.76	0.08	7.15	0.07	28	136	2145	202
С	1	3	20	1.5	0.15	0.72	0.13	6.14	0.09	11	63.1	1721	196
С	2	1	10	1.3	0.08	0.76	0.15	7	0.08	41	151	1748	182
С	2	1	20	1.5	0.09	0.74	0.17	6.52	0.06	9.5	55.9	1425	189
С	2	2	10	1.3	0.07	0.76	0.18	6.53	0.04	29	144	1434	196
С	2	2	20	1.4	0.15	0.69	0.16	5.81	0.04	5.2	59.5	1474	233
С	2	3	10	1.4	0.16	0.73	0.11	6.76	0.08	28	150	1865	202
С	2	3	20	1.5	0.13	0.7	0.17	5.59	0.07	7.8	69.5	1344	194
С	3	1	10	1.3	0.09	0.76	0.16	6.97	0.05	62	180	1808	204
С	3	1	20	1.4	0.09	0.74	0.17	6.84	0.05	12	67.4	1598	204
С	3	2	10	1.3	0.08	0.65	0.27	7.32	0.05	50	194	2050	195
С	3	2	20	1.4	0.17	0.69	0.14	7	0.04	13	66.6	1409	188
С	3	3	10	1.4	0.18	0.7	0.12	6.92	0.07	137	153	2094	201
С	3	3	20	1.4	0.15	0.62	0.24	4.21	0.05	14	55.9	1681	216
S	1	1	10	1.3	0.04	0.81	0.15	7.51	0.1	39	58.7	2348	281
S	1	1	20	1.4	0.05	0.78	0.17	7.48	0.06	7.6	31.9	1877	323
S	1	2	10	1.3	0.06	0.77	0.18	7.52	0.07	25	65.8	2019	263
S	1	2	20	1.4	0.13	0.74	0.13	7.37	0.04	5.2	43.8	1956	400
S	1	3	10	1.3	0.1	0.77	0.13	7.51	0.07	35	88	2027	307
S	1	3	20	1.5	0.1	0.76	0.14	6.75	0.06	8.1	54.2	1998	379
S	2	1	10	1.4	0.1	0.76	0.15	5.5	0.06	19	143	1613	192
S	2	1	20	1.4	0.07	0.69	0.24	7.04	0.08	8	53.1	1377	181
S	2	2	10	1.4	0.09	0.75	0.17	5.68	0.07	22	105	1674	192
S	2	2	20	1.4	0.14	0.7	0.16	7.14	0.07	8.1	63.9	1417	224
S	2	3	10	1.4	0.18	0.73	0.09	6.4	0.11	46	99.9	2174	175
S	2	3	20	1.5	0.17	0.71	0.12	6.5	0.09	11	64	1622	205
S	3	1	10	1.3	0.07	0.75	0.18	7.48	0.08	23	105	1755	300
S	3	1	20	1.5	0.07	0.68	0.25	6.65	0.07	5.3	59.7	1611	435

S	3	2	10	1.3	0.08	0.75	0.17	7.44	0.07	23	79.3	2191	307
S	3	2	20	1.4	0.18	0.68	0.14	6.65	0.05	10	58.7	1827	472
S	3	3	10	1.4	0.16	0.73	0.12	7.34	0.11	28	97.4	2251	278
S	3	3	20	1.4	0.17	0.67	0.16	6.72	0.08	8.9	65.1	1983	403
W	1	1	10	1.2	0.09	0.75	0.16	7.48	0.05	15	60.9	1731	324
W	1	1	20	1.4	0.09	0.76	0.15	7.8	0.04	3.4	32.1	1578	320
W	1	2	10	1.2	0.09	0.76	0.16	7.37	0.06	20	63.9	1947	371
W	1	2	20	1.4	0.18	0.7	0.12	7.65	0.05	8.3	41.7	2023	423
W	1	3	10	1.4	0.16	0.71	0.13	6.75	0.08	22	102	1902	401
W	1	3	20	1.4	0.13	0.69	0.18	6.42	0.07	8.5	75.1	1709	420
W	2	1	10	1.3	0.08	0.73	0.19	7.04	0.05	16	91.3	1464	394
W	2	1	20	1.4	0.09	0.74	0.17	6.54	0	9.5	46.3	1165	410
W	2	2	10	1.3	0.08	0.74	0.19	7.14	0.05	18	93	1715	443
W	2	2	20	1.4	0.15	0.67	0.18	7.3	0.04	4.6	70	1793	540
W	2	3	10	1.4	0.15	0.73	0.12	6.5	0.08	30	96.9	1677	380
W	2	3	20	1.5	0.12	0.72	0.16	7.07	0.06	13	65.5	1960	456
W	3	1	10	1.3	0.1	0.66	0.24	6.65	0.05	20	91.6	1339	222
W	3	1	20	1.4	0.07	0.71	0.22	6.58	0.05	8.6	47.8	1521	201
W	3	2	10	1.3	0.09	0.75	0.16	6.65	0.05	24	102	1588	255
W	3	2	20	1.4	0.18	0.7	0.12	6.85	0.05	9.5	45.6	1553	214
W	3	3	10	1.5	0.17	0.7	0.13	6.72	0.09	29	120	1819	387
W	3	3	20	1.5	0.15	0.71	0.14	6.96	0.08	14	82.5	1999	420
Eco	plot	rep	depth	Su	Na	Fe	Mn	Zn	Cu	OM	TotN	TotC	CN
С	1	1	10	10	32.3	229	379	1.2	1.7	29	1203	12.5	10.4
С	1	1	20	17	41.8	156	417	0.5	1.2	22	821	8.01	9.75
С	1	2	10	8.7	26.9	281	268	1.5	1.4	29	1272	13.2	10.4
С	1	2	20	8.7	40.8	282	299	1	1.6	25	1020	10.7	10.4
С	1	3	10	14	22.1	355	383	1.2	1.8	34	1383	14.2	10.3
С	1	3	20	43	54.4	194	431	0.7	1.6	24	1008	9.14	9.07
С	2	1	10	14	20.3	162	470	1.3	1.5	32	1426	14.7	10.3
С	2	1	20	13	33.5	145	364	0.6	1.3	23	934	8.7	9.32
С	2	2	10	12	19.6	147	388	0.9	1.4	32	1417	13	9.17
С	2	2	20	33	34.2	112	282	0.2	1.3	23	898	6.67	7.43
С	2	3	10	15	24.9	150	416	1.1	1.6	33	1350	13.2	9.77
С	2	3	20	54	48.7	136	336	0.3	1.3	27	1045	8.27	7.91
С	3	1	10	8.1	13.9	160	406	2.1	1.6	32	1356	14.1	10.4
С	3	1	20	14	26.6	142	428	1.3	1.7	25	973	9.28	9.54
С	3	2	10	8.3	15.7	179	481	1.6	1.7	38	1346	14.5	10.8
С	3	2	20	18	26.1	132	372	0.4	1.1	22	863	7.61	8.82

С	3	3	10	17	26.5	223	424	2.2	1.6	32	1358	14.5	10.7
С	3	3	20	14	40.8	114	308	0.5	1.3	19	737	6.16	8.37
S	1	1	10	8.3	26	382	189	1.3	1.6	27	1175	13.2	11.2
S	1	1	20	5.1	35.8	321	204	0.9	1.6	19	785	8.88	11.3
S	1	2	10	7.2	21.6	283	230	1.5	1.7	28	1161	14.4	12.4
S	1	2	20	5.2	45.3	181	288	1	1.8	18	594	6.81	11.5
S	1	3	10	11	24.3	355	205	1.7	1.8	28	1124	13.4	11.9
S	1	3	20	6.4	46.8	229	289	0.7	1.8	18	569	7.11	12.5
S	2	1	10	9.8	23.6	139	444	1	1.4	33	1355	14.9	11
S	2	1	20	50	39.4	124	268	0.4	1.1	28	955	8.41	8.8
S	2	2	10	9.6	26	151	491	1	1.2	35	1431	14.7	10.3
S	2	2	20	26	37.7	129	298	0.6	1.2	26	985	8.88	9.02
S	2	3	10	16	25.9	183	511	1.3	1.4	37	1519	18.2	12
S	2	3	20	14	34.5	164	556	0.8	1.3	29	977	9.54	9.77
S	3	1	10	7.5	20.6	157	498	0.9	1.6	34	1359	14.1	10.4
S	3	1	20	9.8	51	112	558	0.4	1.4	22	782	6.28	8.04
S	3	2	10	8.3	29.5	194	533	1.3	1.8	31	1222	13.3	10.9
S	3	2	20	7.4	60.2	130	640	0.6	1.4	25	886	7.88	8.89
S	3	3	10	13	24.6	206	553	1.5	2	37	1434	16.4	11.4
S	3	3	20	8.1	50.5	122	682	0.6	1.6	25	910	7.75	8.52
W	1	1	10	7.3	25.1	219	391	2.1	1.6	28	1171	12.8	10.9
W	1	1	20	4.8	53.5	148	552	1.5	1.4	16	478	4.92	10.3
W	1	2	10	8.8	34.5	258	465	2.7	1.8	32	1214	13.1	10.8
W	1	2	20	8.2	68.9	184	605	2.3	1.8	23	819	9.54	11.7
W	1	3	10	15	31.3	207	579	1.9	1.9	34	1326	14.5	10.9
W	1	3	20	22	60.2	122	328	0.5	1.5	49	787	7.85	9.97
W	2	1	10	8.2	21.3	152	550	1.3	1.6	30	1208	12.3	10.2
W	2	1	20	29	50.2	150	562	0.7	1.2	21	786	6.6	8.39
W	2	2	10	11	30.9	181	580	1.7	1.9	32	1279	13.4	10.5
W	2	2	20	5.8	78.2	113	358	0.5	1.6	20	714	5.25	7.35
W	2	3	10	15	25.2	286	422	2.6	2	33	1380	14.7	10.6
W	2	3	20	11	56.6	195	468	1	1.6	23	739	8.07	10.9
W	3	1	10	11	20.9	172	465	1.5	1.5	29	1239	12.9	10.4
W	3	1	20	11	43.7	136	347	0.5	1.3	19	710	6.59	9.28
W	3	2	10	13	26.3	196	521	2	1.7	33	1468	14.4	9.8
W	3	2	20	8.8	41.4	138	582	1.1	1.5	21	799	7	8.77
W	3	3	10	15	29	235	514	2.4	2.2	33	1281	13.9	10.8
W	3	3	20	14	52.1	196	591	2.4	2.1	26	952	9.88	10.4

The 2012 Monthly Data Set for Soil Respiration. Treatment (Treat), Replication (Rep), 0-6cm Volumetric water content (Vwc, cm<sup>3</sup> cm<sup>-3</sup>), 2cm soil temperature, (Temp2cm, °C), 10cm soil temperature (Temp10cm, °C), and CO<sub>2</sub> flux (Flux, μmol m<sup>-2</sup> s<sup>-1</sup>) Month

(2012)	Treat	Rep	Vwc	Temp2cm	Temp10cm	Flux
jan	С	1	0.462	2.1	3.19	0.968
jan	С	2	0.39	1	3.2	0.389
jan	С	2	0.385	1.5	3.36	0.353
jan	С	2	0.281	1	2.64	0.385
jan	С	3	0.284	1	3.24	0.465
jan	С	3	0.356	0.9	2.36	0.111
jan	С	3	0.226	1.1	2.44	0.356
jan	S	1	0.46	4.2	3.85	-0.0164
jan	S	1	0.453	3.4	3.73	-1.69
jan	S	1	0.446	1.6	3.79	0.36
jan	S	2	0.401	1.6	3.08	0.185
jan	S	2	0.401	2.3	3.86	0.254
jan	S	2	0.445	2.5	3.17	0.138
jan	S	3	0.457	1.1	3.33	-1.09
jan	S	3	0.445	2.5	3.52	-1.09
jan	S	3	0.434	1.5	3.425	0.453
jan	W	1	0.433	3.2	3.72	0.191
jan	W	1	0.426	3.5	3.74	0.3
jan	W	1	0.43	2	3.16	0.167
jan	W	2	0.437	2.2	3.15	0.472
jan	W	2	0.383	4.7	3.25	0.284
jan	W	2	0.457	4.6	3.17	0.343
jan	W	3	0.402	1.9	3.42	-0.349
jan	W	3	0.367	1.7	2.92	0.349
jan	W	3	0.417	2.9	3.33	0.25
feb	С	1	0.479	10.3	7.98	0.37
feb	С	1	0.479	10.7	7.37	0.397
feb	С	1	0.418	11.4	7.58	0.63
feb	С	2	0.372	10.9	6.93	1.73
feb	С	2	0.367	11	7.36	2.49
feb	С	2	0.374	10.9	7.79	1.17
feb	С	3	0.326	12.7	8.46	0.632
feb	С	3	0.354	12	8.27	1.81
feb	С	3	0.35	12.3	8.38	4.93
feb	S	1	0.417	12.7	9.33	0.421
feb	S	1	0.415	12.2	9.06	0.206
feb	S	1	0.406	13.5	10.16	0.744

feb	S	2	0.409	7.8	7.51	0.408
feb	S	2	0.407	10.6	7.75	0.36
feb	S	2	0.389	9.1	7.57	0.625
feb	S	3	0.404	12.1	8.48	0.458
feb	S	3	0.428	11.8	8.8	0.416
feb	S	3	0.432	15	9.36	1.72
feb	W	1	0.46	10.9	8.82	0.418
feb	W	1	0.44	12.3	8.36	0.43
feb	W	1	0.42	12.2	7.91	0.438
feb	W	2	0.406	12.8	9.13	0.564
feb	W	2	0.407	15.2	9.42	0.387
feb	W	2	0.419	11.3	9	1.15
feb	W	3	0.444	15	9.09	0.444
feb	W	3	0.68	12.9	9.15	0.68
feb	W	3	0.34	13.2	8.9	0.34
mar	С	1	0.449	22.7	19.5	1.82
mar	С	1	0.436	22	19.22	0.593
mar	С	1	0.462	21.9	19.36	0.327
mar	С	1	0.463	22	19.29	0.398
mar	С	1	0.453	22	0	0
mar	С	2	0.379	21	18.64	2.94
mar	С	2	0.381	21.4	18.95	1.92
mar	С	2	0.401	21.3	19.38	1.91
mar	С	2	0.378	21.1	18.91	0.945
mar	С	2	0.389	22	19.27	1.61
mar	С	3	0.323	24.1	19.42	9.19
mar	С	3	0.367	23.1	19.26	6.82
mar	С	3	0.432	22.9	19.47	0.0863
mar	С	3	0.296	23.5	19.62	2.84
mar	С	3	0.378	23.7	19.04	2.21
mar	S	1	0.414	24.8	21.04	2.08
mar	S	1	0.38	25.6	21.1	2.39
mar	S	1	0.407	26.1	20.42	0.398
mar	S	1	0.411	24.3	20.25	0.481
mar	S	1	0.419	29.1	29.28	0.553
mar	S	2	0.396	19.8	18.71	1.65
mar	S	2	0.412	20.4	18.41	2.19
mar	S	2	0.405	20.7	18.73	1.19
mar	S	2	0.435	20.9	18.85	0.611
mar	S	2	0.429	21.1	19.05	1.33
mar	S	3	0.406	24.4	20.39	0.799
mar	S	3	0.405	25.7	20.57	2.31

mar	S	3	0.397	25.6	21.07	0.78
mar	S	3	0.344	29.6	21.01	3.84
mar	S	3	0.396	28.8	20.84	5.77
mar	W	1	0.456	22.8	19.45	1.02
mar	W	1	0.449	23.2	19.1	0.944
mar	W	1	0.452	22.1	19.1	-1.12
mar	W	1	0.457	21.8	19.21	1.28
mar	W	1	0.459	22.4	19.41	1.4
mar	W	2	0.429	24.4	19.74	1.27
mar	W	2	0.421	23.3	20.57	2.22
mar	W	2	0.453	26.8	20.06	2.6
mar	W	2	0.38	26.3	20.13	1.93
mar	W	2	0.443	26.6	20.11	1.79
mar	W	3	0.431	21.2	19.59	0.828
mar	W	3	0.433	22.3	20.5	1.45
mar	W	3	0.447	24.7	19.93	0.635
mar	W	3	0.424	24.1	19.34	2.33
mar	W	3	0.424	22.4	20.08	1.59
apr	С	1	0.278	17.8	15.14	2.67
apr	С	1	0.285	20.9	16.62	1.95
apr	С	1	0.25	17.7	14.9	1.43
apr	С	1	0.295	17.8	15.98	2.09
apr	С	1	0.255	18.7	15.54	1.9
apr	С	2	0.281	16.7	15.01	1.22
apr	С	2	0.265	18	14.78	1.12
apr	С	2	0.256	17.8	14.34	1.63
apr	С	2	0.246	16	14.63	1.82
apr	С	2	0.271	16.8	14.95	1.47
apr	С	3	0.24	21.2	16.45	1.59
apr	С	3	0.274	19.4	15.66	1.8
apr	С	3	0.281	20	15.99	1.91
apr	С	3	0.258	19.7	16.02	2.17
apr	С	3	0.286	21.2	17.06	1.84
apr	S	1	0.315	19.1	16.32	2.64
apr	S	1	0.296	21.6	17.29	2.72
apr	S	1	0.307	22.5	17.66	3.2
apr	S	1	0.346	21.8	17.99	3.06
apr	S	1	0.311	23	18.13	2.47
apr	S	2	0.367	13.3	13.75	1.82
apr	S	2	0.362	15.4	13.7	1.6
apr	S	2	0.6	13.8	14.01	1.55
apr	S	2	0.355	13.5	13.72	1.63

apr	S	2	0.345	14.3	13.39	1.96
apr	S	3	0.351	25.3	17.47	1.98
apr	Ŝ	3	0.33	21	16.78	2.73
apr	S	3	0.332	19.7	17.15	1.96
apr	S	3	0.337	21.9	17.06	3.05
apr	S	3	0.329	22.2	16.98	2.81
apr	W	1	0.266	18.7	14.68	2.99
apr	W	1	0.279	17.2	14.73	2.29
apr	W	1	0.316	15.9	14.12	1.51
apr	W	1	0.291	15.3	14.21	2.52
apr	W	1	0.267	16.3	15.44	1.14
apr	W	2	0.298	24.3	17.56	2.46
apr	W	2	0.336	16.6	14.54	2.22
apr	W	2	0.209	20.3	15.49	2.63
apr	W	2	0.299	16.5	15.01	1.59
apr	W	2	0.289	19.1	15.57	1.82
apr	W	3	0.227	15.8	14.74	2.19
apr	W	3	0.324	15.1	14.9	1.63
apr	W	3	0.26	18.6	15.26	2.47
apr	W	3	0.291	15.6	14.52	1.98
apr	W	3	0.267	17.3	15.31	4.19
may	С	1	0.214	34	26.3	4.39
may	С	1	0.19	32.6	26.79	3.17
may	С	1	0.187	36.6	28.09	3.55
may	С	1	0.199	33.3	27.46	4.03
may	С	1	0.246	33.1	26.82	3.67
may	С	2	0.064	35.8	27.82	2.48
may	С	2	0.112	33.5	27.86	6.49
may	С	2	0.157	31.5	27.54	4.23
may	С	2	0.163	35.7	27.1	6.04
may	С	2	0.153	34.4	29.2	3.46
may	С	3	0.188	32.6	25.83	3.18
may	С	3	0.191	31.1	25.24	2.91
may	С	3	0.12	34.4	25.99	2.31
may	С	3	0.161	33.2	25.45	6.13
may	С	3	0.118	37.7	27.3	3.65
may	S	1	0.252	22.9	22.16	4.01
may	S	1	0.213	31.6	24.15	4.88
may	S	1	0.227	29.4	24.77	6.21
may	S	1	0.217	26.6	23.12	5.03
may	S	1	0.249	31.1	24.27	5.39
may	S	2	0.178	21.5	20.04	6.06

may	S	2	0.185	25.7	21.14	3.35
may	S	2	0.154	21.6	20.17	1.95
may	S	2	0.165	20.6	19.69	5.08
may	S	2	0.196	21.4	19.87	2.03
may	S	3	0.21	21.1	21.72	1.83
may	S	3	0.174	28.6	23.3	2.56
may	S	3	0.166	26.7	23.38	2.58
may	S	3	0.192	33.4	25.06	3.57
may	S	3	0.194	28.3	22.22	4.29
may	W	1	0.135	25.1	22.09	2
may	W	1	0.142	25	20.71	2.33
may	W	1	0.122	26.5	20.81	2.16
may	W	1	0.143	23.1	20.58	1.99
may	W	1	0.136	31.1	22.34	1.81
may	W	2	0.148	33.2	24.71	3.84
may	W	2	0.127	25.4	21.49	2.93
may	W	2	0.147	34.1	24.49	2.18
may	W	2	0.144	25.2	21.65	2.27
may	W	2	0.131	28.7	23.58	3.25
may	W	3	0.113	26.2	22.74	1.86
may	W	3	0.189	26	22.08	4.76
may	W	3	0.151	23.2	21.36	2.64
may	W	3	0.127	28.3	22.88	1.56
may	W	3	0.148	23.8	21.82	2.32
jun	С	1	0.034	41		3.58
jun	С	1	0.055	43.4	39.72	1.56
jun	С	1	0.063	45	36.96	3.22
jun	С	2	0.19	47.5	36.73	2.85
jun	С	2	0.045	46.2		4.96
jun	С	2	0.028	48.9	38.4	1.87
jun	С	3	0.032	43.7		2.52
jun	С	3	0.068	46.8		3.52
jun	С	3	0.068	42.9		2.12
jun	S	1	0.124	37.9		3.47
jun	S	1	0.138	36.8	29.55	3.67
jun	S	1	0.102	39.2	32.29	3.25
jun	S	2	0.07	45.2	32.91	2.68
jun	S	2	0.12	43	34.42	1.93
jun	S	2	0.091	39.7	37.23	4.65
jun	S	3	0.124	36.9	29.99	3.12
jun	S	3	0.138	39.2	40.46	3.97
jun	S	3	0.102	40.2	46.85	2.65

jun	W	1	0.088	41.7	34.15	1.34
jun	W	1	0.102	38.8		1.81
jun	W	1	0.113	41.8		1.28
jun	W	2	0.115	37.5	31.55	1.84
jun	W	2	0.1	44.1	36.52	2.12
jun	W	2	0.106	35.3	38.36	1.92
jun	W	3	0.074	35.4	30.45	1.26
jun	W	3	0.091	38.8	32.09	2.25
jun	W	3	0.097	34	30.72	2.07
jul	С	1	0.316	33.5	39.36	9.99
jul	С	1	0.284	34	30.92	9.15
jul	С	1	0.25	34	31.52	10.6
jul	С	1	0.194	32.5	29.84	8.85
jul	С	1	0.318	37.8	31.17	8.31
jul	С	2	0.211	31.8	29.08	4.41
jul	С	2	0.08	39.2	32.99	5.15
jul	С	2	0.133	31.9	30.03	5.43
jul	С	2	0.127	32.5	30.69	7.28
jul	С	2	0.07	39.6	33.1	4.01
jul	С	3	0.173	34.1	29.35	11.8
jul	С	3	0.173	31.8	28.9	10.7
jul	С	3	0.072	42.3	31.86	4.99
jul	С	3	0.292	33.2	29.62	10.6
jul	С	3	0.267	34.2	29.78	9.86
jul	S	1	0.292	29.5	28.3	8.33
jul	S	1	0.42	30.3	28.88	2.35
jul	S	1	0.236	28.2	27.59	8.29
jul	S	1	0.346	31.6	29.3	9.64
jul	S	1	0.337	29.9	27.3	7.3
jul	S	2	0.217	34.1	29.58	5.73
jul	S	2	0.218	33.6	29.94	7.39
jul	S	2	0.204	31.6	29.72	5.78
jul	S	2	0.218	33.5	29.54	8.66
jul	S	2	0.161	34.8	30.71	8.48
jul	S	3	0.28	31.9	28.11	7.18
jul	S	3	0.245	31.7	29.74	6.25
jul	S	3	0.19	30.2	28.92	9.49
jul	S	3	0.263	34.2	28.58	8.4
jul	S	3	0.315	32.5	30.26	10.5
jul	W	1	0.345	37.4	30.5	4.33
jul	W	1	0.277	34.8	31.37	5.7
jul	W	1	0.273	33.3	31.02	6.15

jul	W	1	0.281	39.4	33.06	5.39
jul	W	1	0.293	31.6	29.98	5.34
jul	W	2	0.261	33.18	33.26	6.27
jul	W	2	0.175	33.67	33.78	6.52
jul	W	2	0.204	31.54	31.48	6.66
jul	W	2	0.163	32.11	32.1	4.99
jul	W	2	0.225	33.96	33.94	8.64
jul	W	3	0.247	37	31.13	4.1
jul	W	3	0.215	36.2	30.82	5.86
jul	W	3	0.232	34.2	30.24	8.67
jul	W	3	0.258	32.6	31.5	6.47
jul	W	3	0.217	34.4	31.07	6.28
aug	С	1	0.022	35.3	31.4	4.33
aug	С	1	0.113	34.1	31.02	4.7
aug	С	1	0.071	35.5	29.04	4.19
aug	С	1	0.112	33.7	28.81	4.73
aug	С	1	0.097	36	32.4	3.35
aug	С	2	0.031	44	40.43	3.53
aug	С	2	0.036	41.5	31.89	2.84
aug	С	2	0.044	38	31.65	2.49
aug	С	2	0.078	39.2	33.17	2.13
aug	С	2	0.023	35.2	30.46	1.94
aug	С	3	0.092	33.7	29	4.49
aug	С	3	0.05	39.2	31.31	3.17
aug	С	3	0.094	33.5	30.91	4.06
aug	С	3	0.06	34.8	28.34	3.61
aug	С	3	0.075	34.6	29.71	3.76
aug	S	1	0.075	38.7	25.3	1.91
aug	S	1	0.164	26.8	25.28	4.43
aug	S	1	0.268	23.4	24.73	1.95
aug	S	1	0.115	28.9	27.37	3.87
aug	S	1	0.156	27.7	25.85	3.13
aug	S	2	0.103	34.9	30.89	12.8
aug	S	2	0.076	33.9	29.72	2.2
aug	S	2	0.088	34.7	28.46	2.75
aug	S	2	0.068	38.1	33.49	2.01
aug	S	2	0.098	41.7	35.05	2.28
aug	S	3	0.05	30.5	26.4	2.98
aug	S	3	0.124	29.4	28.49	6.16
aug	S	3	0.058	30	26.46	5.26
aug	S	3	0.071	34.1	28.26	3.24
aug	S	3	0.139	31.8	29.41	4.22

aug	W	1	0.093	37.3	33.14	1.07
aug	W	1	0.089	31.4	29.5	2.56
aug	W	1	0.12	33.7	29.36	1.81
aug	W	1	0.102	33	30.11	2.2
aug	W	1	0.136	32.3	31.24	1.96
aug	W	2	0.081	34.5	34.05	1.37
aug	W	2	0.092	34.9	32.68	2.4
aug	W	2	0.131	39.1	35.35	2.14
aug	W	2	0.107	33.7	32.97	1.57
aug	W	2	0.112	33.2	32.62	2.93
aug	W	3	0.073	33.6	30.17	1.81
aug	W	3	0.103	36.3	32.64	1.38
aug	W	3	0.086	33.2	30.64	2.09
aug	W	3	0.105	31.7	30.59	1.86
aug	W	3	0.116	33.2	29.12	2.44
sep	С	1	1.88	24.9	23.36	3.17
sep	С	1	0.743	26.1	23.88	4.2
sep	С	1	0.201	26.2	23.66	3.41
sep	С	1	0.148	25.8	24.28	4.9
sep	С	2	0.076	27.9	25.71	4.57
sep	С	2	0.132	26.1	24.8	1.19
sep	С	2	0.118	24.66	25.6	2.52
sep	С	3	0.094	23.8	23.45	4.12
sep	С	3	0.169	23.8	23.64	3.27
sep	С	3	0.085	23.8	24.3	2.66
sep	S	1	0.304	24.5	23.19	6.08
sep	S	1	0.326	27.6	24.25	4.95
sep	S	1	0.253	25.9	23.59	3.88
sep	S	1	0.287	26	23.58	3.7
sep	S	1	0.29	24.4	23.35	3.64
sep	S	2	0.212	23.8	23.8	3.34
sep	S	2	0.255	25.1	24.59	4.59
sep	S	2	0.238	24.7	24.01	5.16
sep	S	3	0.216	23.2	22.76	2.97
sep	S	3	0.157	23.2	22.69	3.61
sep	S	3	0.246	23.6	22.9	5.18
sep	W	1	0.144	27.6	25	2.59
sep	W	1	0.144	27.7	24.74	3.54
sep	W	1	0.183	26.6	24.55	3.34
sep	W	1	0.169	28.4	25.32	3.3
sep	W	1	0.137	24.6	25	3.65
sep	W	2	0.097	26.6	26.12	2.31

sep	W	2	0.12	25.6	25.9	2.69
sep	W	2	0.143	25.4	24.91	2.18
sep	W	3	0.153	25.3	24.89	2.7
sep	W	3	0.127	25.4	25.8	3.03
sep	W	3	0.154	25.7	25.12	2.48
sep	W	3	0.131	25	25.2	3.1
sep	W	3	0.131	24.3	24.53	1.82
oct	С	1	0.382	23.1	17.9	2.04
oct	С	1	0.306	23.1	18.4	2.25
oct	С	1	0.376	21.6	18.8	2.75
oct	С	1	0.356	23.1	19.2	1.56
oct	С	1	0.325	21.2	18.6	1.68
oct	С	2	0.201	23.1	19.4	3.22
oct	С	2	0.223	23.2	19.4	2.8
oct	С	2	0.235	23.3	19.7	3.04
oct	С	2	0.294	23.4	18.9	1.95
oct	С	2	0.254	23	19.3	3.41
oct	С	3	0.3	22.1	18.1	1.86
oct	С	3	0.323	22.6	18.4	2.78
oct	С	3	0.342	21.3	17.9	2.39
oct	С	3	0.348	20.3	17.7	1.68
oct	С	3	0.241	24.4	18.6	2.84
oct	S	1	0.423	16	16.3	1.79
oct	S	1	0.42	16.1	16.3	1.17
oct	S	1	0.432	16.1	16.05	0.675
oct	S	1	0.346	16.3	16.6	1.56
oct	S	1	0.33	16.7	16.7	1.72
oct	S	2	0.233	19.1	17.7	1.53
oct	S	2	0.319	18.9	17.6	2.69
oct	S	2	0.286	18.8	17.4	4.89
oct	S	2	0.313	17.8	17.2	1.9
oct	S	2	0.132	17.9	17.2	3.47
oct	S	3	0.353	15.8	16	2.81
oct	S	3	0.377	15.8	15.9	1.03
oct	S	3	0.355	15.8	15.9	1.46
oct	S	3	0.375	16.1	16.2	2.43
oct	S	3	0.383	16.5	16.3	1.77
oct	W	1	0.294	17.8	17.6	1.49
oct	W	1	0.285	21.7	17.8	2.11
oct	W	1	0.291	18.7	17.4	2.16
oct	W	1	0.296	18.4	17.4	2.28
oct	W	1	0.298	18	17.1	1.98

oct	W	2	0.287	21.2	19.8	2.87
oct	W	2	0.307	21.1	19	2.81
oct	W	2	0.294	21.1	18.5	1.95
oct	W	2	0.264	22.4	19.3	3.19
oct	W	2	0.3	25.3	21.6	2.89
oct	W	3	0.25	19.2	17.8	1.7
oct	W	3	0.333	19.2	17.7	1.44
oct	W	3	0.269	19.2	17.7	2.2
oct	W	3	0.255	18.9	17.8	1.75
oct	W	3	0.32	17.7	17.3	2.11
nov	С	1	0.328	16.2	12	1.16
nov	С	1	0.354	16.2	12	1.31
nov	С	1	0.328	16.2	12	0.959
nov	С	1	0.465	16.2	12	1.26
nov	С	1	0.396	16.2	12	1.21
nov	С	2	0.215	17.6	11.6	1.08
nov	С	2	0.175	15.7	12.3	1.39
nov	С	2	0.322	15.4	12	1.08
nov	С	2	0.199	16.2	12	1.91
nov	С	2	0.168	16.2	12	1.23
nov	С	3	0.286	14	11.3	2.03
nov	С	3	0.317	13.3	11.1	1.22
nov	С	3	0.268	14.2	11	0.766
nov	С	3	0.255	12.9	11.1	1.34
nov	С	3	0.219	13.3	11.3	1.58
nov	S	1	0.326	12.7	9.5	1.41
nov	S	1	0.444	13.1	9.5	0.562
nov	S	1	0.423	12.3	9.6	0.657
nov	S	1	0.413	11.1	9.1	0.568
nov	S	1	0.38	11.1	9.3	4.24
nov	S	2	0.288	14	11.1	1.5
nov	S	2	0.292	12.7	10.6	1.02
nov	S	2	0.278	11.2	10.2	1.52
nov	S	2	0.374	13	11.1	1.43
nov	S	2	0.228	13	11.2	0.43
nov	S	3	0.343	12.3	9.6	0.74
nov	S	3	0.321	11.2	9.6	1.15
nov	S	3	0.306	11.1	9.5	1.14
nov	S	3	0.247	11.7	9.3	1.01
nov	S	3	0.322	11.8	9.8	0.938
nov	W	1	0.267	11.1	10	0.93
nov	W	1	0.273	11.5	10.2	0.696

nov	W	1	0.3	12.4	10.8	0.653
nov	W	1	0.347	12.1	10.7	1.09
nov	W	1	0.256	13.1	10.5	1.08
nov	W	2	0.252	12.1	10	1.05
nov	W	2	0.36	11.7	10.4	1.48
nov	W	2	0.283	11.1	9.6	1.21
nov	W	2	0.316	10.8	9.6	0.926
nov	W	2	0.292	12	9.8	1.18
nov	W	3	0.312	11.7	9.8	1.2
nov	W	3	0.271	12.6	10.3	1.25
nov	W	3	0.354	12.2	10.3	1.32
nov	W	3	0.301	12.7	10.2	1.29
nov	W	3	0.277	11.9	10.8	1.92
dec	С	1	0.413	4.6	5.6	0.595
dec	С	1	0.401	5.3	6	0.515
dec	С	1	0.44	5.4	6	0.776
dec	С	2	0.378	5.3	6	0.594
dec	С	2	0.321	5	6	0.826
dec	С	2	0.367	5.6	6	0.684
dec	С	3	0.361	5.6	6.4	0.357
dec	С	3	0.354	6.9	7.4	0.792
dec	С	3	0.32	6.5	6.6	1.07
dec	S	1	0.385	6.7	6.6	0.203
dec	S	1	0.41	8	7	0.869
dec	S	1	0.412	7.4	7.5	0.397
dec	S	2	0.417	6.4	7	0.364
dec	S	2	0.366	6.6	7.6	0.493
dec	S	2	0.381	5.7	6.4	0.775
dec	S	3	0.385	7.1	7.1	0.42
dec	S	3	0.41	8	7.2	0.641
dec	S	3	0.412	8.1	7.3	0.319
dec	W	1	0.423	7	7.3	0.555
dec	W	1	0.367	7.3	8	0.53
dec	W	1	0.32	7.2	7	0.582
dec	W	2	0.41	7.6	7.2	0.518
dec	W	2	0.384	6.8	7.4	0.959
dec	W	2	0.41	7	7.4	0.953
dec	W	3	0.337	8.1	8.7	-1.61
dec	W	3	0.392	7.3	8	0.705
dec	W	3	0.371	7	7.5	0.412

The 2013 Monthly Data Set for Soil Respiration. Treatment (Treat), Replication (Rep), 0-6cm Volumetric water content (Vwc, cm<sup>3</sup> cm<sup>-3</sup>), 2cm soil temperature, (Temp2cm, °C), 10cm soil temperature (Temp10cm, °C), and CO<sub>2</sub> flux (Flux,  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>)

Month						
(2013)	Treat	Rep	VWC	Temp2cm	Temp10cm	Flux
jan	С	1	0.452	4.8	5.1	0.948
jan	С	1	0.575	5.4	5	0.742
jan	С	1	0.494	5.3	5.3	0.675
jan	С	1	0.55	5	5.2	0.902
jan	С	1	0.466	5.7	5.5	0.386
jan	С	2	0.362	5.3	5	0.562
jan	С	2	0.367	5.5	5.3	0.507
jan	С	2	0.391	5	5	0.802
jan	С	2	0.38	5.3	5.7	0.0998
jan	С	2	0.338	5.4	5.2	0.21
jan	С	3	0.356	5.7	5.2	0.325
jan	С	3	0.41	5.5	5.5	0.475
jan	С	3	0.419	5.3	5.4	0.466
jan	С	3	0.342	5.7	5.4	0.5
jan	С	3	0.284	6	5.7	0.436
jan	S	1	0.452	4.2	5	0.259
jan	S	1	0.473	4.3	5	0.258
jan	S	1	0.522	4.4	5	0.221
jan	S	1	0.401	4.6	5.1	0.299
jan	S	1	0.381	5	5.2	0.302
jan	S	2	0.319	5.4	5	0.292
jan	S	2	0.383	5	5	0.52
jan	S	2	0.345	5	5	0.315
jan	S	2	0.457	5	5	0.466
jan	S	2	0.407	6	5.3	0.404
jan	S	3	0.391	4.5	4.9	0.395
jan	S	3	0.359	4.7	5	0.413
jan	S	3	0.357	4.7	5.4	0.378
jan	S	3	0.347	5.1	5.1	0.362
jan	S	3	0.378	5	5	0.368
jan	W	1	0.402	5	5.1	0.445
jan	W	1	0.426	5.5	5.3	0.455
jan	W	1	0.378	4.6	4.9	0.197
jan	W	1	0.4	4.7	5	0.206
jan	W	1	0.385	5	5.3	0.287
jan	W	2	0.38	4.7	5	0.404

jan	W	2	0.325	5.5	5.3	0.366
jan	W	2	0.291	5	5.1	0.381
jan	W	2	0.314	5	5.3	0.349
jan	W	2	0.412	4.6	4.9	0.49
jan	W	3	0.331	4.6	5	0.295
jan	W	3	0.349	4.7	5.5	0.483
jan	W	3	0.352	4.9	5.3	0.381
jan	W	3	0.427	5.6	5.4	0.349
jan	W	3	0.47	4.8	5.5	0.49
feb	С	1	0.497	4.7	5.6	0.572
feb	С	1	0.501	5	6	0.544
feb	С	1	0.409	4.7	5.7	0.612
feb	С	2	0.388	4.2	5.3	0.614
feb	С	2	0.402	4.2	5.9	0.353
feb	С	2	0.433	4	5	0.671
feb	С	3	0.409	4.7	5.5	0.358
feb	С	3	0.417	5.2	5.5	0.592
feb	S	1	0.371	5	6.2	0.592
feb	S	1	0.495	4.7	5.8	0.428
feb	S	1	0.406	5.2	6	0.277
feb	S	2	0.429	4.9	6	0.287
feb	S	2	0.451	4.6	5.8	0.261
feb	S	2	0.413	4.9	6.4	0.221
feb	S	3	0.454	5	5.9	0.461
feb	S	3	0.346	5.7	6.6	0.446
feb	W	1	0.407	5.5	5.9	0.259
feb	W	1	0.449	5.5	6.5	0.409
feb	W	1	0.407	5.3	6	0.0118
feb	W	2	0.35	5.2	6	0.106
feb	W	2	0.4	5.6	6.5	0.54
feb	W	2	0.47	4.7	6	0.46
feb	W	3	0.424	4.2	4.2	0.417
feb	W	3	0.362	4.2	4.6	0.369
feb	W	3	0.392	5.7	5	0.436
mar	С	1	0.539	11	7.7	1.5
mar	С	1	0.532	10.2	7.1	3.08
mar	С	1	0.532	9.5	7.3	1.06
mar	С	2	0.552	10.3	7.6	1.12
mar	С	2	0.557	9.3	7.6	1.5
mar	С	2	0.554	9.1	6.8	1.65
mar	С	3	0.537	6.4	6.4	2.16
mar	С	3	0.543	7.5	6.2	1.64

mar	С	3	0.543	7.1	6.4	1.07
mar	S	1	0.534	10.5	7.4	0.771
mar	S	1	0.517	8.9	7	0.386
mar	S	1	0.531	10.9	7.5	0.307
mar	S	2	0.431	9.2	7.8	1.74
mar	S	2	0.442	8.1	7.2	0.0166
mar	S	2	0.444	10.6	7	-0.25
mar	S	3	0.395	11.7	8.7	0.852
mar	S	3	0.424	12.2	8.7	1.19
mar	S	3	0.407	11.1	8.1	0.564
mar	W	1	0.437	9.8	7	0.704
mar	W	1	0.429	8.4	7	0.333
mar	W	1	0.443	9	7	0.76
mar	W	2	0.431	7	6	0.256
mar	W	2	0.43	5.1	5.3	-0.0172
mar	W	2	0.443	6	5.7	0.874
mar	W	3	0.4	7	5.6	0.957
mar	W	3	0.395	8.7	5.6	0.675
mar	W	3	0.399	7.3	6.4	0.534
apr	С	1	0.399	24.1	19.1	2.14
apr	С	1	0.392	26	19.4	4.25
apr	С	1	0.336	27	20.2	3.77
apr	С	1	0.392	26	20	5.49
apr	С	1	0.357	25.1	20.1	2.69
apr	С	2	0.241	23.6	19.7	2.17
apr	С	2	0.328	24.7	19.3	2.24
apr	С	2	0.313	22.7	18.7	2.73
apr	С	2	0.347	24.4	20.7	4.72
apr	С	2	0.284	25.1	20.1	3.12
apr	С	3	0.346	23.4	19	3.7
apr	С	3	0.375	23	18.7	7.18
apr	С	3	0.409	23	18.6	2.9
apr	С	3	0.4	23.7	18.9	2.94
apr	С	3	0.263	24	19	7.84
apr	S	1	0.33	18.2	16.4	1.32
apr	S	1	0.357	17.6	16.4	1.13
apr	S	1	0.35	17.5	16.4	0.635
apr	S	1	0.396	19	16.4	2.01
apr	S	1	0.384	18.1	16.4	2.25
apr	S	2	0.336	22.7	19.1	2.02
apr	S	2	0.328	20.6	18.1	2.69
apr	S	2	0.38	22	18.2	1.52

apr	S	2	0.404	22.4	18.5	2.28
apr	S	2	0.354	21	18.6	3.77
apr	S	3	0.399	18.5	17	2.53
apr	S	3	0.356	18.3	16.6	1.71
apr	S	3	0.384	18.4	16.5	1.4
apr	S	3	0.354	19.5	16.9	3.69
apr	S	3	0.363	19.1	16.8	2.53
apr	W	1	0.392	21.5	17.9	2.49
apr	W	1	0.388	19	16.6	2.13
apr	W	1	0.413	17.4	16.1	1.92
apr	W	1	0.4	19	16.1	2.04
apr	W	1	0.376	19.8	16.4	2.37
apr	W	2	0.343	17.7	16.5	1.83
apr	W	2	0.264	18.6	16.4	2.03
apr	W	2	0.343	17.5	16.4	2.47
apr	W	2	0.285	18.5	16.4	6.15
apr	W	2	0.337	18.3	16.2	1.06
apr	W	3	0.284	20	16.8	2.44
apr	W	3	0.358	20.5	17	1.54
apr	W	3	0.435	17.7	16	1.26
apr	W	3	0.337	19.7	16.5	1.08
apr	W	3	0.233	19.9	17	3.93
may	С	1	0.285	27	23.8	2.15
may	С	1	0.375	26.7	23.7	0.997
may	С	1	0.218	25	24	3.63
may	С	1	0.157	27.2	24	4.37
may	С	1	0.332	27.9	23.7	1.54
may	С	2	0.18	27	24.1	4.71
may	С	2	0.217	27	23.3	2.54
may	С	2	0.167	26.5	23	3.48
may	С	2	0.25	26.8	23.7	2.11
may	С	2	0.266	27.7	24	2.82
may	С	3	0.176	28.4	25.3	2.2
may	С	3	0.203	29	25	2.8
may	С	3	0.21	28	25	2.26
may	С	3	0.221	29.3	25	2.66
may	С	3	0.182	29.4	24.8	4.33
may	S	1	0.331	26.8	24.2	7.77
may	S	1	0.316	28.6	25.4	6.42
may	S	1	0.361	30.7	26.5	6.98
may	S	1	0.379	28.5	24.9	5.64
may	S	1	0.327	30.2	26	4.11

may	S	2	0.295	24	23	7.68
may	S	2	0.293	23.6	23	6.58
may	S	2	0.286	24.4	22.9	4.85
may	S	2	0.342	23.4	22.8	5.22
may	S	2	0.3	24.7	23.4	4
may	S	3	0.333	27	24.7	3.99
may	S	3	0.292	26.6	23.9	7.09
may	S	3	0.248	26.3	23.5	4.62
may	S	3	0.309	29.7	24.9	8.17
may	S	3	0.263	27	23.9	5.6
may	W	1	0.211	26.4	24	5.19
may	W	1	0.202	24.6	22.2	3.83
may	W	1	0.192	23.7	22	3.64
may	W	1	0.152	22.5	21.5	3.51
may	W	1	0.212	23.7	21.9	3.19
may	W	2	0.17	25.7	23.1	8.06
may	W	2	0.203	23.6	22.1	4.38
may	W	2	0.161	24.6	22.6	2.94
may	W	2	0.173	25.4	22.6	3.81
may	W	2	0.134	26.7	24.4	1.77
may	W	3	0.148	25	22.9	2.76
may	W	3	0.149	24.1	22.7	2.74
may	W	3	0.179	24.9	22.9	7.78
may	W	3	0.17	23.7	22	4.61
may	W	3	0.168	29.4	25.4	3.4
jun	С	1	0.387	36	30.9	4.16
jun	С	1	0.418	35.8	31.2	5.04
jun	С	1	0.395	36	30.8	5.36
jun	С	1	0.359	36.5	31.5	4.08
jun	С	1	0.449	35.9	32	3.46
jun	С	2	0.335	38	31.4	6.82
jun	С	2	0.36	37.1	32.1	3.67
jun	С	2	0.404	36	32	6.98
jun	С	2	0.365	37.6	31.7	6.45
jun	С	2	0.328	37.1	32.7	4.27
jun	С	2	0.264	36.6	32.2	4.54
jun	С	3	0.276	38	32.9	4.12
jun	С	3	0.312	35.5	31.3	3.53
jun	С	3	0.289	37.4	31.8	5.4
jun	С	3	0.24	36.9	32	5.31
jun	С	3	0.398	30.4	25.6	
jun	S	1	0.46	28.3	25.4	

jun	S	1	0.477	28.8	25.5	
jun	S	1	0.466	29.2	25.5	0.696
jun	S	1	0.416	29.2	25.5	6.24
jun	S	1	0.416	29.1	25.7	4.33
jun	S	2	0.322	27.1	26.3	5.46
jun	S	2	0.366	28.8	25.1	4.88
jun	S	2	0.396	32.8	26.4	6.85
jun	S	2	0.334	27.7	25	6.46
jun	S	2	0.248	28.7	27	6.14
jun	S	3	0.3	31.8	28.6	6.55
jun	S	3	0.316	28.8	27	7.15
jun	S	3	0.282	29.6	28	5.64
jun	S	3	0.298	29.7	27.65	6.8
jun	S	3	0.362	27.7	25	3.7
jun	W	1	0.336	26.3	24	2.98
jun	W	1	0.338	33.2	27.2	4.55
jun	W	1	0.35	32	29	5.3
jun	W	2	0.417	34.6	25.7	5.4
jun	W	2	0.287	31.8	24.7	3.7
jun	W	2	0.288	31.2	26.1	4.68
jun	W	2	0.348	32.4	27.6	2.73
jun	W	2	0.318	29	25	2.9
jun	W	3	0.4	25.6	24.28	3.46
jun	W	3	0.325	30.4	20.6	2.63
jun	W	3	0.37	33	26.3	3.45
jun	W	3	0.353	29.2	24.8	3.53
jun	W	3	0.402	29	25.4	5.34
jul	С	1	0.404	32.7	29.6	7.15
jul	С	1	0.308	37.9	32.3	6.17
jul	С	1	0.39	37.5	31	2.48
jul	С	1	0.33	34.1	31	10.6
jul	С	1	0.286	36	32.1	8.97
jul	С	2	0.244	36.7	31	7.94
jul	С	2	0.256	36.4	31.4	10.2
jul	С	2	0.244	34	31	9.7
jul	С	2	0.22	36.5	32.5	10.5
jul	С	2	0.232	37	44.7	9.585
jul	С	2	0.24	24	30	8.68
				07.4	20.0	E 2E
jul	С	3	0.279	27.4	30.8	5.35
jul jul	C C	3 3	0.279 0.312	27.4 31.2	30.8 30.6	5.35 13.5
jul jul jul	C C C	3 3 3	0.279 0.312 0.33	27.4 31.2 31	30.8 30.6 28.6	5.35 13.5 6.65

jul	С	3	0.348	27.2	26.5	4.8
jul	S	1	0.384	27.7	26.7	4.7
jul	S	1	0.332	27.1	26	4.99
jul	S	1	0.356	28	26	5.06
jul	S	1	0.338	27	25.8	4.61
jul	S	1	0.302	29.4	27	7.47
jul	S	2	0.266	31.7	28.4	7.04
jul	S	2	0.324	29	26.7	5.56
jul	S	2	0.322	30.8	28.1	7.03
jul	S	2	0.302	28.3	27	6.65
jul	S	2	0.351	26	25	3.73
jul	S	3	0.339	25.6	25	3.09
jul	S	3	0.349	26.4	25	4.74
jul	S	3	0.362	27.4	26	5.68
jul	S	3	0.287	26	25.2	4.21
jul	S	3	0.369	28	25	3.55
jul	W	1	0.29	26.3	25	2.8
jul	W	1	0.34	28.3	26.5	2.98
jul	W	1	0.384	28.9	26.9	2.64
jul	W	1	0.332	24.3	29	4.11
jul	W	2	0.312	26.7	25.7	2.89
jul	W	2	0.266	30.7	26.9	3.25
jul	W	2	0.368	33.2	27.7	6.35
jul	W	2	0.353	28	26.6	3.34
jul	W	2	0.334	26.8	28.6	3.88
jul	W	3	0.432	28	26.3	2.39
jul	W	3	0.416	27	25.4	2.98
jul	W	3	0.326	27.6	26.2	3.8
jul	W	3	0.372	27.6	25.9	3.16
jul	W	3	0.336	28.4	26.7	4
aug	С	1	0.195	27.2	23.9	3.9
aug	С	1	0.153	27	24.5	3.62
aug	С	1	0.156	26	25	4.21
aug	С	1	0.152	27.6	24.1	3.71
aug	С	1	0.138	26.6	24.4	2.97
aug	С	2	0.096	25.9	23.7	3.35
aug	С	2	0.054	26.7	24.2	3.33
aug	С	2	0.036	27.1	25	3.16
aug	С	2	0.112	25.4	23.5	3.46
aug	С	2	0.125	25.5	23.7	3.54
aug	С	3	0.098	27.6	25.7	3.36
aug	С	3	0.082	26	25	2.46

aug	С	3	0.101	29	25.4	4.08
aug	С	3	0.131	25.4	23.9	4.32
aug	С	3	0.114	26.5	23.7	4.39
aug	S	1	0.186	26.2	24.9	6.71
aug	S	1	0.306	25.7	24.3	3.9
aug	S	1	0.238	29	27	3.82
aug	S	1	0.223	27.3	25.6	2.9
aug	S	2	0.236	22.6	22.4	6.41
aug	S	2	0.232	22.6	22	4.53
aug	S	2	0.226	23.2	22.8	4.49
aug	S	2	0.206	22.9	22.1	3.71
aug	S	2	0.263	23.7	23.6	5.3
aug	S	3	0.244	25.7	24	4.21
aug	S	3	0.224	26.8	24.5	3.47
aug	S	3	0.252	25.6	24	5.54
aug	S	3	0.218	24.9	23.6	3.07
aug	W	1	0.11	26.6	25.3	2
aug	W	1	0.208	25	24	2.77
aug	W	1	0.118	24	25.2	1.92
aug	W	1	0.152	26	25	2.56
aug	W	1	0.126	25.3	24.2	2.96
aug	W	2	0.158	22.5	25.4	4.64
aug	W	2	0.078	28.1	26.8	2.48
aug	W	2	0.102	28.3	26	4.45
aug	W	2	0.096	29	26	2.03
aug	W	2	0.163	27.4	25.7	3.35
aug	W	3	0.09	26.8	24.5	2.48
aug	W	3	0.197	26	23.8	2.71
aug	W	3	0.133	32	26.7	5.77
aug	W	3	0.176	29	25.6	3.71
aug	W	3	0.182	29	25.9	1.68
sep	С	1	0.007	30.6	27	2.03
sep	С	1	0.005	35	30.7	1.64
sep	С	1	0.02	34.5	27.4	1.77
sep	С	1	0.034	31.4	26.4	1.88
sep	С	1	0.05	28.7	24.6	1.52
sep	С	2	0.03	29	26	2.14
sep	С	2	0.022	30.8	26.4	1.785
sep	С	2	0.008	31.1	30.4	1.64
sep	С	2	0.006	27.7	24	1.89
sep	С	2	0.018	28.3	24.1	1.47
sep	С	3	0.02	27.1	23.8	1.82

sep	С	3	0.008	26	22	1.83
sep	С	3	0.019	30.5	26	3.67
sep	С	3	0.022	29	25	0.765
sep	С	3	0.003	33.9	25.4	0.987
sep	S	1	0.054	19.6	19	1.74
sep	S	1	0.078	19.4	19.1	1.34
sep	S	1	0.073	19.6	19	1.4
sep	S	1	0.06	20.7	19.9	1.6
sep	S	1	0.036	22	20.8	2.28
sep	S	2	0.004	33.1	24.1	3.51
sep	S	2	0.054	23	22	1.53
sep	S	2	0.078	30.2	26	3.43
sep	S	2	0.042	24.7	21.9	2.26
sep	S	3	0.061	19.1	18.9	1.97
sep	S	3	0.06	20.5	19.6	1.49
sep	S	3	0.032	21	19.6	1.58
sep	S	3	0.074	19.5	18.6	2.16
sep	S	3	0.096	19	19.175	1.37
sep	W	1	0.06	24.7	22.7	0.606
sep	W	1	0.053	23	21.6	1.14
sep	W	1	0.088	25	22.4	0.489
sep	W	1	0.049	24.6	21.9	0.71
sep	W	1	0.081	26.9	24	0.688
sep	W	2	0.058	25	21.9	0.596
sep	W	2	0.063	22	21.7	1.52
sep	W	2	0.043	23	22	1.11
sep	W	2	0.092	26.3	22.5	1.04
sep	W	2	0.055	23.5	22.4	1.31
sep	W	3	0.038	28	25.4	0.552
sep	W	3	0.061	25.6	22.5	0.055
sep	W	3	0.04	23	22	1.21
sep	W	3	0.045	28.2	24	1.2
sep	W	3	0.054	32.9	25.1	1.3
oct	С	1	0.322	20.4	17.5	2.25
oct	С	1	0.532	22.3	19.3	1.98
oct	С	1	0.343	22.8	17.9	2.37
oct	С	1	0.399	20.9	17.2	3.03
oct	С	1	0.41	19.5	16.3	2.48
oct	С	2	0.265	19.2	16.9	2.24
oct	С	2	0.28	19.9	17.1	1.95
oct	С	2	0.285	20.4	19.1	2.095
oct	С	3	0.248	19.2	15.9	2.67

oct	С	3	0.294	17.4	14.9	1.45
oct	С	3	0.305	20.3	16.9	2.63
oct	С	3	0.283	19.5	16.4	1.99
oct	С	3	0.283	21.8	16.6	1.91
oct	S	1	0.198	14	13	1.56
oct	S	1	0.344	13.9	13.1	1.57
oct	S	1	0.343	13.6	13	3.61
oct	S	1	0.295	14.4	13.5	3.11
oct	S	1	0.3	15	13.9	3.03
oct	S	2	0.306	22.4	16.5	1.83
oct	S	2	0.267	18.7	16	3.78
oct	S	2	0.307	17.7	15.2	3.57
oct	S	2	0.293	20.7	17.1	2.39
oct	S	2	0.29	17.9	15.2	1.57
oct	S	3	0.303	13.5	13.2	4.33
oct	S	3	0.351	14.4	13.5	3.04
oct	S	3	0.278	15.1	13.5	3.87
oct	S	3	0.311	14	13	4.08
oct	S	3	0.31	13.8	13.3	2.89
oct	W	1	0.262	17.8	15.3	4.65
oct	W	1	0.259	16.5	14.8	3.5
oct	W	1	0.363	17.7	15.4	4.08
oct	W	1	0.295	17.5	15	4.22
oct	W	1	0.295	18.6	16.1	4
oct	W	2	0.255	16.9	14.7	2.14
oct	W	2	0.309	15.4	14.9	3.59
oct	W	2	0.296	16.5	14.5	3.45
oct	W	2	0.287	17.7	15	2.63
oct	W	2	0.287	16.4	15	3.2
oct	W	3	0.249	18.6	16.5	3.13
oct	W	3	0.26	17.8	15	3.14
oct	W	3	0.193	16.2	15	2.56
oct	W	3	0.234	18.9	15.8	3.74
oct	W	3	0.234	21.3	16.4	4.1
nov	С	1	0.331	10.2	8	0.912
nov	С	1	0.426	9.6	7.9	0.525
nov	С	2	0.352	9.3	7.7	0.512
nov	С	2	0.334	9	7.7	0.548
nov	С	2	0.408	9.7	7.8	0.622
nov	С	3	0.336	10.2	7.7	0.687
nov	С	3	0.371	11.2	8	0.733
nov	С	3	0.302	8.8	7.7	0.737

nov	S	1	0.323	8.3	7	0.424
nov	S	1	0.365	8.4	7.1	1.11
nov	S	1	0.416	7.6	7	0.475
nov	S	2	0.242	11.6	8.9	0.552
nov	S	2	0.323	9.6	8.4	1.04
nov	S	2	0.254	12.3	8.41	0.879
nov	S	3	0.392	7.9	7.4	0.341
nov	S	3	0.386	8.2	7.3	0.409
nov	S	3	0.363	9.1	7.4	0.426
nov	W	1	0.39	10.9	7.9	0.771
nov	W	1	0.32	10	7.9	0.671
nov	W	1	0.376	10.4	8.4	0.902
nov	W	2	0.364	8.7	7.4	0.331
nov	W	2	0.273	8.7	7.7	0.817
nov	W	2	0.318	10	7.4	0.67
nov	W	3	0.355	9.2	7.5	0.423
nov	W	3	0.394	10	7.4	0.992
nov	W	3	0.32	9.4	7.9	1.44
dec	С	1	0.379	9.2	6.7	0.42
dec	С	1	0.463	12.6	7	0.203
dec	С	2	0.418	7.2	5	0.435
dec	С	2	0.378	10.8	6	0.438
dec	С	2	0.468	9	5.8	0.819
dec	С	3	0.344	13.4	5.8	0.556
dec	С	3	0.338	8.7	6	0.814
dec	С	3	0.378	8.7	6.7	0.282
dec	S	1	0.416	8	4.4	0.29
dec	S	1	0.493	7	4.7	0.105
dec	S	1	0.48	6.3	4.7	0.529
dec	S	2	0.356	8.6	5.9	0.342
dec	S	2	0.452	9	6	0.42
dec	S	2	0.342	9.6	6.3	0.381
dec	S	3	0.499	5.9	4.7	0.257
dec	S	3	0.456	7.7	5	0.346
dec	S	3	0.426	6.9	4.9	0.2
dec	W	1	0.439	11.4	7	0.482
dec	W	1	0.38	9.6	6	0.53
dec	W	1	0.466	11.2	7	0.333
dec	W	2	0.504	10	6.6	-0.212
dec	W	2	0.422	9	6	0.893
dec	W	2	0.35	8.7	4.9	0.381
dec	W	3	0.494	6.7	5.1	0.314

dec	W	3	0.404	10.7	5.9	0.425
dec	W	3	0.366	9.6	5.2	0.399

Annual Emission data set for both years. Ecosystem (Eco), Plot (Rep), Year: 2012 (1) and 2013 (2), Emission= Kg C ha<sup>-1</sup>.

Eco	Rep	Year	Emissions
с	1	1	10875
с	2	1	8935
с	3	1	11198
S	1	1	9610
S	2	1	8949
S	3	1	9966
W	1	1	6588
W	2	1	8086
W	3	1	7277
с	1	2	9311
с	2	2	9951
с	3	2	9843
S	1	2	8163
S	2	2	9746
S	3	2	9624
W	1	2	7302
W	2	2	7402
W	3	2	7429

2013 Growing Season Microbial Biomass Data Set. Ecosystem (Treat), Plot (Rep), Microbial Biomass Carbon (TOC), Microbial Biomass Nitrogen (TN), and Microbial Biomass C:N ratio (CN).

Month	Treat	Rep	TOC	TN	CN	
apr	С	1	42.14	16.01	2.63	
apr	С	1	0	0	0	
apr	С	1	25.87	4.82	5.37	
apr	С	2	31.18	12	2.6	
apr	С	2	25.23	4.52	5.59	
apr	С	2	39.79	12.62	3.15	
apr	С	3	34.56	13.05	2.65	
apr	С	3	46.91	5.99	7.83	
apr	С	3	23.61	8.58	2.75	
apr	S	1	31.84	14.78	2.15	

apr	S	1	39.95	18.73	2.13
apr	S	1	20.83	3.56	5.86
apr	S	2	45.33	19.71	2.3
apr	S	2	33.16	12.94	2.56
apr	S	2	55.89	15.89	3.52
apr	S	3	22.07	4.1	5.38
apr	S	3	0	0.72	0
apr	S	3	0	2.77	0
apr	W	1	29.66	7.57	3.92
apr	W	1	34.79	9.4	3.7
apr	W	1	24.07	10.03	2.4
apr	W	2	5.64	1.78	3.17
apr	W	2	11.07	3.07	3.6
apr	W	2			
apr	W	3	41.3	4.31	9.58
apr	W	3	54.68	12.08	4.53
apr	W	3	26.67	10.25	2.6
may	С	1	84.76	0	0
may	С	1	85.96	16.44	5.23
may	С	1	87.33	16.09	5.43
may	С	2	84.92	11.47	7.4
may	С	2	77.96	15.57	5.01
may	С	2	95.24	24.11	3.95
may	С	3	89.73	9.68	9.27
may	С	3	78.5	22.58	3.48
may	С	3	78.64	23.27	3.38
may	S	1	88.6	15.09	5.87
may	S	1	89.09	15.83	5.63
may	S	1	92.58	18.44	5.02
may	S	2	84.12	15.79	5.33
may	S	2	88.1	14.99	5.88
may	S	2	87.24	15.2	5.74
may	S	3	103.65	19.17	5.41
may	S	3	80.54	13.98	5.76
may	S	3	91.78	15.16	6.05
may	W	1	163.35	14.78	11.06
may	W	1	89.39	14.79	6.04
may	W	1	99.93	16.08	6.22
may	W	2	67.86	10.04	6.76
may	W	2	95.65	12.09	7.91
may	W	2	84.7	13.11	6.46
may	W	3	108.34	20.42	5.3

may	W	3	106.31	15.95	6.66
may	W	3	96.35	14.75	6.53
june	С	1	111.39	22.71	4.9
june	С	1	91.65	18.58	4.93
june	С	1	101.91	22.03	4.63
june	С	2	81.48	16.68	4.89
june	С	2	87.11	14.69	5.93
june	С	2	52.02	12.15	4.28
june	С	3	85.63	17.56	4.88
june	С	3	91.73	19.2	4.78
june	С	3	81.35	14.05	5.79
june	S	1	27.52	1.66	16.58
june	S	1	91.39	13.57	6.74
june	S	1	127.02	32.77	3.88
june	S	2	75.15	8.43	8.91
june	S	2	78.41	9.16	8.56
june	S	2	76.94	8.66	8.89
june	S	3	80.28	16.02	5.01
june	S	3	42.46	3.68	11.55
june	S	3	92.08	14.99	6.14
june	W	1	30.79	2.59	11.9
june	W	1	64.62	6.76	9.57
june	W	1	69.55	8.26	8.42
june	W	2	47.1	3.4	13.84
june	W	2	104.7	13.67	7.66
june	W	2	71.56	11.81	6.06
june	W	3	81	14.28	5.67
june	W	3	44.25	9.08	4.87
june	W	3	77.26	9.85	7.84
july	С	1	54.56	2.13	25.63
july	С	1	66.37	0	0
july	С	1	65.02	3.52	18.48
july	С	2	46.36	9.78	4.74
july	С	2	83.27	12.89	6.46
july	С	2	50.65	6.53	7.76
july	С	3	15.56	3.83	4.06
july	С	3	36.26	54.6	0.66
july	С	3	29.82	0	0
july	S	1	47.14	15.59	3.02
july	S	1	52.01	5.5	9.45
july	S	1	24.91	14.42	1.73
july	S	2	44.81	11.47	3.91

july	S	2	33.61	10.71	3.14
july	S	2	106.14	25.76	4.12
july	S	3	23.91	4.54	5.26
july	S	3	55.06	10.78	5.11
july	S	3	66.97	12.76	5.25
july	W	1	25.97	8.53	3.04
july	W	1	0	0	0
july	W	1	29.93	9.69	3.09
july	W	2	30.92	7.46	4.14
july	W	2	64.32	11.22	5.73
july	W	2	36.05	9.5	3.8
july	W	3	10.68	0.69	15.52
july	W	3	28.49	0	0
july	W	3	26.06	8.91	2.92
aug	С	1	63.88	4.97	12.84
aug	С	1	82.01	12.8	6.41
aug	С	1	73.19	9.28	7.89
aug	С	2	86.9	8.56	10.15
aug	С	2	82.69	5.2	15.91
aug	С	2	41.31	1.36	30.41
aug	С	3	45.34	2.92	15.53
aug	С	3	79.2	5.62	14.1
aug	С	3	83.12	5.51	15.08
aug	S	1	115.57	12.88	8.97
aug	S	1	56.66	12.6	4.5
aug	S	1	71.42	9	7.94
aug	S	2	62.49	5.59	11.17
aug	S	2	68.59	5.89	11.65
aug	S	2	80.22	7.1	11.29
aug	S	3	34.41	9.12	3.77
aug	S	3	82.21	12.44	6.61
aug	S	3	62.49	10.28	6.08
aug	W	1	97.19	6.63	14.67
aug	W	1	100.6	11.83	8.51
aug	W	1	64.55	5.53	11.67
aug	W	2	65.07	12.52	5.2
aug	W	2	75.79	15	5.05
aug	W	2	94.51	17.5	5.4
aug	W	3	114.3	19.99	5.72
aug	W	3	67.73	6.71	10.1
aug	W	3	68.97	4.86	14.18
sep	С	1	52.68	2.92	18.04

sep	С	1	47.12	3.38	13.95
sep	С	1	39.56	0.85	46.31
sep	С	2	31.16	2.55	12.21
sep	С	2	47.09	2.47	19.09
sep	С	2	50.6	2.94	17.21
sep	С	3	49.53	2.86	17.34
sep	С	3	44.34	1.55	28.61
sep	С	3	76.42	4.45	17.16
sep	S	1	71.79	8.78	8.18
sep	S	1	69.62	7.16	9.72
sep	S	1	68.29	7.66	8.91
sep	S	2	46.21	3.71	12.46
sep	S	2	70.87	6.1	11.61
sep	S	2	52.58	4.02	13.07
sep	S	3	53.4	4.6	11.61
sep	S	3	54.28	4.52	12
sep	S	3	66.24	4.41	15.04
sep	W	1	72.9	4.79	15.21
sep	W	1	41.74	2.46	16.98
sep	W	1	52.08	3.24	16.07
sep	W	2	0	6.31	0
sep	W	2	41.32	3.22	12.85
sep	W	2	27.07	2.34	11.57
sep	W	3	61.01	4.96	12.29
sep	W	3	49.19	3	16.39
sep	W	3	130.42	6.51	20.04
oct	С	1	94.67	11.18	8.47
oct	С	1	90.04	13.19	6.83
oct	С	1	81.97	13.31	6.16
oct	С	2	68.96	9.11	7.57
oct	С	2	89.02	12.9	6.9
oct	С	2	86.8	11.63	7.46
oct	С	3	32.1	0	0
oct	С	3	100.08	13.32	7.52
oct	С	3	85.3	11.94	7.14
oct	S	1	46.64	6.37	7.32
oct	S	1	124.82	17.79	7.02
oct	S	1	84.77	9.4	9.02
oct	S	2	97.64	13.7	7.12
oct	S	2	134.58	20.35	6.61
oct	S	2	118.85	15.88	7.49
oct	S	3	60.98	7.03	8.68

oct	S	3	106.68	16.54	6.45
oct	S	3	76.48	10.14	7.54
oct	W	1	40.62	2.55	15.92
oct	W	1	70.27	8.47	8.3
oct	W	1	101.15	13.43	7.53
oct	W	2	102.4	11.46	8.94
oct	W	2	88.69	14.77	6.01
oct	W	2	116.1	17.13	6.78
oct	W	3	95.06	13.12	7.25
oct	W	3	116.03	16.53	7.02
oct	W	3	47.43	7.81	6.07

Agroecosystem Row Spacing for 2012 and 2013 data set. Space: Row (R) and In-between Row (I) ) Replication (Rep), 0-6cm Volumetric water content (Vwc, cm<sup>3</sup> cm<sup>-3</sup>), 2cm soil temperature, (Temp2cm, °C), 10cm soil temperature (Temp10cm, °C), and CO<sub>2</sub> flux (Flux,  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>)

Month	Space	Rep	VWC	Ttemp2cm	Temp10cm	Flux
may	R	3	0.214	34	26.3	4.39
may	Ι	4	0.19	32.6	26.79	3.17
may	Ι	2	0.187	36.6	28.09	3.55
may	R	1	0.199	33.3	27.46	4.03
may	R	5	0.246	33.1	26.82	3.67
may	Ι	5	0.064	35.8	27.82	2.48
may	R	4	0.112	33.5	27.86	6.49
may	Ι	3	0.157	31.5	27.54	4.23
may	R	2	0.163	35.7	27.1	6.04
may	Ι	1	0.153	34.4	29.2	3.46
may	R	5	0.188	32.6	25.83	3.18
may	R	4	0.191	31.1	25.24	2.91
may	R	3	0.12	34.4	25.99	2.31
may	Ι	2	0.161	33.2	25.45	6.13
may	Ι	1	0.118	37.7	27.3	3.65
june	R	1	0.034	41		3.58
june	Ι	2	0.055	43.4	39.72	1.56
june	Ι	3	0.063	45	36.96	3.22
june	Ι	3	0.19	47.5	36.73	2.85
june	R	2	0.045	46.2		4.96
june	Ι	1	0.028	48.9	38.4	1.87
june	R	1	0.032	43.7		2.52
june	R	2	0.068	46.8		3.52
june	R	3	0.068	42.9		2.12

july	Ι	5	0.316	33.5	39.36	9.99
july	Ι	1	0.284	34	30.92	9.15
july	R	4	0.25	34	31.52	10.6
july	R	3	0.194	32.5	29.84	8.85
july	Ι	2	0.318	37.8	31.17	8.31
july	R	1	0.211	31.8	29.08	4.41
july	Ι	2	0.08	39.2	32.99	5.15
july	R	5	0.133	31.9	30.03	5.43
july	Ι	4	0.127	32.5	30.69	7.28
july	R	3	0.07	39.6	33.1	4.01
july	R	3	0.173	34.1	29.35	11.8
july	R	2	0.173	31.8	28.9	10.7
july	Ι	1	0.072	42.3	31.86	4.99
july	Ι	5	0.292	33.2	29.62	10.6
aug	Ι	5	0.022	35.3	31.4	4.33
aug	Ι	1	0.113	34.1	31.02	4.7
aug	R	4	0.071	35.5	29.04	4.19
aug	R	3	0.112	33.7	28.81	4.73
aug	Ι	2	0.097	36	32.4	3.35
aug	R	2	0.031	44	40.43	3.53
aug	Ι	3	0.036	41.5	31.89	2.84
aug	R	4	0.044	38	31.65	2.49
aug	Ι	1	0.078	39.2	33.17	2.13
aug	R	5	0.023	35.2	30.46	1.94
aug	R	5	0.092	33.7	29	4.49
aug	R	1	0.05	39.2	31.31	3.17
aug	Ι	2	0.094	33.5	30.91	4.06
aug	Ι	3	0.06	34.8	28.34	3.61
sep	Ι	4	1.88	24.9	23.36	3.17
sep	Ι	5	0.743	26.1	23.88	4.2
sep	R	2	0.201	26.2	23.66	3.41
sep	R	3	0.148	25.8	24.28	4.9
sep	R	1	0.076	27.9	25.71	4.57
sep	Ι	1	0.132	26.1	24.8	1.19
sep	R	2	0.118	24.66	25.6	2.52
sep	R	3	0.094	23.8	23.45	4.12
sep	R	3	0.169	23.8	23.64	3.27
sep	Ι	2	0.085	23.8	24.3	2.66
oct	Ι	2	0.382	23.1	17.9	2.04
oct	Ι	3	0.306	23.1	18.4	2.25
oct	R	1	0.376	21.6	18.8	2.75
oct	R	4	0.356	23.1	19.2	1.56

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oct	I	5	0.325	21.2	18.6	1.68
oct	R	1	0.201	23.1	19.4	3.22
oct	Ι	2	0.223	23.2	19.4	2.8
oct	R	4	0.235	23.3	19.7	3.04
oct	Ι	3	0.294	23.4	18.9	1.95
oct	R	5	0.254	23	19.3	3.41
oct	R	5	0.3	22.1	18.1	1.86
oct	R	4	0.323	22.6	18.4	2.78
oct	Ι	3	0.342	21.3	17.9	2.39
oct	Ι	2	0.348	20.3	17.7	1.68
nov	R	5	0.328	16.2	12	1.16
nov	Ι	4	0.354	16.2	12	1.31
nov	R	3	0.328	16.2	12	0.959
nov	Ι	2	0.465	16.2	12	1.26
nov	Ι	1	0.396	16.2	12	1.21
nov	Ι	1	0.215	17.6	11.6	1.08
nov	Ι	2	0.175	15.7	12.3	1.39
nov	Ι	3	0.322	15.4	12	1.08
nov	R	4	0.199	16.2	12	1.91
nov	R	5	0.168	16.2	12	1.23
nov	Ι	1	0.286	14	11.3	2.03
nov	R	2	0.317	13.3	11.1	1.22
nov	R	3	0.268	14.2	11	0.766
nov	Ι	4	0.255	12.9	11.1	1.34
nov	Ι	5	0.219	13.3	11.3	1.58

Conclusion

## Conclusion

This study demonstrated that switchgrass and cottonwood grown as bioenergy feedstocks in the LMAV did not increase soil respiration relative to a traditional soybean-grain sorghum agroecosystem. Although 2012 and 2013 differed greatly in precipitation, similar trends for soil temperature, moisture and  $CO_2$  flux were observed for both years. As predicted, all treatments showed general trends in  $CO_2$  flux throughout the year. Carbon dioxide fluxes increased from winter lows throughout the spring, peaking during summer months, then falling during autumn back to winter lows. Additionally, the greatest  $CO_2$  flux measured was in the agroecosystem, as predicted. However, collar placement did not affect measurements in the agroecosystem for either year.

In general, the agroecosystem and the switchgrass ecosystem were similar in CO<sub>2</sub> fluxes throughout both years, while the cottonwood had generally lower fluxes throughout both years. This trend was also evident when annual emissions from the ecosystems were quantified. Contrary to what was hypothesized, annual emissions from the agroecosystem and switchgrass ecosystem were similar, while the cottonwood had significantly lower annual emissions. Soil MBC and MBN did not differ significantly by treatment for the 2013 growing season, unlike what was predicted, however, the agroecosystem did have the greatest C:N ratio

As predicted, soil temperature and moisture play large roles in controlling soil respiration, and can be used to account for a large portion of the variation in soil respiration. However, this study showed that, when only accounting for growing-season  $CO_2$  flux, when soil temperature are more consistent, other parameters, such as MBN, should be taken into account. This suggests that, during optimum soil temperature conditions, other variables form a more complex set of controlling factors. This study suggests that switchgrass and cottonwood, grown
as bioenergy feedstocks in the LMAV, do not increase soil respiration compared to a regionally common agroecosystem. In addition, cottonwood grown as bioenergy feedstock may decrease soil respiration, which may eventually help to increase soil C stocks in the LMAV.