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Transition to second generation cellulosic biofuel production systems reveals limited negative impacts on the soil microbial community structure.

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ABSTRACT

Currently we lack knowledge on how the soil microbial community responds to the transition from traditional grain production to cellulosic feedstock production systems with residue removal for biofuel production. Implementation of second-generation biofuel production systems could create a challenge in meeting the goal of providing cellulose biomass if the processes impact soil's function as part of ecosystem services. Our goal was to assess the transition period as we move from a grain harvest system with residue return into biomass production systems with both residue and grain removal. The existing traditional systems were continuous corn (CC) and corn–soybean rotations (corn–soybean–corn (R1) or soybean–corn–soybean (R2)). The biomass production systems include a previously established mixed tall grass prairie dominated by big bluestem (*Andropogon gerardii*; PR) where the above ground biomass is now harvested when it had been previously burned, corn–soybean rotations transitioned to residue recovery based on: *Miscanthus* × *giganteus* (MS), dual-purpose sorghum (*Sorghum bicolor*; SG), upland switchgrass (*Panicum virgatum*; c.v. *Shawnee*; SW) and a tilled continuous corn changed to continuous no-till corn (*Zea mays*; CR) with the residue removed. Over a three-year transition period, biological indicators of soil health included microbial biomass and community structure profile using phospholipid fatty acid (PLFA) biochemical markers, and an assessment of the saprotrophic fungal diversity using denaturing gradient gel electrophoresis (DGGE) of polymerase chain reaction amplified ITS rRNA genetic marker. Over the course of the transition, bacterial biomass dominated soils in the annual systems of CC, R1, R2, CR and SG, and the perennial system of MS. While fungal biomass dominated soils under PR. Fungal PLFA signatures and ITS-DGGE profiles demonstrated similarity between PR and the transitioned SW system, and to a lesser extent SG. In contrast, MS did not statistically alter the microbial biomass, community structure or fungal diversity away from patterns observed in the corn–soybean system. Our findings support microbial community stability and resilience as we found no negative impacts in any of the biomass recovery systems (PR, MS, SG, SW and CR) relative to the traditional systems (CC, R1 and R2), as indicated by measures of soil health. We did find that the selection of the plant types used in the biomass system could have a significant impact on the structure of both bacterial and fungal communities. With respect to soil health indicators including the fungal community, our findings suggest the dual-purpose SG system provides an encouraging alternative to continuous maize when considering plant biomass production. The selection of this dedicated perennial biomass system offers an avenue to enhance indicators of soil health and associated ecosystem services while supporting the bioenergy economy.

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1. Introduction

The possible widespread introduction of second-generation cellulosic ethanol production within the Midwestern Corn Belt offers a unique opportunity for landscape diversification, but the impacts

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on soil health are unclear. In 2012, fermentation of corn grain was the predominant agricultural source for ethanol (first-generation biofuel), with the United States producing 13.3 billion gallons of ethanol using this process (Renewable Fuels Association, 2013). However, the use of grain has raised concerns related to food market competition and hidden environmental costs; consequently the development of bioethanol utilizing pre-existing crop residues and/or dedicated plant biomass have been in development (Hill et al., 2006; Hill, 2007). The cellulosic ethanol platform is now an emerging technology with some 20 facilities and projects under development across the U.S. (Advanced Ethanol Council, 2013). Cellulosic ethanol offers flexibility as materials can originate either from pre-existing grasslands or annual cropland, or from the establishment of dedicated perennial biomass systems. Inclusive of the production system chosen, it is important to identify and understand ecosystem impacts on the soil quality and health as the crop residues, a major nutrient input to soil, are removed.

The quality of the soil resource is a fundamental component of agroecosystem sustainability, driving system stability and the long-term capacity to produce plants. In turn, soil microbial dynamics drive soil quality, and changes in soil biology can serve as predictive indicator of negative impacts of a purposed change (Kennedy and Smith, 1995). In the discussion of agroecosystem sustainability, the term soil health is used interchangeably with soil quality. However, we use soil health to emphasize the living nature of soil that is dynamic in both functional capacity and response to management changes (Doran and Zeiss, 2000). Soil health is a function of the soil microbial community's ability to recover (i.e., resilience), and the soils buffering capacity (i.e., resistance) (Holling, 1987). Therefore, the expression of both resilience and resistance is unique to soil type, environmental conditions, and the legacy effects of past management (Wertz et al., 2007).

With regard to best management practices for cellulose biofuel production, changes in the type and abundance of above and belowground organic inputs may influence soil biology by shifting metabolic niche diversity with C input changes (Blagodatsky et al., 2010; Jangid et al., 2011; Coleman and Whitman, 2005). Transitioning to perennial plant species or no-till practices may change microbial abundance and community structure by changing soil structure through increased aggregate formation (Drijber et al., 2000; Frey et al., 1999). Though some reports have demonstrated strong plant species effects (Aira et al., 2010; Kuske et al., 2002; Grayston et al., 2001; Hartmann et al., 2009; Heděnec et al., 2014; Smalla et al., 2001), others have reported that changes in vegetation had no effect on soil biology (Girvan et al., 2003; Hedlund et al., 2003; Kennedy et al., 2004.)

A clear case of the environmental detriment is the decline in soil organic carbon (SOC) under increased stover harvest in continuous maize production systems has been shown in multiple studies (Blanco-Canqui and Lal, 2007; Benjamin et al., 2010; Lal, 2005; Lemus and Lal, 2005; Orr, 2012). In light of the identified risks associated with the possible loss of SOC, distinct knowledge gaps concerning the impacts on soil biology and more broadly soil health associated with proposed cellulosic feedstock production systems exist.

In this paper, we report on a side-by-side production system comparison of how biological measures of soil health shifts with the management changes (e.g., system changes away from corn–soybean and the use of residue removal). The application of both phospholipid fatty acid (PLFA) biochemical profiling and molecular fingerprinting of the saprotrophic fungal community by polymerase chain reaction denaturing gradient gel electrophoresis (PCR-DGGE) can provide a thorough evaluation on the effect of environmental drivers such as plant species and disturbance on the soil microbial community composition (Agnelli et al., 2004; Acosta-Martinez et al., 2010; Anderson and Cairney, 2004; Blume et al., 2002; Brodie et al., 2003; Coleman and Whitman, 2005; García-Orenes et al., 2013; Gil et al., 2011; Heděnec et al., 2014; Liang et al., 2012). Combining biochemical and genetic profiling provides complementary surveys, with PLFA signatures distinguishing abundance of broad microbial groups (e.g., Gm−/+ bacteria, fungi), while resolution of PCR-DGGE can be refined to target populations of interest (e.g., soil fungi) to the genus level (Anderson and Cairney, 2004) as saprotrophic fungi are an important functional group in litter decomposition and soil formation. Recent advances in molecular techniques have expanded evaluation of shifts in diversity as additional early indicators of changes in soil health (Duchicela et al., 2013). Notwithstanding conflicting findings, consideration of management practices on soil microbiology is a valuable component in assessing soil health related to long-term ecosystem sustainability.

Our assessments of cellulosic biofuel production systems are relative to traditional corn–soybean systems typical of the Midwest, USA. We anticipated shifts in the soil community structure related to the start of residue removal and choice of cellulosic biofuel feedstock compared to traditional grain corn–soybean production systems. Therefore, the objectives were to examine: (i) if a conversion from corn-soybean to cellulosic biofuel systems affected microbial group abundance and community structure and (ii) compare annual and perennial biofuel crop production system impacts on soil biology. This effort was part of lager study investigating production system changes and possible impacts on nutrient losses to drainage water and conducted on the long-term plots at Purdue's Water Quality Field Station.

2. Materials and methods

2.1. Site description

Candidate cellulosic feedstock production systems were established on a subset of field plots at the Purdue University Water Quality Field Station (WQFS; 40.498667, −86.998111) located at the Agronomy Center for Research and Education (ACRE). A detailed description of WQFS can be found in Hofmann et al. (2004) and Hernandez-Ramirez et al. (2009). The WQFS was initially established in 1992 with four field replicates representing twelve agronomic management treatments in a randomized complete-block design. Field plots are located on top of drainage lysimeters (10 m × 48.5 m) which were constructed by interlocking bentonite clay walls into the subsurface glacial till. Each plot contains a drainage collection tile drain at a depth of 0.9 m. To quantify the impacts of the cropping strategies linked to residue removal in 2007 five corn or soybean plots were converted to biofueld production systems (Table 1) and were grown adjacent to conventional corn–soybean systems (with residue return).

2.2. Transition to cellulosic biofuel systems

Pre-existing but residue recovery modified systems included continuous corn and tall grass big bluestem dominated prairie. The prairie system (PR) was seeded with big bluestem (*Andropogon gerardii* Vitman), indiangrass (*Sorghastrum nutans* (L.) Nash), and native forbs in 1992. The PR system was burned annually until 2006, and transitioned to biomass harvest in 2008. A continuous corn treatment was converted to no-till in the fall of 2007 to offset the impact of residue removal (CR). Prior to the planting of dual-purpose sorghum (*Sorghum bicolor*; SG), the treatment plots had been in continuous corn production. The most substantial land use change was associated with establishing perennial monocultures biomass feedstocks of upland ecotype Shawnee switchgrass (*Panicum virgatum*; SW) and *Miscanthus* × *giganteus* (MS) into plots

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