Field Scale Variability of Nitrogen and δ15N in Soil and Plants

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Field Scale Variability of Nitrogen and δ¹⁵N in Soil and Plants

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ABSTRACT

Understanding the factors that influence soil and plant nitrogen (N) spatial variability may improve our ability to develop management systems that maximize productivity and minimize environmental hazards. The objective of this study was to determine the field (65 ha) scale spatial variability of N and δ¹⁵N in soil and corn (Zea mays). Soil, grain, and stover samples were collected from grids that ranged in size from 30 by 30 m to 60 by 60 m. Plant samples, collected following physiological maturity in 1995, were analyzed for total N and δ¹⁵N. Soil samples, collected prior to planting in the spring of 1995 and 1996, were analyzed for inorganic-N, total N, and δ¹⁵N. All parameters showed strong spatial relationships. In an undrained portion of the field containing somewhat poorly and poorly drained soils there was a net loss of 95 kg N ha⁻¹, while in an adjacent area that was tile drained there was a net gain of 98 kg N ha⁻¹. Denitrification and N mineralization most likely were responsible for losses and gains, respectively. Differences between the N balances of these areas (193 kg N ha⁻¹) provide a relative measure of the impact of tile drainage on plant N availability and greenhouse gas production in a wet year.
Site-specific management provides a link between new technologies and sound agronomic practices. Applying optimum amounts of nutrients at specific points within a field is a sound concept and has the potential to increase profitability and simultaneously reduce the impact of agricultural activities on the environment. Our current ability to attain this goal is limited by our inability to accurately predict plant responses at specific field locations (Kitchen et al., 1995; Lamb et al., 1995).

Many current fertilizer recommendations conceptualize N recommendations as balancing a checkbook, with fertilizer recommendations being equal to shortages after N credits are subtracted from needs (N required for a given yield goal). Nitrogen credits may include residual soil nitrate-N, irrigation water nitrate, apparent legume N enhancements, and estimates of N mineralization from organic amendments. However, the ability of the soil to mineralize N is influenced by complex interactions between chemical, biological, and physical processes, which are influenced by landscape position (Rice et al., 1986). To fully take advantage of site specific N management, an improved understanding of the fate of soil and fertilizer N at different landscape positions is required.

Differences in the fertilizer and soil $^{15}$N/$^{14}$N ratios make it theoretically possible to evaluate the fate of different N sources in plant and soil systems. However, because $^{15}$N/$^{14}$N ratios of plant and soil samples contain spatial variability (Karamonos et al., 1981), traditional statistical approaches that assume random variation may not be appropriate. Sutherland et al. (1993) measured $\delta^{15}$N spatial variability in soil and plant samples in a 1.12 ha field. In their study, samples were collected from a 10 by 10 m grid. They reported that: (i) topography had a significant influence on the landscape-scale patterns of $\delta^{15}$N in soil and durum wheat (*Triticum durum* Desf.) samples; (ii) soil $\delta^{15}$N variability was less than plant $\delta^{15}$N variability; and (iii) the extreme outliers in plant $\delta^{15}$N were associated with portions of the field with the lowest Eh values.

Selles et al. (1986) evaluated natural $^{15}$N abundance spatial variability in cultivated and native prairie soils. They reported that the number of samples required to estimate the true mean of total N at the 1 and 5% probability levels was nearly double that for estimating the $\delta^{15}$N value, and that the spatial variability of $\delta^{15}$N values of total N most likely reflected isotopic composition of N substances entering the soil system or changes in the isotopic composition of soil N due to the humification processes, probably induced by variations in topography.

Clay (1997) compared fertilizer use by corn (*Zea mays*) as estimated by the difference (fertilizer N used = N contained in fertilized plants - N contained in unfertilized plants) and the natural abundance $\delta^{15}$N approaches for a well drained soil in 1992 and 1993. Nitrogen fertilizer rates in the replicated field experiment were 0 and 15.7 g N m$^{-2}$. Clay (1997) reported that fertilizer efficiency as estimated by the difference and $\delta^{15}$N natural abundance approaches were similar when the crop responded to N fertilizer; however, when an N response was not observed,
the difference method underestimated the amount of fertilizer N contained in the plant and overestimated soil N contained in the plant. This research showed that the difference and $\delta^{15}$N natural abundance approaches complement rather than duplicate each other. Due to the complexity of watersheds, isotopic as well as nonisotopic information will be required to evaluate the impact of landscape position on crop responses. The objective of this study was to determine the field (65 ha) scale spatial variability of N and $\delta^{15}$N in soil and corn (*Zea mays*).

**MATERIALS AND METHODS**

A 65 ha field, located in eastern South Dakota having latitude and longitude values of 44.17°N and 96.62°W, respectively, was used for this study. Corn (*Zea mays* L.) was planted in the spring of 1995 at a row spacing of 56 cm. Urea (46-0-0) fertilizer was broadcast applied on November 8, 1994, at the rate of 108 kg N ha$^{-1}$. During planting, 12 kg N were applied as a popup fertilizer and an additional 4 kg N were applied with a herbicide application. Total amount of N fertilizer applied was 124 kg-ha$^{-1}$. This field had a corn/soybean (*Glycine max*) rotation and a no-tillage management system. The dominant soil series on the ridge top and shoulders were a Doland loam (fine-loamy, mixed Pachic Udic Haploboroll) and a Vienna loam (fine-loamy, fixed Pachic Haploboroll). These soils had slopes ranging between 2 and 6%. The dominant soil series on the sideslopes or backslopes was a Kranzburg silty clay loam (fine-silty, mixed, Udic Haploboroll). The Kranzburg had slopes ranging between 2 and 6%. Waubay silty clay loam (fine-silty, mixed Pachic Udic Haploboroll) was the dominant soil series in the footslope or swale areas. The pH values (water) of soils on ridge tops ranged from 6 to 7, while in valley areas pH values ranged from 7 to 8. The organic C in soils on ridge tops ranged between 26 and 32, while in valley areas, C ranged from between 30 and 40 g·kg$^{-1}$.

Soil samples from a 30 by 30 m grid were collected from the 0- to 15- and 15- to 60-cm depths in April 1995. Each sample contained 15 individual cores from the 0- to 15-cm and 15- to 60-cm soil depths. The 15 individual cores were collected at sample points located every 11.4 cm along a transect that transversed 3 corn rows. Samples from the 0- to 15-cm depth were dried, ground (ball milled), and analyzed for NH$_4^+$-N and NO$_3^-$-N on a Wescan ammonia analyzer (Carlson, 1978), and for total N and $\delta^{15}$N of total N on a Europa Ratio Mass Spectrometer (Europa Scientific Ltd, UK) (Barrie et al., 1995). The standard error of the mean for $\delta^{15}$N analysis were less than 0.2‰ (Clay, 1997). Samples from the 15- to 60-cm soil depth were analyzed for NH$_4^+$-N and NO$_3^-$-N. The elevation, latitude and longitude coordinates for each grid point was determined with a rod transit and a GPS system, respectively. In April 1996 the field was resampled on a 60 by 60 m grid following the same procedure as described above. Samples from the 0- to 15- and 15- to 60-cm soil depths were dried, ground, and analyzed for NO$_3$-N and NH$_4^+$-N.
Stover and corn grain samples were collected at plant maturity (October 1995) from a 30 by 60 m grid. At each grid point, grain was collected from 10 plants, stover was collected from 5 plants, and plant populations were measured over a 8.13 m² area. Samples were dried, weighed, ground, and analyzed in duplicate for total N and δ¹⁵N on a Europa 20-20 ratio mass spectrometer (Europa Scientific, Ltd, UK). The δ¹⁵N value was calculated by the following equation:
FIELD SCALE VARIABILITY OF NITROGEN AND $\delta^{15}N$

\[
(\delta^{15}N = (((N/14N_{sample}) / (15N/14N_{atmosphere}) - 1)*1000))\%_0
\]

Plant $\delta^{15}N$ values were normalized by the following equation:

\[
N\delta^{15}N = ((\delta^{15}N_{soil} - \delta^{15}N_{plant}) / (\delta^{15}N_{soil}))
\]

where, soil $\delta^{15}N_{soil}$ was the $\delta^{15}N$ of total N contained in the 0- to 15-cm soil depth, and $\delta^{15}N_{plant}$ was the $\delta^{15}N$ value of N contained in the grain and stover.

Three areas of the field were selected for additional characterization (Figure 1). Areas 1 and 2 contained similar soils and were poorly or somewhat poorly drained. The difference between the two areas were that area 1 was not tile drained while area 2 was tile drained. Area 3 was located on the ridge between areas 1 and 2 and contained moderate to well drained soils. The net N balances for areas 1, 2, and 3 were determined by the following equation:

\[
\text{Net N balance} = SN_{96} + PBN - SN_{95} - FN_{95}
\]

where $SN_{95}$ is the soil NO$_3$-N plus NH$_4$-N in the spring of 1996, PBN is the N contained in the grain and stover at harvest in 1995, $SN_{95}$ is the soil NO$_3$-N plus NH$_4$-N in the spring of 1995, and $FN_{95}$ is the N fertilizer applied after soil sampling (16 kg N ha$^{-1}$). These calculations assume that all of the fall applied urea was hydrolyzed during the six month period between application and sampling.

Semi-variances, semi-variograms, skewness, and kurtosis were calculated using Geo-eas (Englund and Sparks, 1991). Surfer: Version 6 (Golden Software, Inc., Golden, CO) was used to produce contour maps. The ratio of semi-variance values at 30 to 540 m was used as a indicator of spatial dependance. If the ration was less than 0.25, between 0.25 and 0.75, or greater than 0.75 then the spatial relationships were characterized as strong, moderate, and weak, respectively (Cambardella et al., 1994).

RESULTS AND DISCUSSION

Rainfall amounts between April 1 and September 30 in 1994 and 1995 were 58.3, 63.6 cm, respectively. The growing degree days (GGD=((max °C+min°C)/2)-10) in 1994 and 1995 were 1,297 and 1,288, respectively. Rainfall amounts in 1994 and 1995 were higher than the average value of 43.2 cm, while GGD were less than the average value of 1,413. The high rainfalls when combined with cool temperatures resulted in soils that were relatively wet during most of 1995 and corn yields that were lower than expected.

Spatial Analysis: Soil
The SE ¼ of the field had higher nitrate concentrations than the W ½ and SW ¼ areas (Figure 1). Based on NO$_3$-N concentrations, current N fertilizer recommendations, and a yield goal of 9 Mg grain ha$^{-1}$, the SE ¼ did not require
additional N fertilizer, while the W ½ and SW ¼ required an additional 100 to 150 kg N ha⁻¹. Total N in the surface 15 cm of soil over the entire field ranged from 1 to 3 g N kg⁻¹ (Figure 1). Drainage ways generally had the highest total N concentrations. The δ¹⁵N of total N ranged from 4 to 9%. This range of values were similar to those reported by Sutherland et al. (1993). The mean and variance of δ¹⁵N were 5.98 and 0.794, respectively. The skewness (0.664) and kurtosis (4.172) values indicated that the distribution was nonnormal and was skewed toward small values. Sutherland et al. (1993) skewness and kurtosis values indicated that δ¹⁵N of total N was normally distributed.

Due to the spatial distribution of NO₃-N in the soil, semi-variograms were calculated for both the SE ¼ and for the W ½ and SW ¼ areas (Figure 2). The

FIGURE 2. Semi-variograms of soil samples collected in the spring of 1995 from a 30 by 30 m grid and analyzed for the mg NO₃-N kg⁻¹ of soil (0- to 60-cm), g total-N kg⁻¹ of soil (0- to 15-cm), and soil δ¹⁵N (0- to 15-cm).
semi-variance value for the SE ¼ at 30 m was approximately one third the value at 540 m, which indicated that in this portion of the field NO₃-N had a moderate spatial relationship. In W ¼ and SW ¼ portions of the field the semi-variance value at 30 and 540 m were similar which indicated that in this portion of the field a weak spatial relationship was observed. It is interesting to note, that all of the individual semivariance values were lower for the W ¼ and SW ¼ area than for the SE ¼ area. For total N a moderate spatial relationship was measured.

Soil δ¹⁵N had a moderate spatial relationship because the semi-variance value at 30 m was approximately 30% of the value at 500 m. Sutherland et al. (1993) had similar results and reported soil δ¹⁵N had a strong spatial relationship. However, it is important to note that our semi-variance value at 540 m was similar to the Sutherlands et al. (1993) value at 15 m.

Spatial Analysis: Plants

Plant population, and grain and stover yields had moderate to strong spatial relationships (Figure 3). Plant δ¹⁵N, grain δ¹⁵N, and stover δ¹⁵N all had strong spatial relationships. These findings were similar to those reported by Sutherland et al. (1993).

Plant populations ranged from less than 20,000 plants·ha⁻¹ to over 72,000 plants·ha⁻¹. Grain production in the field ranged from less than 3 to 10 Mg ha⁻¹ (Figure 4). Highest yields were recorded in tile drained or contained well drained soils. Lowest yields were measured in poorly drained areas that were not tile drained. Over the entire field, the average grain yield per plant was 114 g with a variance (s²) of 636. The minimum yield per plant was 20 and the maximum yield was 160 g. Stover yields per plant ranged from 20 to 108 g, had a mean of 73 g, and a variance of 212. The skewness and kurtosis values for grain yields per plant were -1.19 and 4.98, respectively, and for stover yields were -0.663 and 4.093 for, respectively. These values show that grain and stover distributions were skewed toward large values.

The amount of N contained in stover plus grain at harvest was dependent on landscape position and ranged from 15 to 200 kg N ha⁻¹ (Figure 4) or from 0.52 to 2.98 g·plant⁻¹. Mean N content was 107 kg N ha⁻¹ (s²=1135) or 1.96 g·plant⁻¹ (s²=0.215). The skewness and kurtosis values for N removal per ha were -0.485 and 3.43, respectively. The skewness and kurtosis values for N removal per plant were -0.547 and 3.53, respectively. These values indicate that both distributions were normal in appearance.

Plant δ¹⁵N values over the entire field ranged from 2 to 13% with a mean of 5.19, and a variance of 4.93 (Figure 4). The skewness (1.77) and kurtosis (5.92) values indicate that plant δ¹⁵N was skewed toward small values. Sutherland et al. (1993) reported similar skewing.

Over the entire field, biomass (grain plus stover) δ¹⁵N values were positively correlated to stover δ¹⁵N values and negatively correlated to g biomass and grain
FIGURE 3. Plant population (plants/ha), grain yield (g/plant), stover yield (g/plant), ppm plant $\delta^{15}$N, ppm grain $\delta^{15}$N, and ppm stover $\delta^{15}$N semi-variograms in plant samples collected from a 30 by 60 m grid in the fall of 1995.

The negative correlation between biomass $\delta^{15}$N values and grain yields was the direct result of poor grain production in poorly drained soils that were not tile drained.

The NP$\delta^{15}$N values over the entire field were positively correlated to grain yields (Figure 6). The positive relationship could have been caused by a combination of factors that included $^{15}$N enrichment of inorganic N pools caused by denitrification or volatilization (Chien et al., 1977) and/or slow plant growth in poorly drained areas. The highest NP$\delta^{15}$N values were measured in plants harvested from ridge areas, while the lowest values were measured in plants harvested from poorly drained soils that were not tile drained (Figure 7).
FIGURE 4. Grain yields (A), plant N removal (B), and plant $\delta^{15}$N (C) values superimposed on the surface elevation map. Areas 1, 2, and 3 represent portions of the field summarized in Table 1.

Net Nitrogen Balance

The amount of N lost or gained from individual locations within the field was influenced by landscape position (Table 1). Areas 1 and 2 contained somewhat poorly and poorly drained soils and were located on either side of a ridge (area 3). Facts that need to be considered when comparing the three areas includes that: (i) area 1 was undrained while area 2 was tile drained; (ii) 107 kg N ha$^{-1}$ of urea
FIGURE 5. The relationship between plant $\delta^{15}$N and stover $\delta^{15}$N, biomass production, grain yield, and grain N concentration.

FIGURE 6. The relationship between plant NP$\delta^{15}$N $((\delta^{15}_{\text{soil}} - \delta^{15}_{\text{plant}})/\delta^{15}_{\text{soil}})$ and grain yields.
ammonium nitrate fertilizer was applied 6 months prior to soil sampling; (iii) if 
the $\delta^{15}N$ value of mineralized N was similar to the $\delta^{15}N$ value of total soil N 
(Karamonos and Rennie, 1980), then mineralization can be responsible for 
lowering NP$\delta^{15}N$ values to zero, but not below zero; and (iv) if $^{15}N$ enrichment of 
the soil nitrate pool occurs during denitrification (Chien et al., 1977), then the $\delta^{15}N$ of plant material can become larger than the associated soil values, resulting 
in negative NP$\delta^{15}N$ values.

Areas 1 and 2 had higher total organic soil N than area 3, which was located on 
a ridge (data not shown). Erosion was thought to be responsible for these results. 
Nitrate concentrations in 1995 did not match the total organic N results and was 
highest in area 1 and lowest in area 3 (Table 1), while NH$_4^+$-N concentrations 
were similar in the three zones. Total inorganic N in the spring of 1995 ranged 
from 30 ppm (240 kg N ha$^{-1}$) in area 1 to 15 ppm (120 kg N ha$^{-1}$) in area 2. In 
areas one and two, 20% and 56% of the soil inorganic N was in the NH$_4^+$-N form, 
respectively.

Yields, plant populations, the amount of N removed, and NP$\delta^{15}N$ values were 
less in area 1 than 2 (Table 1). The net N balance for area 1 showed that 
approximately 95 kg N ha$^{-1}$ were lost between the spring of 1995 and 1996. 
However, because this estimate does not consider N mineralization, N loss must 
have been much greater than 95 kg ha$^{-1}$. The NP$\delta^{15}N$ values when combined with
TABLE 1. The influence of sampling site on grain yield, plant population, N removed by the corn plant, NP$^{15}$N, nitrate plus ammonium concentrations in the surface 60 cm of soil in the spring of 1995 and 1996, and the net balance of N removed by the crop and remaining in the soil. The 95% confidence interval for each mean is shown in parenthesis.

<table>
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<tr>
<th>Area</th>
<th>Plant g/plt$^1$</th>
<th>Hectare kg:ha$^1$</th>
<th>Population Pl/ha$^4$</th>
<th>Grain yield g-plr</th>
<th>Plant population Pl/ha$^4$</th>
<th>N in corn kg:ha$^4$</th>
<th>NP$^{15}$N %</th>
<th>Soil $^{15}$N g:$^{15}$N</th>
<th>NO$_3$-N mg N g-soil$^4$</th>
<th>NH$_4$-N mg N g-soil$^4$</th>
<th>NH$_3$+NO$_3$ mg N g-soil$^4$</th>
<th>Net N balance kg:ha$^1$</th>
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<td>4700(1140)</td>
<td>50000(6890)</td>
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<td>7.7(0.7)</td>
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<tr>
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<td>6800(460)</td>
<td>55200(2600)</td>
<td>117(8.5)</td>
<td>0.06(0.08)</td>
<td>5.91(0.31)</td>
<td>7.1(0.6)</td>
<td>7.8(0.6)</td>
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<tr>
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<td>6970(411)</td>
<td>58500(3160)</td>
<td>124(6.0)</td>
<td>0.41(0.04)</td>
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</table>

$^1$ g/plt = grams per plant
$^2$ kg:ha = kilograms per hectare
$^3$ Pl/ha = plants per hectare
$^4$ mg N g-soil = milligrams of nitrogen per gram of soil
the N balance information provides clues toward identifying mechanisms responsible for N losses. If fertilizer was preferentially leached from the soil or lost in runoff water, then the plant $\delta^{15}N$ could increase to the soil $\delta^{15}N$ value resulting in a near zero positive NP$\delta^{15}N$ value (Karamonos and Rennie, 1980). Leaching and runoff were discounted as loss mechanisms because negative NP$\delta^{15}N$ values were measured in areas 1 and 2. If ammonia volatilization or denitrification were responsible for the N loss in area 1, then $^{15}N$ enrichment could occur, resulting in plant $\delta^{15}N$ values which were greater than the soil $\delta^{15}N$ values. Under these conditions, negative NP$\delta^{15}N$ values are possible. Even though ammonia volatilization could occurred, it was discounted because N loss occurred 6 months after the fertilizer was applied. If denitrification was responsible for the N loss, then the lowest values should have been measured in area 1 and the highest values in area 3. This was the case and therefore, based on a process of elimination and that the results matched expectations, N losses in area 1 most likely were caused by denitrification.

The net N balance for area 2 showed that 98 kg of N were gained between the springs of 1995 and 1996. This N gain most likely resulted from N mineralization of soil organic matter and soybean biomass (grown at the site in 1994). The near zero and negative NP$\delta^{15}N$ values for this area also suggests that denitrification reduced inorganic N concentrations, and therefore N mineralization most likely exceeded 98 kg N ha$^{-1}$. The difference in the net N balances of areas 1 and 2 (193 kg N ha$^{-1}$) demonstrate the relative importance of tile drainage on N plant availability and greenhouse gas production.

Area 3 which was located on the ridge between areas 1 and 2 had the highest NP$\delta^{15}N$ and lowest $\delta^{15}N_{soil}$ values (Table 1). The amount of N removed per plant was similar in areas 2 and 3. Area 3 had a net N balance of 57 kg N ha$^{-1}$. If the net N balance represents N derived from N mineralization, then more N was mineralized in area 2 than 3. Positive NP$\delta^{15}N$ values in this zone indicate that denitrification did not substantially reduce inorganic N concentrations. If we assume that: (i) denitrification did not occur; (ii) the $\delta^{15}N$ of the fertilizer was zero; (iii) the $\delta^{15}N$ of the soil is similar to the $\delta^{15}N$ value of plants grown in unfertilized soil; and (iv) N$^{15}$ discrimination did not occur during NO$_3^{-}$N leaching (Karamonos and Rennie, 1980), then the NP$\delta^{15}N$ value indicates that approximately 41% of the N contained in the crop at harvest was derived from fertilizer.

CONCLUSIONS

This research shows that topography had a large impact on $\delta^{15}N$ in soil and plant samples, grain and stover yields, nitrate concentrations, and total soil N. The semi-variograms for all of the parameters measured, showed strong spatial relationships. The area of the field (area 3) where denitrification was unlikely to occur had NP$\delta^{15}N$ values ranging from 0.3 to 0.6. Areas 1 and 2 contained poorly or somewhat poorly drained soils and had NP$\delta^{15}N$ values that were near zero or
negative. Negative values can result from preferential denitrification of $^{14}$N relative to $^{15}$N. The NPS$^{15}$N values, N removed by the plant, and inorganic N concentrations were used to evaluate the relative importance of N leaching, mineralization, and denitrification in this complex system. The spatial variability of $\delta^{15}$N in soil and plant samples did not hinder the ability of using natural variation in the $^{15}$N for studying important agronomic processes.

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