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Potential Soil Nitrogen Mineralization in Biofuel Feedstock Crops with Varying Management Intensity

By

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ABSTRACT

In order to test whether or not *in situ* ion-exchange resin strips can be used as a successful indicator of soil nitrogen availability as compared to the wide-spread laboratory potential mineralization method, nine plots in the Great Lakes Bioenergy Research Center located in Hickory Corners, MI were tested using both methods. The nine plots included several different crops systems with different management intensities, such as: continuous corn, continuous corn and cover crop, rotational soybean with a cover crop, rotational corn with a cover crop, switchgrass, hybrid poplar, mixed-species native grasses, successional vegetation, and restored prairie. The two methods compared were the use of ion-exchange resin strips incubated in the field plots for 28 days, and lab incubation of extracted soil cores. The results found that the use of ion-exchange resin strips were an accurate measurement of actual nitrogen flow through the system, as opposed to the potential nitrogen mineralization through the laboratory procedure. The stated hypothesis was proved correct, that soils with more intensive management, such as fertilizer, cover crops, and rotational cropping, will have more nitrogen available for plants and higher nitrogen mineralization rates than unmanaged soils. The strips have been credited to being an accurate representation of real root-soil interactions in the field, which has many potential future benefits for ecological soil studies.

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ACKNOWLEDGEMENTS

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I would also thank to express my appreciation to my project advisor and Environmental Science Division Head Dr. Briggs for agreeing to supervise my writing, and always being a source of knowledge and guidance throughout the four years of my undergraduate studies.

INTRODUCTION

The Great Lakes Bioenergy Research Center (GLBRC) is a part of the Long Term Ecological Research site at the Kellogg Biological Station (KBS) in Michigan. The research at GLBRC is focused on determining how to best grow biofuels under a variety of environmental conditions and management. At this specialized field site, numerous experiments are in place to measure the viability of these crops, as well as their sustainability and impact on the environment and the economy. The 2013 GLBRC site featured several different systems in a randomized complete block design (5 replicate blocks of 30x40m plots) that included the following crops: continuous corn, continuous corn and cover crop, rotational soybean with a cover crop, rotational corn with a cover crop, switchgrass , hybrid poplar, mixed-species native grasses, successional vegetation, and restored prairie (GLBRC, 2014). These crops require different levels of management intensity and chemical inputs to successfully grow and harvest, which include nitrogen-based fertilizer and the implementation of cover crops (Gelfand et al., 2010). These different input levels can affect the nutrient availability in the soils, such as nitrogen.

Nitrogen is an important macronutrient required by plants for essential growth and productivity. The majority of nitrogen found in soil environment is in organic forms unavailable for uptake by the majority of higher plants (Deenik, 2006). The other inorganic forms of nitrogen, such as ammonium and nitrates, are readily accessible for plants. Organic nitrogen needs to be converted into inorganic form before it can be easily accessed by plants through a process known as nitrogen mineralization (Crohn, 2004).

Mineralization occurs when the organic nitrogen in soil organic matter is converted into inorganic forms usable by plants, through the activities of soil microbes (Deenik, 2006). The rate at which this procedure is undertaken is known as the mineralization rate, and is an extremely useful tool for determining Nitrogen availability (Simmons et al., 1995). In order to effectively manage for higher crop yield and productivity, accurately estimating the available nitrogen in soil is critical for understanding the soil's relative fertility (Crohn, 2004). This information can then be used to undertake the necessary management intensities, such as fertilization.

Potential soil nitrogen mineralization is typically measured by taking soil core samples from the field sites and incubating them inside a laboratory, before determining mineralization. This technique is a reliable method for comparing different soils under standard conditions, but it has drawbacks by not allowing the full dynamic nature of the process. For example, it typically causes great disturbances to the soil, and results in static extractions that do not account for kinetic release and transport of nutrients (Dobermann et al., 1994). An alternate procedure to this method involves placing ion resin strips in the field site, and analyzing the nitrogen availability results after one month of incubating in the plot itself (Schoenau and Qian, 2005). This method would ensure to measure the relative amount of all soluble nitrogen in an area, and not just the potential mineralization results obtained from laboratory incubation. The ion resin strip method is based on measuring the relative amount of nutrients in the soil available to plants, and the rate they are released from the soil organic matter (Dobermann et al., 1994). The cation and anion exchange membranes present on each positive and negatively charged strip act as ion sinks or dynamic exchangers and the nutrients present in the soil colloids or solution become adsorbed (Schoenau and Qian, 2005). By placing the ion exchange resin strips in direct contact in the soil, this method has been proven to be an effective way to determine relative ammonium and nitrate release rates by mineralization (Schoenau and Qian, 2005).

The objective of this paper is to examine whether or not *in situ* ion-exchange resin strips can be used as an indicator of soil nitrogen availability as compared to the potential mineralization used in the laboratory. We hypothesized that soils with more intensive management, such as fertilizer, cover crops, and rotational cropping, will have a positive effect on the soil nitrogen availability, and therefore the systems with these management techniques will have higher nitrogen mineralization rates than systems without. To test this question we assessed nitrogen availability in nine replicated agroecosystems with different management gradients ranging from high to low. The two methods used were standard lab nitrogen mineralization essay vs. *in situ* ion-exchange resin strips in the field.

METHODS

The plots utilized in this experiment were located in the first block of the GLBRC and included continuous corn (CC), Continuous Corn with a cover crop (CC+cc), rotational soybean with a cover crop (S+cc), rotational corn with a cover crop (C+cc), switchgrass: *Panicum virgatum* variety Cave-in-rock (SG), native grass mix (NG), hybridized Poplar: *Populus nigra* x *Populous maximowiczii* (POP), old field (OF), and native prairie (NP).

Field Incubation

The ion-exchange resin sheets with cation or anion exchange capacity were cut into 2.5 x 10 cm strips, and hole punched with star shape for positive, and a rectangle for negative. The strips were placed in separate plastic tubs filled with 0.5 M HCl, and shaken at 40 rpm (revolving per minute) for one hour. After the first round of shaking, the strips were removed from the HCl and soaked in 0.5 M NaHCO3 and shaken for five hours at 40 rpm. The solution was changed every hour and discarded. After five hours, the strips were rinsed with deionized water and placed into separate anion and cation Ziploc bags with enough deionized water to keep the strips moist. Stainless steel mesh was cut to fold over each resin strip with half an inch of extra steel on each end (11 x 2.5 cm). The mesh cages were then folded over each strip and stapled together.

The mesh-covered strips were randomly placed in each of the 30x40m replicated plots in the field by creating two vertical slits about 10 cm into the soil, inserting the strips, and then closing the openings with a hand shovel. Each plot had one cation and one anion strip approximately one inch apart, and the exact location was marked with a plastic flag.

In order to compare and check for interference of the stainless steel covering, two mesh-covered strips and two uncovered strips were also placed into undisturbed soil cores (1.9 cm diameter cylinder wet tip hand probe) and kept in laboratory under room temperature relatively moist in accordance with natural weather patterns. The strips were then left in each plot or sample for 28 days.

After 28 days, the strips were extracted from the soil with metal pliers, brushed for excess dirt particles, and placed in separate labeled plastic bags for each plot. In the lab, an anion and cation strips for each plot were placed in individual 237 mL specimen cups with a squirt of deionized water. The cups were vigorously shaken by hand, and then the water was drained to remove excess soil. This process was repeated until there was not more obvious soil left in each cup. After shaking, 35mL of 2.0 M KCl was added to each cup and placed on a shaker for one hour at 40 rpm. After shaking, a syringe was used to filter the extract with a Type A/E 1 µm pore size, glass fiber filter. A labeled scintillation vial was rinsed with extract and then filled up with enough extract to allow expansion of the solution in the freezer (-7 °C) for storage until processing. The used strips can then be regenerated later and re-used for additional experiments.

Mineralization was determined by the following equation:

N Adsorbed = [(conc in µg N per mL) × 70 mL of KCl)] /
(50.8 cm^2 area of the strip × % strip remaining × number of days in the ground)

Nitrogen Mineralization Potential in Laboratory Incubation

Forty grams (0-10 cm depth) of fresh soil samples collected from the replicated plots were sieved and weighed into a tin for determining gravimetric moisture content using the protocol outlined by GLBRC (GLBRC, 2014). Soil water-filled pore space of each sample was determined by the following equation:

$$WFP = [Pw \times (Db/St)] \times 100$$

where:
Pw = water content [(g water/g dry soil) × 100]
Db = bulk density; use 1.46 g/cm^{-3} (mean bulk density in surface soils)
St = total porosity; use 45 at KBS and 50% at Arlington (St = [1-(Db/particle density)]*100, assume mineral particle density of 2.65 g cm^{-3})

The amount of water needed to raise the WFP to 60% in each sample was determined by the following equation:

$$\text{Target g of water} = 0.1875 \times \text{dry wt of remaining soil}$$

where:
dry wt of remaining soil = fresh wt of remaining soil – fresh wt of remaining soil x (g water in subsample/fresh wt of subsample)
mL of water to add = (target g of water) – (g of water in remaining soil)
where g of water in remaining soil = fresh wt – dry wt

The determined amount of deionized water was added to each sample and mixed to evenly distribute. Ten grams of each soil were weighed into six specimen cups, and three cups were set aside for nutrient extraction with 2M KCl using the protocol outlined by the Great Lake Bioenergy Research Center (GLBRC, 2014). The remaining three samples were capped loosely and placed in a dark room with constant temperature. A sub-set of the containers were re-weighed each week and if the weight decreased by >10%, water was added to bring it back up to 10 mL. After 28 days the incubated soil samples were extracted using a 2M KCl solution, using the same protocol outlined by the GLBRC (GLBRC, 2014).

The extracts were analyzed for NO3- and NH4+ and corrected for area and gravimetric water content using the protocol outlined by the Great Lake Bioenergy Research Center (GLBRC, 2014). Mineralization was determined using the following equations:

$$\text{N mineralized} = [(\text{Nitrate}T_{28} + \text{Ammonium}T_{28})-(\text{Nitrate}T_{0} + \text{Ammonium}T_{0})]/28 \text{ days}$$

where $\text{Nitrate}T_{28} = NO_3^-$ concentration at the end of the 28-day incubation
$\text{Ammonium}T_{28} = NH_4^+$ concentration at the end of the 28-day incubation
$\text{Nitrate}T_{0} = NO_3^-$ concentration at the beginning of the incubation
$\text{Ammonium}T_{0} = NH_4^+$ concentration at the beginning of the incubation

RESULTS

The results for the field mineralization showed the largest rate for continuous corn with a cover crop (G2) with 19.5 μg N/cm^2/day and the smallest rate in the native prairie (G10) at 1.4 μg N/cm^2/day (Figure 1). All of the plots with fertilizer treatment, cover crops, or rotational cropping are among the highest values of field mineralization, such as continuous corn with a cover crop (G2), rotational corn with a cover crop (G4), and soybean and a cover crop (G3) (Figure 1). The rest of the plots show very low mineralization rates, such as switchgrass (G5), native grass mix (G7), poplar (G8), native prairie (G10), and old field (G9) (Figure 1). The results of the potential mineralization from the lab also showed the largest value for continuous corn with a cover crop (G2) with 0.94 μg N/g/day, and the smallest value was the native prairie (G10) with 0.16 μg N/g/day (Figure 2).

The results for the regression of lab and field mineralization show a strong positive linear regression with $r^2=0.6587$ (Figure 3).

The differences between the covered and uncovered strips show relatively small differences between the two methods, with covered strips showing higher mineralization values first two plots, and uncovered showing higher mineralization values for the second two plots (Figure 4). The regression for the uncovered and covered strip values showed a very strong positive linear regression with a value of $r^2=0.8295$ (Figure 5).

Plot	Crop Type	Abbreviation
G1	Continuous Corn	CC
G2	Continuous Corn + cover crops	CC+cc
G3	Soybean + cover crops	S+cc
G4	Corn + cover crops	C+cc
G5	Switchgrass	SG
G7	Native Grass Mix	NG
G8	Poplar	P
G9	Old Field	OF
G10	Native Prairie	NP

Table 1. Legend of treatments used in the study

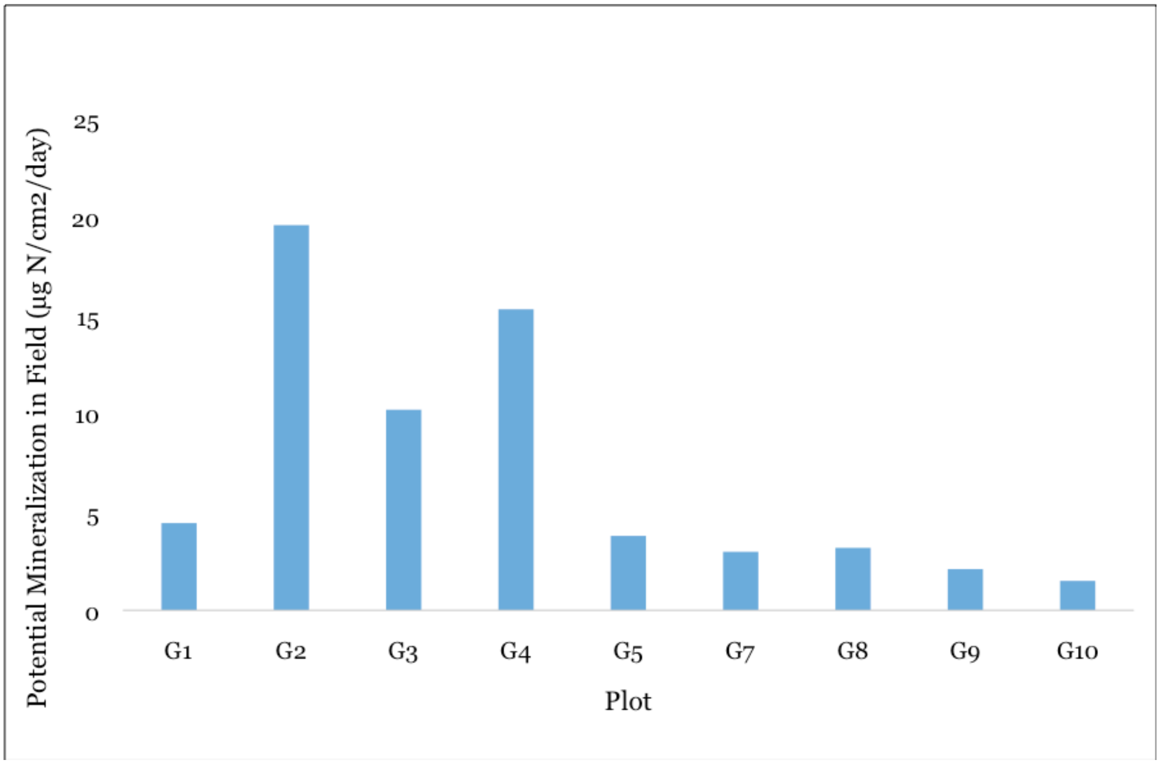

Figure 1. Nitrogen mineralization rates from ion exchange resin strips in the field

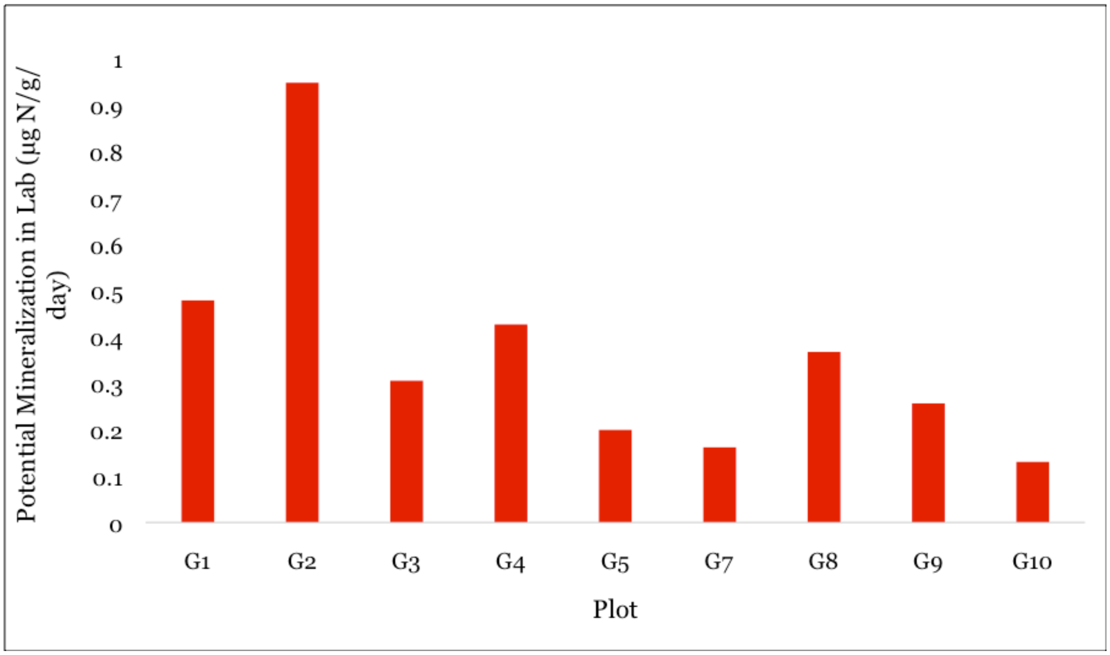

Figure 2. Potential nitrogen mineralization rates from lab incubation

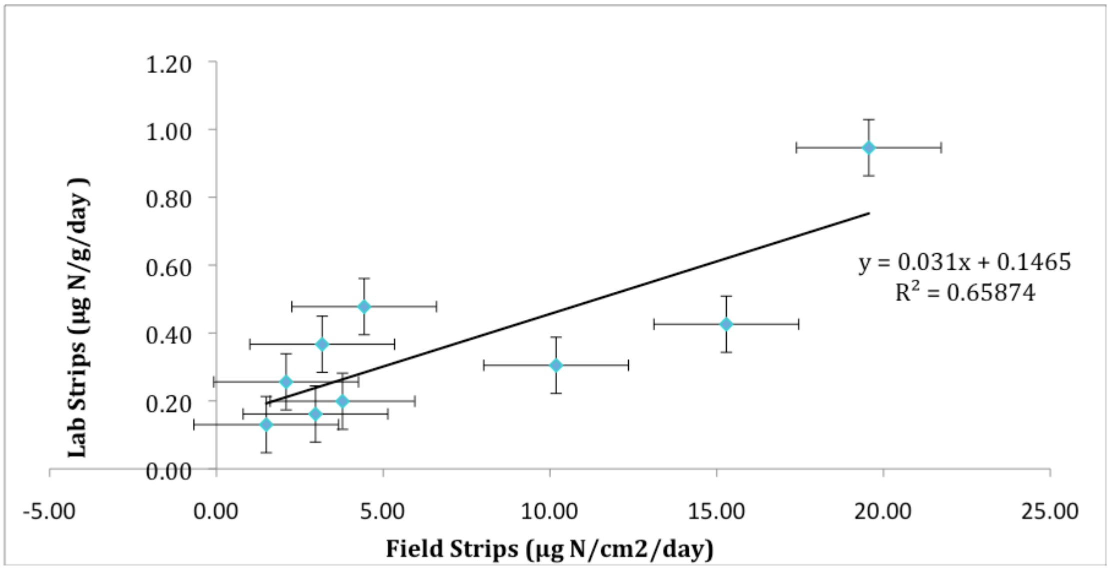

Figure 3. Regression for field mineralization vs. potential nitrogen mineralization rates from lab incubation

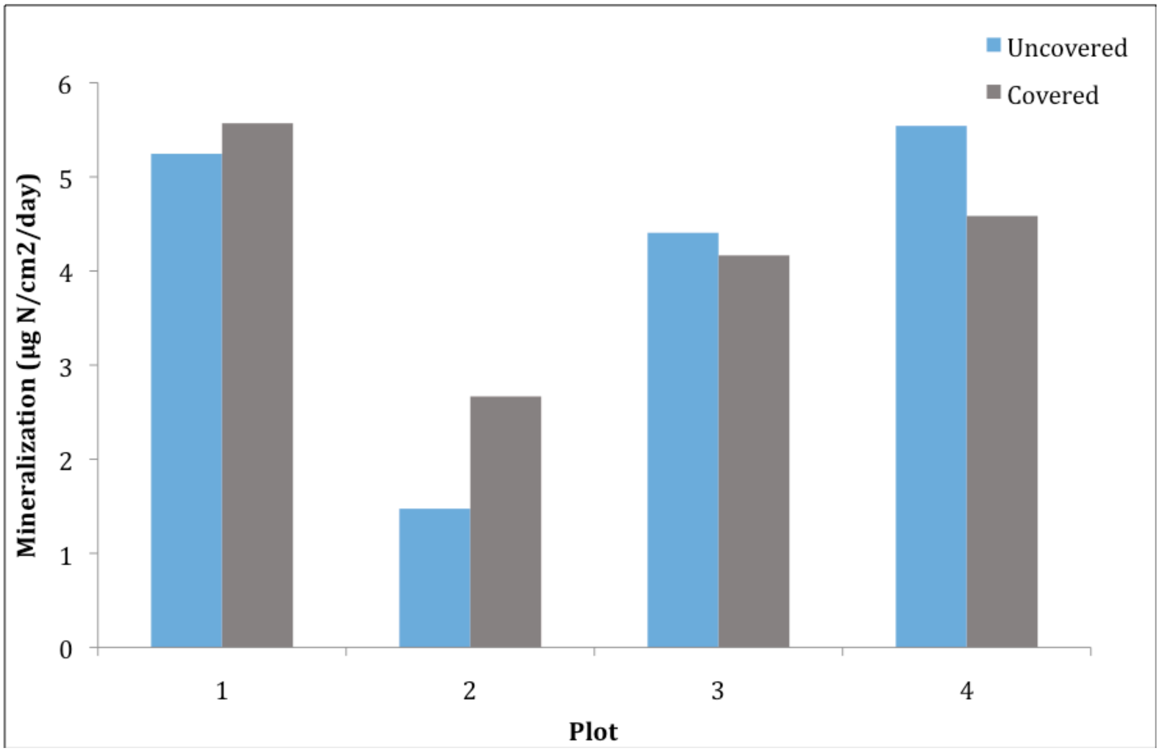

Figure 4. Comparison of mineralization rates of mesh covered strips vs. uncovered strips

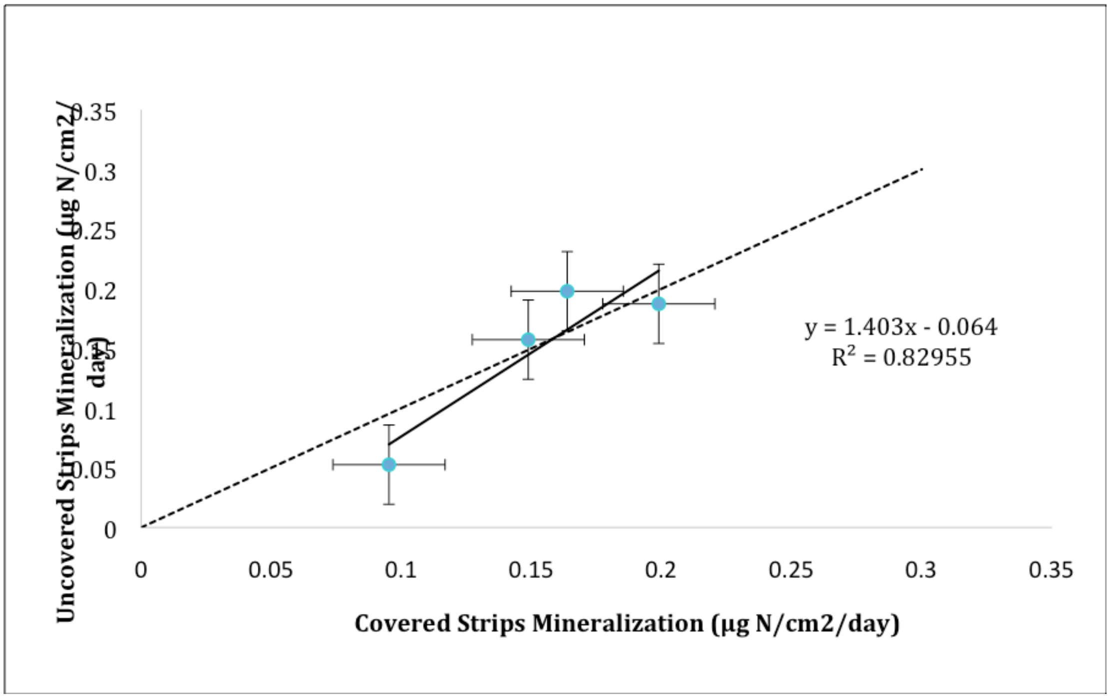

Figure 5. Regression line for mineralization rates of mesh covered strips vs. uncovered strips

DISCUSSION

The results supported the hypothesis that the soils with more intensive management, such as fertilizer, cover crops, and rotational cropping, will have more nitrogen available for plants and higher nitrogen mineralization rates than unmanaged soils. The plots with more intensive management, CC, CC+cc, S+cc, and C+cc, had higher levels of nitrogen mineralization (Figures 1 and 2). This management included the use of cover crops, fertilizer, and rotational cropping. Continuous corn (CC) was the only plot where there were several applications of nitrogen-based fertilizer with ammonium sulfate. These management applications have been known to increase the amount of available soil nitrogen (Veldkamp and Keller, 1997).

The results for the uncovered and covered ion-exchange resin strips also showed that there was only a small difference between the mineralization rate of strips with mesh cover, and strips without (Figure 5). The stainless steel mesh covering the strips was unique to this experiment to try and deter rodents eating part of the strips. The strong positive linear regression found in Figure 5 demonstrates that the mesh coverings can effectively be used in the future to protect the strips from any further rodent damage. There is a need for better validation of the potential interference of the stainless mesh covering, but the relatively high correlation between the two types (Figure 3) show that they can potentially be used interchangeable in the future.

Overall, the mineralization rates measured by the ion strips in the field were considerably higher than the values for the lab method (Figures 3). This result is not surprising due to the fact that the strips in the field are able to experience the full range of environmental conditions, including temperature and lateral water flow through the soil. Due to this distinction, the strips in the field are able to relay the relative amount of all soluble nitrogen, as opposed to the lab method, which can only estimate potential mineralization based on available nitrogen in the sample at the time of extraction. This results in a much more comprehensive mineralization value capable of assessing potential nutrient changes in a system (Qian and Schoenau, 1996). Additionally, even though the scale for the lab and field results are different, there is a clear distinction between the two methods where the higher the potential mineralization shown on figure 3 corresponds with a higher amount of field *in situ* mineralization figure 2. This finding provides further foundation for the use of ion exchange resin strips to accurately determine nitrogen mineralization.

The differences in mineralization results between the separate agroecosystems stemmed mainly from the management including crop rotations and the implementation of cover crops. The plots with the highest mineralization values were CC+cc, and C+cc. These two plots are significant because they both contained cover crops, which are utilized mainly as a form of carbon sequestration and to restore soils of depleted nutrients between crop plantings (University of Minnesota Cooperate Extension, 2013). The higher mineralization rates found in these plots could have been a result of the ability of the cover crops to replenish lost nutrients, such as N, to the soil after harvest. Additionally, C+cc not only utilized cover crops but also was part of a yearly rotation between corn and soybean plants. Crop rotations have also been used as a management tool to replenish soil nutrients after each harvest. The use of soybean as the alternative rotational crop is especially useful, as nitrogen-fixing legumes specifically increases the amount of organic N stored in the soil organic matter (University of Minnesota Cooperate Extension, 2013). This information was consistent with the results found, as the three plots that utilized cover crops and/or rotational plantings all had the highest values for mineralization (Figures 2 and 3).

The plots G5-10 utilized in this study experienced no management techniques such as fertilization, cover crops, or rotations and all expressed relatively low measures of mineralization. This corresponds with a study by Campbell et al. that found that there was a considerable decline in the soil organic nitrogen content and overall N mineralization potential in fallow-based cropping systems (Campbell et al., 1990 as referenced by Schoenau, 1996). This could contribute to the explanation as to why those low-movement intensity plots had the lowest mineralization values.

A similar study by Camill et al., 2004 monitored N mineralization rates in a large-scale prairie restoration in southern Minnesota and found that rates decreased in fields three years or older. They found that a rise in warm-seasons grasses by the third growing season had increased litter rates and C mineralization, but decreased N mineralization (Camill et al., 2004). Their reasoning was that these grasses allocated a greater amount of biomass underground, which could have led to increased fine foot turnover and labile C source in the root zone. They theorize that this increase in C sources could have increased microbial demand for nitrogen (Camill et al., 2004). Their findings are comparable to the lower nitrogen mineralization rates found in the grass systems (SG, MG, NG, POP, OF, NG, NP) utilized in this study. Since the GLBRC is part of a long term ecological research site, these grass systems have been in place for the past 5-6 years, so the nutrient availability would be much less than in the first stages of planting. When the biomass is eventually harvested for biofuel feedstock and the plot is replanted, the nitrogen mineralization rates are bound to recover to higher levels (Camill et al., 2004).

CONCLUSION

The ability to accurately measure nitrogen mineralization in soils is of considerable importance for effectively assessing the nutrient availability of agricultural lands (Schomberg et al., 2009). In response to these estimates, the proper management techniques, such as the appropriate amount of fertilization, can be implemented to avoid excess runoff and nitrate leaching into nearby waterways (Schomberg et al., 2009). This information is not only helpful for maximizing crop productivity in biofuels, but also estimating changes in nutrient availability in response to the changing climate (Simmons et al., 1995). The effects of this study show that the ion-exchange resin strips are a useful indicator of nitrogen mineralization in comparison to the more common laboratory incubation method, and supported the original hypothesis that higher management intensity resulted in higher mineralization rates. The widespread use of ion-exchange strips could result in the increase in mineralization studies, as this method is a unique approach to assessing soil nutrient availability by mimicking what a real plant root would experience. This method could allow numerous agricultural and ecological benefits for the measurement of ions independent to soil type (Qian and Schoenau, 1996).

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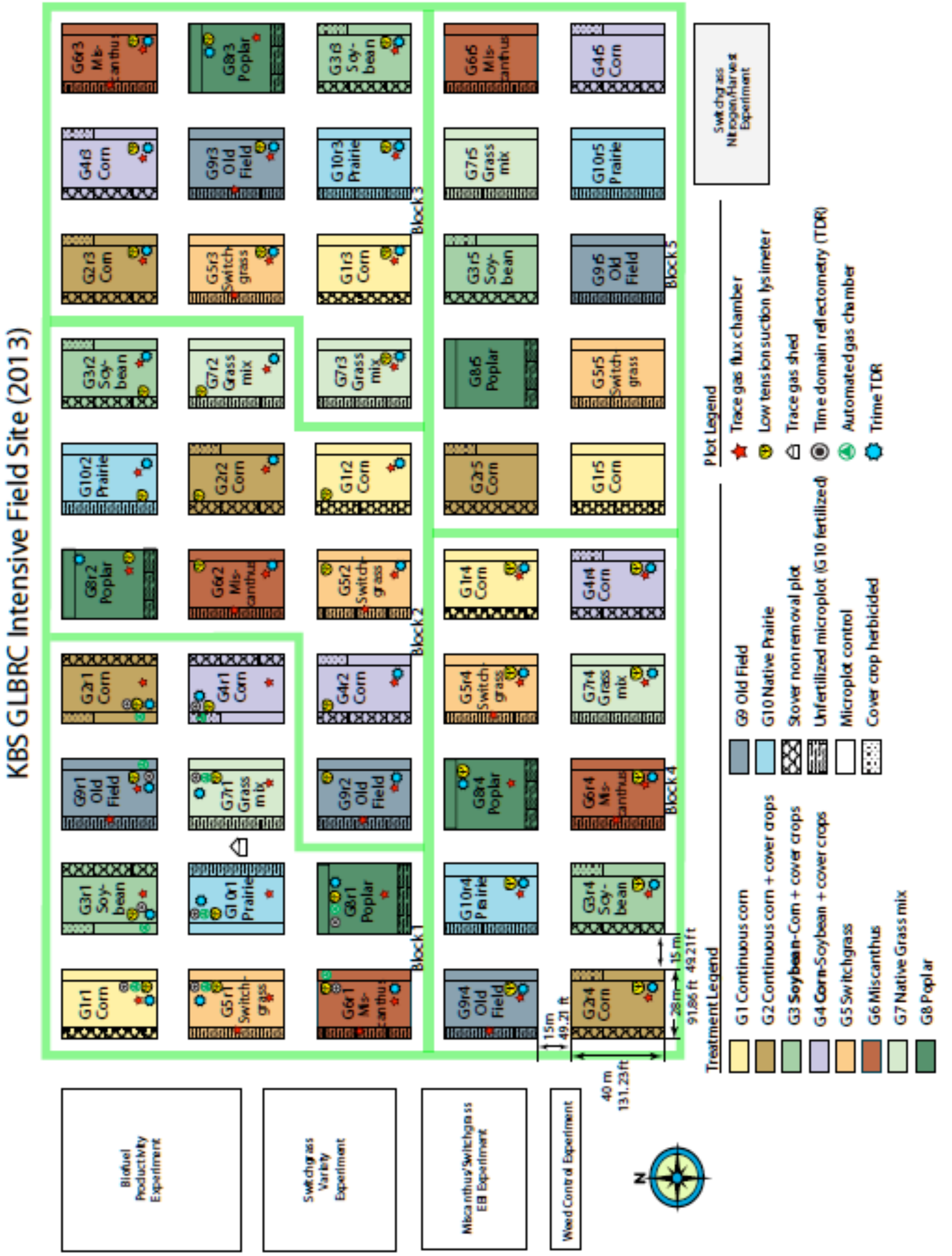
APPENDIX

A. Map of Site



B. Data

Plot	Strip Results (µg N $cm^{-2}day^{-1}$)	Lab Results (µg N/g/day)
G1	4.4296	0.47775
G2	19.5595	0.946083333
G3	10.1842	0.304916667
G4	15.2874	0.426
G5	3.77975	0.199083333
G7	2.9698	0.161583333
G8	3.17075	0.366916667
G9	2.0894	0.256166667
G10	1.49325	0.13025

Uncovered Strips (ug/cm^2/day)	Covered Strips (ug/cm^2/day)
5.246	5.57
1.474	2.667
4.406	4.166
5.542	4.585

Name	NH4 (ppm)	NO3 (ppm)	MIN	rep	plot
G1-1	0.098	3.667	3.765	1	G1
G1-2	1.079	4.447	5.526	2	G1
G1-3	0.143	7.7	7.843	3	G1
G1-4	0.885	1.516	2.401	4	G1
G1-5	0.474	2.139	2.613	5	G1
G2-1	0.101	7.837	7.938	1	G2
G2-2	0.284	15.051	15.335	2	G2
G2-3	0.327	43.405	43.732	3	G2
G2-5	0.085	11.148	11.233	4	G2
G3-1	0.137	11.026	11.163	1	G3
G3-2	0.145	10.374	10.519	2	G3
G3-3	0.176	11.602	11.778	3	G3
G3-4	0.231	12.525	12.756	4	G3
G3-5	0.354	4.351	4.705	5	G3
G4-1	0.342	59.134	59.476	1	G4
G4-2	0.569	5.191	5.76	2	G4
G4-3	0.591	3.953	4.544	3	G4
G4-4	0.239	4.286	4.525	4	G4
G4-5	0.191	1.941	2.132	5	G4

G5-1	0.452	4.111	4.563	1	G5
G5-3	0.493	0.558	1.051	2	G5
G5-4	0.181	8.77	8.951	3	G5
G5-5	-0.012	0.566	0.554	4	G5
G6-1	0.395	12.668	13.063	1	G6
G7-1	0.117	3.629	3.746	1	G7
G7-2	0.076	5.292	5.368	2	G7
G7-3	0.706	1.351	2.057	3	G7
G7-4	0.037	3.265	3.302	4	G7
G7-5	0.253	0.123	0.376	5	G7
G8-1	0.008	1.806	1.814	1	G8
G8-2	-0.001	2.556	2.555	2	G8
G8-3	0.358	1.541	1.899	3	G8
G8-4	0.018	6.397	6.415	4	G8
G9-1	0.044	5.671	5.715	1	G9
G9-2	0.084	2.095	2.179	2	G9
G9-3	0.196	0.254	0.45	3	G9
G9-4	0.085	0.137	0.222	4	G9
G9-5	0.16	1.721	1.881	5	G9
G10-1	0.205	0.203	0.408	1	G10
G10-2	2.245	0.775	3.02	2	G10
G10-4	0.445	0.051	0.496	3	G10
G10-5	1.955	0.094	2.049	4	G10
t1	0.02	5.226	5.246		
t2	-0.025	1.499	1.474		
t3	0.058	4.348	4.406		
t4	0.021	5.521	5.542		
O-1	-0.024	5.594	5.57		
O-2	0.096	2.571	2.667		
O-3	0.416	3.75	4.166		
O-4	0.116	4.469	4.585		

Name	NH4 (ppm)	NO3 (ppm)	deployment days	% area remaining	NH4 N Adsorbed	NO3 N adsorbed	Min
G1-1	0.098	3.667	28	100	4.82283E-05	0.001804626	0.001852854
G1-2	1.079	4.447	28	100	0.000531004	0.002188484	0.002719488
G1-3	0.143	7.7	28	100	7.0374E-05	0.00378937	0.003859744
G1-4	0.885	1.516	28	100	0.000435531	0.000746063	0.001181594
G1-5	0.474	2.139	28	100	0.000233268	0.001052657	0.001285925
G2-1	0.101	7.837	28	100	4.97047E-05	0.003856791	0.003906496
G2-2	0.284	15.051	28	100	0.000139764	0.007406988	0.007546752

G2-3	0.327	43.405	28	100	0.000160925	0.021360728	0.021521654
G2-5	0.085	11.148	28	100	4.18307E-05	0.00548622	0.005528051
G3-1	0.137	11.026	28	100	6.74213E-05	0.005426181	0.005493602
G3-2	0.145	10.374	28	100	7.13583E-05	0.005105315	0.005176673
G3-3	0.176	11.602	28	100	8.66142E-05	0.005709646	0.00579626
G3-4	0.231	12.525	28	100	0.000113681	0.006163878	0.006277559
G3-5	0.354	4.351	28	100	0.000174213	0.00214124	0.002315453
G4-1	0.342	59.134	28	100	0.000168307	0.029101378	0.029269685
G4-2	0.569	5.191	28	100	0.00028002	0.002554626	0.002834646
G4-3	0.591	3.953	28	100	0.000290846	0.001945374	0.00223622
G4-4	0.239	4.286	28	100	0.000117618	0.002109252	0.00222687
G4-5	0.191	1.941	28	100	9.39961E-05	0.000955217	0.001049213
G5-1	0.452	4.111	28	100	0.000222441	0.00202313	0.002245571
G5-3	0.493	0.558	28	100	0.000242618	0.000274606	0.000517224
G5-4	0.181	8.77	28	100	8.90748E-05	0.004315945	0.00440502
G5-5	-0.012	0.566	28	100	-5.90551E-06	0.000278543	0.000272638
G6-1	0.395	12.668	28	100	0.00019439	0.006234252	0.006428642
G7-1	0.117	3.629	28	100	5.75787E-05	0.001785925	0.001843504
G7-2	0.076	5.292	28	100	3.74016E-05	0.002604331	0.002641732
G7-3	0.706	1.351	28	100	0.000347441	0.000664862	0.001012303
G7-4	0.037	3.265	28	100	1.82087E-05	0.001606791	0.001625
G7-5	0.253	0.123	28	100	0.000124508	6.05315E-05	0.000185039
G8-1	0.008	1.806	28	100	3.93701E-06	0.00088878	0.000892717
G8-2	-0.001	2.556	28	100	-4.92126E-07	0.001257874	0.001257382
G8-3	0.358	1.541	28	100	0.000176181	0.000758366	0.000934547
G8-4	0.018	6.397	28	100	8.85827E-06	0.00314813	0.003156988
G9-1	0.044	5.671	28	100	2.16535E-05	0.002790846	0.0028125
G9-2	0.084	2.095	28	100	4.13386E-05	0.001031004	0.001072343
G9-3	0.196	0.254	28	100	9.64567E-05	0.000125	0.000221457
G9-4	0.085	0.137	28	100	4.18307E-05	6.74213E-05	0.000109252
G9-5	0.16	1.721	28	100	7.87402E-05	0.000846949	0.000925689
G10-1	0.205	0.203	28	100	0.000100886	9.99016E-05	0.000200787
G10-2	2.245	0.775	28	100	0.001104823	0.000381398	0.00148622
G10-4	0.445	0.051	28	100	0.000218996	2.50984E-05	0.000244094
G10-5	1.955	0.094	28	100	0.000962106	4.62598E-05	0.001008366
t1	0.02	5.226	28	100	9.84252E-06	0.00257185	0.002581693
t2	-	1.499	28	100	-1.23031E-	0.000737697	0.000725394

	0.025				05		
t3	0.058	4.348	28	100	2.85433E-05	0.002139764	0.002168307
t4	0.021	5.521	28	100	1.03346E-05	0.002717028	0.002727362
O-1	-0.024	5.594	28	100	-1.1811E-05	0.002752953	0.002741142
O-2	0.096	2.571	28	100	4.72441E-05	0.001265256	0.0013125
O-3	0.416	3.75	28	100	0.000204724	0.001845472	0.002050197
O-4	0.116	4.469	28	100	5.70866E-05	0.002199311	0.002256398

treatment	initial_no3	final_no3	initial_nh4	final_nh4	days	net_nit	net_min	net_amm
G1	13.3	34.1	6.5	0.8	28	0.7	0.5	-0.2
G1	13.4	34.1	6.5	0.8	28	0.7	0.5	-0.2
G1	21.1	42.9	9.9	3.8	28	0.8	0.6	-0.2
G1	21.1	42.9	9.9	3.8	28	0.8	0.6	-0.2
G1	19.3	53.6	22.8	1.0	28	1.2	0.4	-0.8
G1	19.2	53.6	22.6	1.0	28	1.2	0.5	-0.8
G1	13.2	28.3	6.5	0.5	28	0.5	0.3	-0.2
G1	13.2	28.3	6.5	0.5	28	0.5	0.3	-0.2
G1	7.1	37.3	6.1	1.8	28	1.1	0.9	-0.2
G1	6.8	37.3	5.9	1.8	28	1.1	0.9	-0.1
G1	16.3	25.4	8.9	0.7	28	0.3	0.0	-0.3
G1	15.4	25.4	8.5	0.7	28	0.4	0.1	-0.3
G10	2.4	7.3	2.5	2.3	28	0.2	0.2	0.0
G10	2.4	7.3	2.5	2.3	28	0.2	0.2	0.0
G10	2.9	14.5	4.4	2.8	28	0.4	0.4	-0.1
G10	2.9	14.5	4.3	2.8	28	0.4	0.4	-0.1
G10	1.7	6.8	2.3	2.5	28	0.2	0.2	0.0
G10	1.7	6.8	2.3	2.5	28	0.2	0.2	0.0
G10	1.1	0.9	1.3	0.6	28	0.0	0.0	0.0
G10	1.1	0.9	1.3	0.6	28	0.0	0.0	0.0
G10	1.5	5.7	2.3	2.2	28	0.1	0.1	0.0
G10	1.6	5.7	2.3	2.2	28	0.1	0.1	0.0
G10	2.7	3.1	4.0	2.4	28	0.0	0.0	-0.1
G10	2.8	3.1	4.1	2.4	28	0.0	-0.1	-0.1
G2	44.2	212.8	68.2	1.2	28	6.0	3.6	-2.4
G2	44.2	212.8	68.2	1.2	28	6.0	3.6	-2.4
G2	22.1	64.4	26.8	1.1	28	1.5	0.6	-0.9
G2	22.2	64.4	26.9	1.1	28	1.5	0.6	-0.9
G2	48.5	129.6	69.1	1.4	28	2.9	0.5	-2.4
G2	49.1	129.6	70.0	1.4	28	2.9	0.4	-2.4

G2	5.9	13.3	3.1	0.8	28	0.3	0.2	-0.1
G2	5.9	13.3	3.1	0.8	28	0.3	0.2	-0.1
G2	52.8	127.3	63.1	1.0	28	2.7	0.4	-2.2
G2	51.9	127.3	61.9	1.0	28	2.7	0.5	-2.2
G2	20.6	66.8	37.5	0.8	28	1.7	0.3	-1.3
G2	20.6	66.8	37.5	0.8	28	1.7	0.3	-1.3
G3	5.6	19.9	2.3	1.3	28	0.5	0.5	0.0
G3	5.5	19.9	2.3	1.3	28	0.5	0.5	0.0
G3	4.6	15.0	2.0	1.0	28	0.4	0.3	0.0
G3	4.6	15.0	1.9	1.0	28	0.4	0.3	0.0
G3	4.3	14.1	2.0	1.1	28	0.3	0.3	0.0
G3	4.3	14.1	2.0	1.1	28	0.3	0.3	0.0
G3	4.1	9.7	1.3	0.7	28	0.2	0.2	0.0
G3	4.1	9.7	1.3	0.7	28	0.2	0.2	0.0
G3	4.7	11.7	1.6	0.8	28	0.2	0.2	0.0
G3	4.7	11.7	1.6	0.8	28	0.2	0.2	0.0
G3	4.5	14.1	1.9	0.7	28	0.3	0.3	0.0
G3	4.5	14.1	1.8	0.7	28	0.3	0.3	0.0
G4	32.6	76.4	32.2	1.1	28	1.6	0.5	-1.1
G4	32.5	76.4	32.1	1.1	28	1.6	0.5	-1.1
G4	17.5	33.3	7.2	1.3	28	0.6	0.4	-0.2
G4	17.4	33.3	7.2	1.3	28	0.6	0.4	-0.2
G4	20.5	91.4	56.4	1.7	28	2.5	0.6	-2.0
G4	20.3	91.4	55.9	1.7	28	2.5	0.6	-1.9
G4	2.9	7.1	1.2	0.7	28	0.2	0.1	0.0
G4	2.9	7.1	1.2	0.7	28	0.2	0.1	0.0
G4	18.9	48.0	3.9	1.4	28	1.0	0.9	-0.1
G4	18.6	48.0	3.8	1.4	28	1.1	1.0	-0.1
G4	30.8	105.6	90.6	8.2	28	2.7	-0.3	-2.9
G4	26.1	105.6	76.8	8.2	28	2.8	0.4	-2.5
G5	3.7	10.2	3.9	1.8	28	0.2	0.2	-0.1
G5	3.6	10.2	3.9	1.8	28	0.2	0.2	-0.1
G5	1.8	9.3	2.9	1.2	28	0.3	0.2	-0.1
G5	1.8	9.3	3.0	1.2	28	0.3	0.2	-0.1
G5	2.6	12.7	3.2	1.5	28	0.4	0.3	-0.1
G5	2.6	12.7	3.3	1.5	28	0.4	0.3	-0.1
G5	1.6	5.8	1.7	1.0	28	0.1	0.1	0.0
G5	1.6	5.8	1.7	1.0	28	0.1	0.1	0.0
G5	2.2	4.7	2.6	1.4	28	0.1	0.0	0.0
G5	2.2	4.7	2.6	1.4	28	0.1	0.0	0.0
G5	3.1	14.8	2.6	1.1	28	0.4	0.4	-0.1

G5	3.1	14.8	2.6	1.1	28	0.4	0.4	-0.1
G6	3.5	13.4	2.3	1.3	28	0.4	0.3	0.0
G6	3.6	13.4	2.4	1.3	28	0.4	0.3	0.0
G6	3.1	14.3	2.3	1.4	28	0.4	0.4	0.0
G6	3.2	14.3	2.3	1.4	28	0.4	0.4	0.0
G6	2.7	13.3	1.9	1.1	28	0.4	0.4	0.0
G6	2.7	13.3	1.9	1.1	28	0.4	0.3	0.0
G6	2.1	9.5	1.6	0.8	28	0.3	0.2	0.0
G6	2.1	9.5	1.6	0.8	28	0.3	0.2	0.0
G6	7.3	21.1	2.1	1.4	28	0.5	0.5	0.0
G6	7.4	21.1	2.2	1.4	28	0.5	0.5	0.0
G6	5.1	21.7	3.3	1.6	28	0.6	0.5	-0.1
G6	5.0	21.7	3.2	1.6	28	0.6	0.5	-0.1
G7	3.2	12.6	4.3	1.8	28	0.3	0.2	-0.1
G7	3.2	12.6	4.4	1.8	28	0.3	0.2	-0.1
G7	3.1	12.0	4.0	2.1	28	0.3	0.2	-0.1
G7	3.1	12.0	4.0	2.1	28	0.3	0.2	-0.1
G7	2.8	11.0	3.4	1.8	28	0.3	0.2	-0.1
G7	2.8	11.0	3.4	1.8	28	0.3	0.2	-0.1
G7	1.6	6.2	1.7	1.0	28	0.2	0.1	0.0
G7	1.6	6.2	1.7	1.0	28	0.2	0.1	0.0
G7	1.9	7.7	3.0	1.7	28	0.2	0.2	0.0
G7	1.9	7.7	3.1	1.7	28	0.2	0.2	0.0
G7	3.2	2.8	3.5	2.3	28	0.0	-0.1	0.0
G7	3.3	2.8	3.7	2.3	28	0.0	-0.1	0.0
G8	3.2	18.8	3.9	2.2	28	0.6	0.5	-0.1
G8	3.1	18.8	3.7	2.2	28	0.6	0.5	-0.1
G8	1.7	10.5	2.7	1.8	28	0.3	0.3	0.0
G8	1.6	10.5	2.6	1.8	28	0.3	0.3	0.0
G8	1.8	13.7	2.4	1.6	28	0.4	0.4	0.0
G8	1.8	13.7	2.4	1.6	28	0.4	0.4	0.0
G8	1.4	8.8	1.1	0.7	28	0.3	0.3	0.0
G8	1.4	8.8	1.1	0.7	28	0.3	0.3	0.0
G8	1.8	12.7	2.3	1.5	28	0.4	0.4	0.0
G8	1.8	12.7	2.4	1.5	28	0.4	0.4	0.0
G8	3.0	16.1	3.5	1.7	28	0.5	0.4	-0.1
G8	2.9	16.1	3.4	1.7	28	0.5	0.4	-0.1
G9	3.2	9.3	3.4	2.4	28	0.2	0.2	0.0
G9	3.3	9.3	3.5	2.4	28	0.2	0.2	0.0
G9	3.4	18.6	3.5	1.9	28	0.5	0.5	-0.1
G9	3.3	18.6	3.4	1.9	28	0.5	0.5	-0.1

G9	2.0	11.4	3.0	2.2	28	0.3	0.3	0.0
G9	2.1	11.4	3.1	2.2	28	0.3	0.3	0.0
G9	1.3	4.2	1.6	1.0	28	0.1	0.1	0.0
G9	1.3	4.2	1.6	1.0	28	0.1	0.1	0.0
G9	4.9	12.9	4.0	2.5	28	0.3	0.2	-0.1
G9	4.9	12.9	4.0	2.5	28	0.3	0.2	-0.1
G9	3.4	11.5	2.9	2.1	28	0.3	0.3	0.0
G9	3.5	11.5	2.9	2.1	28	0.3	0.3	0.0
G1	2.0	9.3	1.4	0.7	28	0.3	0.2	0.0
G1	2.0	9.3	1.4	0.7	28	0.3	0.2	0.0
G1	2.5	7.6	2.6	0.4	28	0.2	0.1	-0.1
G1	2.5	7.6	2.6	0.4	28	0.2	0.1	-0.1
G1	1.8	6.2	1.3	0.6	28	0.2	0.1	0.0
G1	1.9	6.2	1.3	0.6	28	0.2	0.1	0.0
G1	2.2	8.7	1.2	0.6	28	0.2	0.2	0.0
G1	2.3	8.7	1.3	0.6	28	0.2	0.2	0.0
G1	1.5	7.3	1.0	0.5	28	0.2	0.2	0.0
G1	1.6	7.3	1.1	0.5	28	0.2	0.2	0.0
G10	1.8	7.2	3.6	1.6	28	0.2	0.1	-0.1
G10	1.8	7.2	3.6	1.6	28	0.2	0.1	-0.1
G10	2.1	11.5	3.3	1.4	28	0.3	0.3	-0.1
G10	2.1	11.5	3.3	1.4	28	0.3	0.3	-0.1
G10	0.7	3.0	2.4	1.3	28	0.1	0.0	0.0
G10	0.7	3.0	2.4	1.3	28	0.1	0.0	0.0
G10	1.0	2.4	2.0	0.9	28	0.1	0.0	0.0
G10	0.9	2.4	2.0	0.9	28	0.1	0.0	0.0
G10	1.8	10.2	2.4	0.9	28	0.3	0.2	-0.1
G10	1.7	10.2	2.4	0.9	28	0.3	0.3	-0.1
G2	1.3	6.3	1.2	0.7	28	0.2	0.2	0.0
G2	1.3	6.3	1.2	0.7	28	0.2	0.2	0.0
G2	1.8	8.5	1.6	0.7	28	0.2	0.2	0.0
G2	1.7	8.5	1.6	0.7	28	0.2	0.2	0.0
G2	2.0	8.2	1.9	0.9	28	0.2	0.2	0.0
G2	2.0	8.2	1.9	0.9	28	0.2	0.2	0.0
G2	1.7	6.5	1.2	0.7	28	0.2	0.2	0.0
G2	2.1	6.5	1.5	0.7	28	0.2	0.1	0.0
G2	1.1	7.4	1.2	0.6	28	0.2	0.2	0.0
G2	1.2	7.4	1.2	0.6	28	0.2	0.2	0.0
G3	1.3	7.6	1.4	0.8	28	0.2	0.2	0.0
G3	1.3	7.6	1.4	0.8	28	0.2	0.2	0.0
G3	1.3	7.6	1.4	0.8	28	0.2	0.2	0.0

G3	1.3	7.6	1.4	0.8	28	0.2	0.2	0.0
G3	1.7	7.7	1.8	0.8	28	0.2	0.2	0.0
G3	1.6	7.7	1.7	0.8	28	0.2	0.2	0.0
G3	1.7	6.3	1.1	0.6	28	0.2	0.1	0.0
G3	1.7	6.3	1.1	0.6	28	0.2	0.1	0.0
G3	1.3	7.5	1.3	0.5	28	0.2	0.2	0.0
G3	1.3	7.5	1.3	0.5	28	0.2	0.2	0.0
G4	1.2	9.6	2.3	0.8	28	0.3	0.2	-0.1
G4	1.2	9.6	2.4	0.8	28	0.3	0.2	-0.1
G4	1.1	7.6	1.4	0.7	28	0.2	0.2	0.0
G4	1.1	7.6	1.4	0.7	28	0.2	0.2	0.0
G4	0.9	5.9	1.2	0.5	28	0.2	0.2	0.0
G4	0.9	5.9	1.2	0.5	28	0.2	0.2	0.0
G4	1.5	9.4	1.8	1.0	28	0.3	0.3	0.0
G4	1.4	9.4	1.7	1.0	28	0.3	0.3	0.0
G4	1.1	4.0	1.0	0.5	28	0.1	0.1	0.0
G4	1.1	4.0	1.0	0.5	28	0.1	0.1	0.0
G5	1.2	7.0	2.3	1.0	28	0.2	0.2	0.0
G5	1.1	7.0	2.2	1.0	28	0.2	0.2	0.0
G5	1.5	5.7	2.3	0.9	28	0.2	0.1	0.0
G5	1.5	5.7	2.3	0.9	28	0.2	0.1	0.0
G5	1.3	6.1	2.1	1.2	28	0.2	0.1	0.0
G5	1.3	6.1	2.1	1.2	28	0.2	0.1	0.0
G5	1.7	6.4	2.1	0.8	28	0.2	0.1	0.0
G5	1.7	6.4	2.1	0.8	28	0.2	0.1	0.0
G5	1.9	9.4	2.3	0.6	28	0.3	0.2	-0.1
G5	1.7	9.4	2.1	0.6	28	0.3	0.2	-0.1
G6	1.7	5.3	2.1	1.1	28	0.1	0.1	0.0
G6	1.7	5.3	2.0	1.1	28	0.1	0.1	0.0
G6	1.4	6.1	1.9	0.8	28	0.2	0.1	0.0
G6	1.4	6.1	1.9	0.8	28	0.2	0.1	0.0
G6	1.8	10.5	2.8	1.0	28	0.3	0.2	-0.1
G6	1.8	10.5	2.8	1.0	28	0.3	0.2	-0.1
G6	2.1	13.2	1.8	0.7	28	0.4	0.4	0.0
G6	2.2	13.2	1.8	0.7	28	0.4	0.4	0.0
G6	1.6	7.8	2.1	0.7	28	0.2	0.2	0.0
G6	1.6	7.8	2.1	0.7	28	0.2	0.2	0.0
G7	1.8	9.8	3.0	1.1	28	0.3	0.2	-0.1
G7	1.8	9.8	3.0	1.1	28	0.3	0.2	-0.1
G7	1.9	8.9	3.4	1.3	28	0.3	0.2	-0.1
G7	1.9	8.9	3.3	1.3	28	0.3	0.2	-0.1

G7	1.8	12.1	3.3	1.2	28	0.4	0.3	-0.1
G7	1.8	12.1	3.3	1.2	28	0.4	0.3	-0.1
G7	1.2	5.9	2.5	1.0	28	0.2	0.1	-0.1
G7	1.2	5.9	2.5	1.0	28	0.2	0.1	-0.1
G7	1.5	3.3	2.2	0.9	28	0.1	0.0	0.0
G7	1.5	3.3	2.1	0.9	28	0.1	0.0	0.0
G8	2.7	12.9	2.6	1.3	28	0.4	0.3	0.0
G8	2.7	12.9	2.6	1.3	28	0.4	0.3	0.0
G8	1.9	9.1	2.4	1.1	28	0.3	0.2	0.0
G8	1.9	9.1	2.5	1.1	28	0.3	0.2	-0.1
G8	2.0	11.7	2.2	1.0	28	0.3	0.3	0.0
G8	2.1	11.7	2.2	1.0	28	0.3	0.3	0.0
G8	2.4	11.4	2.4	0.7	28	0.3	0.3	-0.1
G8	2.4	11.4	2.4	0.7	28	0.3	0.3	-0.1
G8	2.6	11.5	2.6	1.1	28	0.3	0.3	-0.1
G8	2.6	11.5	2.7	1.1	28	0.3	0.3	-0.1
G9	2.9	8.3	2.5	1.4	28	0.2	0.2	0.0
G9	2.4	8.3	2.0	1.4	28	0.2	0.2	0.0
G9	2.3	11.9	2.8	1.2	28	0.3	0.3	-0.1
G9	2.3	11.9	2.8	1.2	28	0.3	0.3	-0.1
G9	2.0	10.4	1.9	1.3	28	0.3	0.3	0.0
G9	2.0	10.4	1.9	1.3	28	0.3	0.3	0.0
G9	3.1	14.4	2.9	1.7	28	0.4	0.4	0.0
G9	3.1	14.4	2.9	1.7	28	0.4	0.4	0.0
G9	2.6	13.1	2.3	2.1	28	0.4	0.4	0.0
G9	2.6	13.1	2.2	2.1	28	0.4	0.4	0.0