



# Can soil nitrogen dynamics explain the yield benefit of crop diversification?

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## ABSTRACT

Diversification of grain-based cropping systems with forage legumes is commonly observed to enhance grain yields, yet the specific causes of this benefit remain poorly understood. One proposed cause is greater N availability, particularly late in the growing season, as these systems typically include organic N inputs such as legume residues and manure that may release mineral N over an extended period. In this study, we utilized a long-term cropping systems experiment in Iowa, USA to determine if differences in soil N dynamics could explain greater corn yield and N uptake in diversified cropping systems. The experiment compared a simple 2-year corn-soybean rotation system with two more diverse systems: a 3-year rotation of corn-soybean-oats/red clover and a 4-year rotation of corn-soybean-oats/alfalfa-alfalfa. The simple system relied on inorganic N fertilizers, whereas the diversified systems received a combination of inorganic N fertilizers and composted cattle manure, as well as forage legume residues. Measurements included long-term (12-year) corn yields, corn N uptake, leaf N concentration, and soil inorganic N pools (0–30 cm) over two growing seasons, and anaerobic potentially mineralizable N (PMN) and gross ammonification (0–20 cm) over a single growing season. Relative to the simple cropping system, corn yields and maximum corn N content in the diversified cropping systems were enhanced by 4% ( $0.85 \text{ Mg ha}^{-1}$ ) and 6–20% ( $10\text{--}28 \text{ kg N ha}^{-1}$ ), respectively, confirming the benefit of crop diversification. The diversified systems also reduced soil inorganic N pools by an average of 11–28% ( $5\text{--}13 \text{ kg N ha}^{-1}$ ) and enhanced anaerobic potentially mineralizable N by 18–33% ( $13\text{--}24 \text{ kg N ha}^{-1}$ ). However, neither soil inorganic N nor PMN were related to corn N uptake or yield, suggesting that inorganic N pools and net N mineralization were not responsible for differences among treatments in corn yields. After accounting for spatial differences in soil organic C, gross ammonification measured late in the season was positively related to corn leaf N concentration and total N content. However, this relationship was not specific to the diversified cropping systems and thus did not explain the crop diversification effect. Overall, we reject the hypothesis that soil N availability plays a major role in boosting corn yields in the diversified systems, and we suggest several alternative lines of investigation for future research, including crop-microbe interactions and soil physical properties.

## 1. Introduction

Diversified crop rotations regularly enhance grain crop yields by 10% or more relative to simple rotations (Bennett et al., 2012), a phenomenon that can be referred to as ‘the diversification effect.’ Diversified rotations are also capable of achieving high yields with fewer external inputs, such as synthetic N fertilizer (Crews and Peoples, 2004; Davis et al., 2012). Reduced reliance on external N inputs can in turn reduce the soil inorganic N pool size and the associated risk of N pollution (Di and Cameron, 2002; Drinkwater and Snapp, 2007). In the US Corn Belt, diversified crop rotations have been shown to maintain or increase crop yields while reducing environmental N losses to air and water resources (Matson et al., 1997; Tomer and Liebman, 2014). Greater soil N cycling activity in diversified cropping systems has been

proposed as an explanation for these effects (Copeland and Crookston, 1992; Smith et al., 2008; Bennett, et al., 2012). Concordant with this hypothesis are observations that crop rotations including leguminous or small grain crops can increase mineralization of soil organic N (SON), thereby increasing N availability to subsequent crops in the rotation sequence (Mengel, 1996; Kramer et al., 2002; Gaudin et al., 2015).

Characterizations of the supply of crop available N produced by N mineralization have generally focused on measurements of net N mineralization (Carpenter-Boggs et al., 2000; Deng and Tabatabai, 2000; Sanchez et al., 2001; Russell et al., 2006; Spargo et al., 2011). Previous studies have often assumed that positive net N mineralization and a large pool of inorganic N are required for supplying adequate amounts of N to a crop (e.g. Magdoff, 1991). However, researchers have begun to recognize the importance of gross ammonification and inorganic N

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immobilization in agricultural systems. Several studies have demonstrated that gross ammonification (the gross flux of  $\text{NH}_4^+$  produced by microbial mineralization of organic N compounds) may in fact provide N to plants beyond that available through net N mineralization, as plants can compete effectively with the other major consumers of soil N, the microbes responsible for N immobilization (Kaye and Hart, 1997; Xu et al., 2007; Inselsbacher et al., 2010). Gross ammonification is often an order of magnitude greater than net N mineralization (Booth et al., 2005; Osterholz et al., 2017b), so net N mineralization may significantly underestimate plant available N supply if plants can access even small portions of the total gross ammonification flux. Additionally, if diversified cropping systems favor high gross N fluxes while maintaining small soil inorganic N pools, the soil N supply may be more resilient to loss, as the high fluxes could readily replenish the small pool as it is gradually depleted by N consumption processes (e.g. plant N uptake, microbial immobilization, N losses to the environment). Thus, gross ammonification may play a key role in a more efficient crop N supply strategy. Nonetheless, despite its potential importance to the supply of plant available N, relatively little research has explored gross ammonification in diversified cropping systems (but see Burger and Jackson, 2003).

In addition to the quantity of the plant available N, timing of N availability may be an important aspect of how cropping systems affect crop productivity (Kramer et al., 2002). Grain crops such as corn (*Zea mays*) respond to N limitation by senescence of leaves and a decrease in photosynthetic capacity in the latter part of the growing season (Wolfe et al., 1988; Dwyer et al., 1995). The crop's ability to delay senescence and maintain photosynthetic capacity late in the growing season (deemed the 'stay-green' effect) can be an important yield determining factor in corn (Duvick and Cassman, 1999; Subedi and Ma, 2005b). As a greater and more extended soil N supply can enhance the "stay-green" effect (Wolfe et al., 1988; Subedi and Ma, 2005a), diversified cropping systems may be able to supply additional N to crops late in the growing season through enhanced mineralization of organic N.

In a recent investigation of gross ammonification in a long-term cropping systems experiment in the US Midwest we observed that diversified cropping systems enhanced crop N uptake, but did not enhance gross ammonification at a single time point in late July (Osterholz et al., 2017b). While this study highlighted the large potential for gross ammonification to contribute to crop N uptake (gross ammonification exceeded maize N uptake by > 500%), but high spatial variability limited comparisons between cropping systems. The study also revealed that a more rigorous investigation of soil N cycling processes and crop N dynamics at multiple time points is needed to better characterize linkages between soil N measurements and crop responses.

The present study assessed the importance of soil N cycling processes for determining the diversification effect at a long-term cropping systems experiment. We utilize several measurements of gross ammonification, PMN, soil inorganic N, and crop N to examine the relationships between cropping system diversity, soil N dynamics, and crop N uptake. The relationships were further explored after accounting for the influence of spatially variable concentrations of SOC. Our study examined three specific hypotheses: (1) gross and net N mineralization rates will be greater in diversified cropping systems while inorganic N pools will be smaller; (2) diversified cropping systems enhance corn N uptake and yield despite lower inorganic N inputs; and (3) differences in corn N uptake among the cropping systems can be explained by soil N mineralization, particularly late in the growing season.

## 2. Methods

### 2.1. Experiment description

These hypotheses were tested by comparing measurements of corn N uptake and soil N dynamics in a long-term cropping systems experiment in Iowa, USA, at Iowa State University's Marsden Farm. The

experiment, which contains three cropping systems differing in crop rotation complexity and fertility sources, was designed to compare both specialized grain production systems and integrated crop-livestock production systems (Liebman et al., 2008; Davis et al., 2012). The experiment was established in 2002 in Boone County in central Iowa, USA (42°01' N, 93°47' W). Soils at the 9-ha experimental site consist of a catena of moderately-well drained to poorly drained loam and clay-loam Mollisols that is among the most important agricultural soil associations in Iowa. Dominant soil types at the site were Clarion loam (fine-loamy, mixed, superactive, mesic, Typic Hapludoll), Nicollet loam (fine-loamy, mixed, superactive, mesic, Aquic Hapludoll) and Webster silty clay loam (fine-loamy, mixed, superactive, mesic, Typic Endoaquoll), with smaller areas of Harps loam (fine-loamy, mixed, superactive, mesic Typic Calciaquoll), and Canisteo silty clay loam (fine-loamy, mixed, superactive, calcareous, mesic Typic Endoaquoll). The experiment was blocked to account for soil differences caused by the catena. However, plots within blocks differed substantially in SOC concentration due to fine scale topographic variability over the experimental site (data not shown). Only a small amount of subsurface tile drainage is present at the experiment site.

The crop rotations present were a short 2-year corn-soybean (*Glycine max*) rotation, and two more extended rotation systems: a 3-year corn-soybean-oat (*Avena sativa*)/red clover (*Trifolium pratense*) rotation and a 4-year corn-soybean-oat/alfalfa (*Medicago sativa*)-alfalfa rotation. The oat crop was undersown with the forage legumes in the extended 3-year and 4-year rotations. The experimental design was a randomized complete block design with four replicates, and all crops within each of the rotations were present in each replicate block in each year of the study. Experiment plots measured 18 m × 84 m.

All cropping systems received both pre-planting and supplemental in-season fertilization using N sources relevant to the cropping system, and the strategy used in each system sought to achieve high corn yields while minimizing over-fertilization. In addition to contributions of N from forage legumes, the two diversified cropping systems also received composted manure in order to represent cropping systems that integrate crop and livestock production. The 2-year system received 112 kg N ha<sup>-1</sup> as urea or urea-ammonium-nitrate fertilizer during the corn phase at corn planting. In the 3- and 4-year systems, forage legume residues were incorporated in late fall preceding corn planting. Composted cattle manure was also applied to the 3- and 4-year systems in the fall preceding corn planting. Over the course of the experiment (2003–2014) an average of 8.6 Mg dry weight ha<sup>-1</sup> of composted manure was applied prior to corn, with an average total N content of 107 kg N ha<sup>-1</sup>. Approximately 16 kg N ha<sup>-1</sup> was available to the subsequent corn crop from the composted manure, assuming 15% N availability (Eghball and Power, 1999). In late spring of the corn year in all cropping systems, soil testing was used to determine application rates of supplemental inorganic N fertilizer. Following a well-established adaptive management program based on soil  $\text{NO}_3^-$  pool size, soil  $\text{NO}_3^-$  was measured from soil samples collected from 0 to 30 cm depth when corn plants developed five to six collared leaves (Magdoff, 1991; Blackmer et al., 1997). In the 2-year cropping system, supplemental fertilizer application was triggered when the soil  $\text{NO}_3^-$  concentration dropped below a threshold of 22–25 mg  $\text{NO}_3^-$ -N kg<sup>-1</sup> soil. The threshold was lower (15–18 mg  $\text{NO}_3^-$ -N kg<sup>-1</sup> soil) in the diversified cropping systems as they received manure and legume N inputs. The lower threshold values were used in years when April–May rainfall was more than 20% greater than the 30-year average. Supplemental fertilization rates were calculated from the difference between the measured soil  $\text{NO}_3^-$  pool and the critical threshold. Supplemental N fertilizer as urea ammonium nitrate (32% liquid N) was applied at recommended rates (2003–2014 averages of 46, 31, and 21 kg N ha<sup>-1</sup> in the 2-, 3-, and 4-year systems, respectively) between May 29 and June 28. Additional management details for the 2013 and 2014 growing seasons can be found in Table 1.

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