Soil Organic Nitrogen Enrichment Following Soybean in an Iowa Corn–Soybean Rotation

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ABSTRACT

Understanding soil organic N (ON) pool enrichment may help explain why rates of N fertilization required to attain maximum corn (*Zea mays* L.) yields are usually lower for corn following soybean [*Glycine max* (L.) Merr.] than for corn following corn. Our objectives were to quantify the ON pools within a 16-ha Iowa field and to correlate those results with corn yield. Spring and fall measurements of ON content (0–15 cm soil) as amino acids (AAs), amino sugars (ASs), and NH_4^+ were made using samples collected between 1997 and 1999 from 10 soil map units. The chemical extraction method determined an average 87% of the total N content ($n = 10$ soils) as identified ON but gave reduced ON recovery from depression soils that experienced periods of water ponding. The total AA concentrations measured in May were positively correlated ($r^2 = 0.84$, $P < 0.01$) with corn yield during a dry year (1997) and 7 out of 10 soils provided near maximum yields. A wetter 1999 boosted overall corn yields 6.6% but resulted in a poorer relationship between May AA concentrations and corn yield. Microbial N compounds measured (May 1997) as glucosamine, galactosamine, and ornithine were also positively correlated with corn yield ($r^2 = 0.84$, $\rho < 0.01$; $r^2 = 0.94$, $P < 0.001$; $r^2 = 0.93$, $P < 0.001$, respectively). The ON concentration decreased during corn production from May to September 1997 an average of 367 kg N ha^{-1} but increased following soybean growth in 1998 by 320 kg N ha^{-1}. The chemical extraction methodology identified soils that may not require the full amount of N fertilizer currently being applied, thus decreasing the potential for N loss to surface and ground water resources without decreasing opportunities to achieve optimum yield.

P UBLIC CONCERN that excessive N fertilization may contribute to NO_3^- enrichment of ground and surface water has stimulated interest in developing analytical methods that can improve our understanding of N cycling and the accuracy of N fertilizer recommendations for corn (Mulvaney et al., 2001). Estimating plant-available soil N not derived from current fertilizer applications is very difficult because of the complicated nature of the soil N cycle. For example, mineralization of indigenous soil N has been shown to increase with application of N fertilizers (Broadbent, 1965; Kissel et al., 1977; Hills et al., 1978; Martens and Dick, 2003). Power et al. (1986) using labeled fertilizer and plant residues found that over 80% of the N assimilated by corn during a growing season originated from indigenous soil N. Unfortunately, identification and measurement of a soil N fraction that could provide sufficient N to support plant growth has been difficult because this fraction clearly responds to seasonal temperature and moisture variations.

Predicting the formation of plant-available N is complicated by the opposing processes of mineralization and immobilization and their interactions with seasonal temperature and precipitation (Jansson and Persson, 1982). Numerous attempts have been made to identify a labile pool of soil ON through chemical fractionation of the soil N hydrolysates (Keeney and Bremner, 1964; Porter et al., 1964; Kahn, 1971; Meints and Peterson, 1977), but with little tangible success. Recently, Khan et al. (2001) reported that determination of soil ASs released by chemical hydrolysis provided a means to separate soils that respond and do not respond to N fertilizer during corn production. Currently, preplant and presidedress N tests are recommended by Minnesota and Iowa, respectively, to provide an estimate of plant-available N and to guide fertilizer recommendations needed to optimize corn grain yield, but inherent limitations with those tests have resulted in frequent failures to predict actual N needs (Mulvaney et al., 2001).

Soybean in Iowa can be given a credit of up to 56 kg N ha^{-1} for a subsequent corn crop (Blackmer et al., 1997). This credit was based on N rate tests that showed optimum N fertilizer rates were less for corn following soybean than for corn following corn and postulated to be due to N fixation by soybean or a change in mineralization patterns (Green and Blackmer, 1995). However, the N credit theory has been questioned because estimates of N fixation and subsequent removal with the grain suggest that soybean actually results in a net removal of N from many soils (Heichel and Barnes, 1984). Recent research by Mayer et al. (2003) has found that legume growth resulted in a large influx of N to the soil in the form of root exudates. Use of ^{15}N isotopes showed that at legume maturity, 15 to 21% of belowground ^{15}N was identified in microbial biomass or as identifiable root fragments, while the remaining 79 to 85% of belowground ^{15}N was not identified. The N forms in the soybean exudates were not evaluated but would be expected to be composed mainly of AAs, the main form of N found in mature soybean tissue (Martens and Loeffelmann, 2003). Accurate estimates of the amount of soybean N returned to the soil for utilization by the following corn crop would greatly improve the prediction of N fertilizer needed for optimum yields.

Recently, improved methodology for analysis of soil ON composition has shown that up to 92% of the total N present in well-drained soils can be identified as AAs, ASs, and NH_4^+ originating from fixed soil NH_4^+ and AA extraction (Martens and Loeffelmann, 2003). Our

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Abbreviations: AA, amino acid; AS, amino sugar; GalX, galactosamine; GluX, glucosamine; MSA, methanesulfonic acid; ON, organic N.

objectives were to apply the new methodology to soil samples from a 16-ha, long-term corn–soybean field study to follow the soil ON pools and the relationship between spring and fall hydrolyzed N and crop yield.

MATERIALS AND METHODS

Site Description

The soils, which formed in calcareous glacial till (Andrews and Didericksen, 1981), were obtained from a 16-ha field in central Iowa that is classified as being in the Clarion (fine-loamy, mixed, superactive, mesic Typic Hapludolls)–Nicollet (fine-loamy, mixed, superactive, mesic Aquic Hapludolls)–Webster (fine-loamy, mixed, superactive, mesic Typic Endoaquolls) soil association. The field is characterized by low relief swell and swale topography representative of the Des Moines lobe of the Cary substage of glaciation (Ruhe, 1969). The field is tile drained, but there are no surface inlets and runoff frequently accumulates and remains in closed depressions for several days after high intensity and/or high volume precipitation events. Detailed information on the soils can be found in Steinwand and Fenton (1995) and Steinwand et al. (1996).

Soils

Soil samples were obtained from the center of representative sites for the soil type noted (5 cm diameter by 15 cm deep) by a Giddings hydraulic sampler (Giddings Machine Co., Ft. Collins, CO) from 10 of 228 yield plots (each plot 12.1 by 3.0 m) in the spring and fall of 1997 and 1998 and in the spring of 1999 (Table 1). The 10 sampling sites had been chosen for more intensive monitoring because they represented the full range in crop yield and the soil map units that had been identified through several years of research within the field (Colvin et al., 1997; Steinwand et al., 1996). All soil was air dried following sampling and passed through a 2-mm sieve before analyses. Soil pH was measured in 0.01 M CaCl$_2$ and soil texture by a pipette method described by Gee and Bauder (1986). Total organic C and total N contents were determined by dry combustion with a PerkinElmer 2400 C–H–N analyzer (PerkinElmer, Inc., Fullerton, CA).

Crop Management

The privately managed central Iowa field with soils typical of the area has been monitored by the USDA-ARS National Soil Tilth Laboratory, Ames, IA, since 1989 with all records collected and management practices performed by local farmers since 1932 (Kaspar et al., 2004). Field management was considered typical for Iowa and was described in detail by Karlen and Colvin (1992) and Colvin et al. (1997). The field has been in a 2-yr corn–soybean rotation since 1957 with corn planted in odd-numbered years. From 1932 to 1981, the primary tillage was fall moldboard plowing followed by disking and harrowing. Since 1981, the primary tillage has been fall chisel plowing or disking followed by harrowing before soybean planting. In recent years, only one or two passes with a field cultivator have occurred before corn planting with N applied as anhydrous NH$_3$. Considered typical, fertilizer management consisted of an average 168–38–93 kg N–P–K ha^{-1} (185 kg N, 1996; 157 kg N, 1998) applied in the fall after soybean since 1980. 'Hills 2660' soybean seed was planted in 1996 and Dekalb 'CX289' in 1998. Corn varieties planted in 1997 and 1999 were Pioneer Brand '3489' and Dekalb '621BT', respectively. Soybean and corn yields were obtained on each of the plots using a modified commercial combine equipped with a weigh-tank and capacitance moisture probe to obtain grain weight and moisture (Colvin, 1990). Complete yield details from 1989 are found in Kaspar et al. (2004). Rainfall during the May through August growing season was used as an indicator of plant available water (Kaspar et al., 2003). Daily precipitation totals were obtained from an Iowa State University research farm located 7 km south of the study area (Herzmann, 2004).

Organic Nitrogen Extraction

Soil (250 mg) was treated with 2 mL of 4 M methanesulfonic acid (MSA) for ON extraction and the samples were autoclaved for 60 min at 136°C (112 kPa) (Martens and Loeffelmann, 2003). After sample cooling, centrifugation, and collection of the supernatant, the residue was washed with two 1-mL aliquots of deionized (DI) water and centrifuged between each addition and the three supernatants combined for amino compound analysis. All combined supernatants were titrated to pH 4 to 5 with 5 M KOH, centrifuged to remove precipitate, and then an aliquot was diluted for analysis.

Amino Compound Analysis

The AAs and ASs released as hydrolysis products of acid digestion were separated on a Dionex DX-500 (Dionex Corp. Sunnyvale, CA) ion chromatograph equipped with an AminoPac PA10 column (2 mm i.d.). Separation was achieved with a tertiary water, NaOH (5 to 80 mM), and sodium acetate (125 to 200 mM) gradient for AAs and ASs (Martens and Loeffelmann, 2003). Pulsed amperometric detection was by a Dionex ED-40 electrochemical detector set in the integrated pulsed mode with a gold working electrode. The AAs (catalog no. AA-S-18) and AS standards were obtained from Sigma (Sigma Chemical Company, St. Louis, MO). The chromatographic parameters for the AminoPac column including AA and AS retention times, column capacity factor, and precision and limits of amperometric detection were given by Martens and Frankenberger (1992).

Table 1. Properties of soils used in this study.†

Soil	Classification	Texture	Organic C	Total N	C/N ratio	pH	Sand	Clay
			g kg^{-1}					g kg^{-1}
Zenor	Hapludoll	Sandy loam	10.3	0.70	14.7	7.35	765	76.0
Clarion 1	Hapludoll	Sandy loam	10.4	0.70	14.9	4.99	579	206
Clarion 2	Hapludoll	Sandy clay loam	14.4	0.92	15.6	7.88	604	145
Webster 1	Haplaquoll	Sandy loam	14.2	1.10	12.9	5.21	683	147
Webster 2	Haplaquoll	Sandy loam	18.7	1.43	13.1	6.21	531	161
Webster 3	Haplaquoll	Loam	19.9	1.31	15.1	5.42	513	199
Webster 4	Haplaquoll	Loam	32.2	2.52	12.8	6.81	398	254
Okoboji	Haplaquoll	Silty clay loam	40.3	3.30	12.2	6.18	104	351
Harps 1	Calciaquoll	Silty clay loam	42.3	3.11	13.6	7.46	103	433
Harps 2	Calciaquoll	Silty clay	50.4	3.91	12.9	6.47	186	360

† See Material and Methods for description of analyses used.

The NH_4^+ concentration in the digestion supernatant was determined by steam distillation (Keeney and Nelson, 1982). Total ON recovered from the soil digestions was calculated as AA-N, AS-N, and NH_4^+ released by acid digestion and recovery efficiency was calculated by the ratio of N recovered/total N content.

RESULTS AND DISCUSSION

The combination of management and landscape position has resulted in a wide range of soil properties (100–770 g sand kg^{-1} soil; 76–430 g clay kg^{-1} soil; 10–50 g organic C kg^{-1} soil) representative of the spatial variability present in the 16-ha field (Table 1). The soils were sampled in early May before warm spring soil temperatures resulted in rapid organic matter mineralization and then again in late September (except 1999) following crop harvest. The spring and fall sampling times potentially provide an index of net ON mineralization during the growing season.

The AA and AS components of soil ON content for the 10 soils at each of the five sample times are presented in Table 2. The extraction and detection methodology used consistently detected 18 AAs in all soils that can be classified as protein AA. Three compounds—one nonprotein AA, ornithine, which is a microbial breakdown product of arginine, and two ASs, galactosamine (GalX) and glucosamine (GluX), of microbial origin—were also noted. Inconsistently, very low concentrations of other microbial amino compounds, muramic and diaminopimelic acids were detected in the extracts, but due to the very low concentrations detected, the compounds were not included in the report. It is of interest that in these soils, the concentration of the S-containing AAs, methionine and cystine, were very low or not detected, which may suggest a shortage of available S. The S amino acids are a ready source of S for plants and microbes (Tabatabai and Bremner, 1972).

The sum of the AA-N, AS-N, and NH_4^+–N released by the MSA digestion process (totaled as ON) and the percentage of the total N content for each soil are presented in Table 3. Greenfield (1992) reported that acid extraction of 34 soils released an average 13% of total N content as fixed NH_4^+. In addition to a small amount of potentially clay fixed NH_4^+–N, the NH_4^+–N recovered was the result of AA degradation during the dehydration process that released AA monomers during the acid digestion. Release of NH_4^+ occurs due to the instability of AAs such as glutamine and asparagine (recovered as glutamic acid and aspartic acid, respectively) in the acid digestion conditions (Martens and Loeffelmann, 2003). The total ON as a percentage of soil total N content ranged from 93.1% (Clarion 1 soil) to 66.1% (Harps 2 soil) with an average ON percentage of 87% for the 10 soils for the five dates. The results suggest that the acid digestion and detection methodology employed for this study successfully quantified the soil ON composition as AAs and AS compounds.

Martens and Loeffelmann (2003) reported that increasing digestion times from 1 h to 16 h (standard digestion time for protein analysis) did not increase the recovery of AA-N from the soils with high clay per-

centages. It was also noted in these 10 soils that the recovery of AA-N as a percentage of the total N decreased with high clay contents ($r = 0.72$, $P < 0.05$, data not presented). High clay content of certain soils may degrade the AAs that were released in the acid digestion process. Another factor in the high clay soils studied (lowest areas in the field) was the presence of water-logged soils (Harps 1 and 2 and Okoboji) with heavy spring precipitation. Prolonged wet conditions may result in anaerobic abiotic NO_3^- immobilization into nonAA-N compounds (Davidson et al., 2003) that would result in a lower percentage of the total N content recovered as AA-N.

The average and the standard deviation of the percentage of total amino compound and total amino compound–N concentration of each individual AA and AS from the 10 soils are presented in Fig. 1. Since the amino compound composition percentage from the five sampling dates (May 1997–May 1999) for each soil was nearly identical (Martens and Loeffelmann, 2003), the amino compounds were averaged for the five sampling dates and then each amino compound percentage was averaged over the 10 soils from the 16-ha field. The percentage of each amino compound/total amino compound for the majority of the AAs showed small standard deviations and suggested that the AAs and ASs increased or decreased concomitantly as the total amino compound concentration changed due to crop and time of season. Senwo and Tabatabai (1998) also found similar AA composition uniformity between 10 diverse Iowa surface soils. The six AAs that composed the highest total amino compound concentration percentage in the soils tested (Fig. 1A) were aspartic acid (10.9%), alanine (10.7%), arginine (10.3%), glutamic acid (8.5%), lysine (7.3%), and glycine (6.8%) for a cumulative total of 54.5%. The microbial N compounds GluX (11.9%), GalX (7.5%), and ornithine (4.1%) composed an additional 23.5% of the amino compound composition for a nine amino compound total of 78% of total AA and AS concentrations. When converted to an N equivalent basis (Fig. 1B), these nine compounds (of the 21 AA and AS compounds detected) accounted for 80.9% of the total ON. Two of the AAs, arginine and lysine, accounted for 31.7% of the total ON-N. Laird et al. (2001) reported that positively charged arginine and lysine accounted for nearly all of the AA-N found on the negatively charged clay fractions of a Webster soil because of the cationic nature of these AAs.

Soybean (1996 and 1998) and corn (1997 and 1999) yields for the 10 sampling sites are given in Table 4. The lack of yield data for the two Harps soils for 1996 (555 mm rain May through August), 1998 (528 mm rain May through August), and 1999 (595 mm rain May through August) and the reduced Okoboji yields (1998 and 1999) were due to early season water-logged conditions, which reduced or eliminated plant populations, plant growth, and yield in those two low landscape positions (Colvin et al., 1997). The harvest of corn yield from these depression soils in 1997 (384 mm rain May through August) was due to a lack of ponded water during the spring and early summer while maintaining favorable soil water contents for plant growth. The remaining eight

Table 2. Amino acid composition for the 10 soils sampled from May 1997 to May 1999.

Soil†	Arg	Orn	Lys	GalX	GluX	Ala	Thr	Gly	Val	Hpr	Ser	Pro	Ile	Leu	Met	His	Phe	Glu	Asp	Cys	Tyr
Zenor S97	0.30	0.08	0.17	0.17	0.28	0.23	0.07	0.15	0.07	0.07	0.05	0.03	0.03	0.06	ND	0.06	0.09	0.25	0.31	0.02	0.03
Zenor F97	0.45	0.11	0.20	0.20	0.32	0.31	0.10	0.18	0.09	0.08	0.06	0.02	0.03	0.06	ND	0.05	0.10	0.33	0.45	0.02	0.03
Zenor S98	0.24	0.05	0.11	0.13	0.20	0.12	0.04	0.11	0.04	0.04	0.03	0.03	0.02	0.03	ND	0.03	0.06	0.13	0.22	0.01	ND
Zenor F98	0.44	0.10	0.21	0.22	0.34	0.33	0.10	0.20	0.08	0.16	0.06	0.04	0.03	0.06	ND	0.04	0.10	0.30	0.38	0.02	0.03
Zenor S99	0.40	0.14	0.22	0.23	0.36	0.34	0.12	0.25	0.09	0.11	0.08	0.02	0.05	0.08	ND	0.05	0.10	0.42	0.54	0.03	0.05
Clar1 S97	0.35	0.15	0.31	0.31	0.61	0.42	0.14	0.26	0.12	0.13	0.10	0.03	0.05	0.08	ND	0.08	0.10	0.41	0.42	0.03	0.04
Clar1 F97	0.36	0.16	0.34	0.34	0.58	0.36	0.11	0.22	0.08	0.09	0.07	0.03	0.03	0.05	ND	0.06	0.07	0.34	0.34	0.02	ND
Clar1 S98	0.33	0.17	0.38	0.38	0.60	0.34	0.12	0.22	0.10	0.09	0.06	0.04	0.03	0.05	ND	0.06	0.07	0.31	0.30	0.02	0.02
Clar1 F98	0.42	0.16	0.35	0.34	0.59	0.37	0.15	0.28	0.12	0.13	0.09	0.03	0.05	0.09	ND	0.08	0.10	0.31	0.29	0.02	0.04
Clar1 S99	0.45	0.17	0.37	0.38	0.66	0.41	0.15	0.31	0.11	0.14	0.10	0.02	0.05	0.10	ND	0.11	0.10	0.39	0.37	0.03	0.05
Clar2 S97	0.32	0.14	0.26	0.26	0.43	0.42	0.15	0.28	0.13	0.15	0.09	0.01	0.04	0.09	ND	0.07	0.10	0.48	0.56	0.03	0.06
Clar2 F97	0.24	0.12	0.17	0.18	0.28	0.20	0.09	0.19	0.08	0.07	0.05	0.05	0.03	0.05	ND	0.04	0.04	0.24	0.27	0.01	ND
Clar2 S98	0.10	0.04	0.06	0.06	0.09	0.05	0.03	0.09	0.03	0.03	0.02	0.03	ND	ND	ND	0.04	0.06	0.07	0.14	0.01	0.02
Clar2 F98	0.18	0.08	0.16	0.13	0.24	0.17	0.10	0.17	0.08	0.08	0.05	0.05	0.03	0.06	ND	0.05	0.06	0.17	0.22	0.05	0.04
Clar2 S99	0.32	0.13	0.24	0.23	0.42	0.30	0.14	0.25	0.11	0.14	0.09	0.03	0.07	0.09	ND	0.07	0.09	0.37	0.41	0.03	0.05
Web1 S97	0.64	0.24	0.44	0.51	0.85	0.96	0.25	0.37	0.15	0.06	0.22	0.12	0.08	0.14	ND	0.13	0.17	0.58	0.65	0.04	0.07
Web1 F97	0.50	0.20	0.35	0.43	0.71	0.45	0.19	0.30	0.14	ND	0.07	0.03	0.06	0.10	ND	0.10	0.11	0.53	0.63	0.04	0.04
Web1 S98	0.48	0.20	0.33	0.41	0.69	0.45	0.20	0.31	0.14	0.13	0.12	0.02	0.06	0.10	ND	0.08	0.12	0.44	0.54	0.03	0.04
Web1 F98	0.64	0.24	0.39	0.43	0.75	0.51	0.23	0.35	0.15	0.17	0.15	0.05	0.07	0.12	ND	0.11	0.12	0.58	0.67	0.04	0.06
Web1 S99	0.57	0.21	0.38	0.43	0.79	0.54	0.21	0.32	0.16	0.16	0.13	0.03	0.07	0.12	ND	0.09	0.11	0.57	0.58	0.04	0.05
Web2 S97	0.70	0.33	0.65	0.60	0.91	0.96	0.31	0.61	0.23	0.25	0.19	0.09	0.10	0.19	0.02	0.14	0.14	0.68	0.79	0.04	0.09
Web2 F97	0.62	0.35	0.61	0.51	0.76	0.68	0.25	0.56	0.22	0.16	0.15	0.05	0.09	0.13	ND	0.07	0.13	0.67	0.93	0.03	0.03
Web2 S98	0.53	0.24	0.39	0.30	0.46	0.48	0.22	0.40	0.24	0.12	0.13	0.03	0.06	0.11	ND	0.06	0.24	0.35	0.49	0.03	0.05
Web2 F98	0.85	0.35	0.62	0.62	0.96	0.91	0.30	0.53	0.27	0.23	0.18	0.08	0.09	0.16	ND	0.08	0.24	0.71	0.92	0.04	0.07
Web2 S99	0.82	0.29	0.57	0.56	0.90	0.84	0.28	0.49	0.24	0.23	0.17	0.07	0.08	0.17	0.01	0.08	0.22	0.66	0.83	0.05	0.08
Web3 S97	0.84	0.29	0.61	0.57	0.98	0.85	0.27	0.52	0.21	0.23	0.16	0.06	0.09	0.16	ND	0.09	0.13	0.55	0.82	0.05	0.07
Web3 F97	0.63	0.24	0.44	0.45	0.77	0.61	0.15	0.35	0.13	0.11	0.10	0.04	0.04	0.07	ND	0.07	0.07	0.38	0.59	0.02	ND
Web3 S98	0.71	0.26	0.47	0.48	0.75	0.73	0.24	0.47	0.19	0.17	0.14	0.04	0.07	0.13	ND	0.08	0.11	0.45	0.69	0.04	0.05
Web3 F98	0.83	0.24	0.54	0.51	0.86	1.49	0.25	0.50	0.18	0.22	0.15	0.05	0.08	0.15	ND	0.09	0.10	0.49	0.75	0.04	0.07
Web3 S99	0.76	0.25	0.56	0.53	0.89	0.67	0.23	0.45	0.19	0.17	0.13	0.03	0.07	0.12	ND	0.08	0.10	0.45	0.69	0.04	0.05
Web4 S97	0.94	0.36	0.66	0.65	0.96	1.09	0.37	0.67	0.26	0.28	0.22	0.12	0.13	0.22	ND	0.16	0.24	0.82	1.45	0.08	0.10
Web4 F97	0.89	0.34	0.63	0.67	0.95	0.99	0.34	0.62	0.22	0.26	0.20	0.10	0.12	0.20	ND	0.12	0.21	0.70	1.30	0.07	0.09
Web4 S98	0.90	0.35	0.63	0.60	0.81	0.98	0.35	0.68	0.22	0.26	0.20	0.10	0.13	0.22	0.01	0.10	0.22	0.72	1.51	0.06	0.09
Web4 F98	1.04	0.38	0.70	0.68	1.01	1.26	0.40	0.72	0.29	0.31	0.24	0.14	0.15	0.25	0.01	0.12	0.22	0.77	1.49	0.06	0.11
Web4 S99	0.97	0.32	0.62	0.60	0.98	0.98	0.36	0.65	0.24	0.29	0.22	0.12	0.13	0.22	ND	0.12	0.18	0.73	1.33	0.07	0.10
Oko S97	0.97	0.44	0.73	0.72	1.17	1.11	0.35	0.65	0.30	0.27	0.22	0.12	0.12	0.22	0.01	0.08	0.23	0.85	1.02	0.06	0.10
Oko F97	1.10	0.50	0.92	0.85	1.35	1.29	0.40	0.76	0.34	0.27	0.25	0.15	0.14	0.24	0.02	0.12	0.27	0.92	1.08	0.06	0.10
Oko S98	1.08	0.53	0.82	0.75	1.21	1.31	0.38	0.77	0.30	0.05	0.35	0.25	0.15	0.25	0.02	0.08	0.26	0.74	0.94	0.04	0.12
Oko F98	0.86	0.40	0.60	0.48	0.85	0.95	0.32	0.69	0.29	0.04	0.30	0.20	0.14	0.23	0.02	0.08	0.30	0.52	0.75	0.05	0.10
Oko S99	1.32	0.39	0.96	0.82	1.37	0.82	0.42	0.81	0.43	0.09	0.38	0.28	0.17	0.31	0.02	0.11	0.33	0.96	1.06	0.07	0.15
Harp1 S97	0.78	0.41	0.57	0.67	0.93	0.96	0.39	0.56	0.25	0.28	0.23	0.13	0.15	0.28	0.03	0.08	0.06	0.78	0.72	0.04	0.11
Harp1 F97	0.68	0.40	0.53	0.64	0.83	0.91	0.37	0.56	0.29	0.24	0.22	0.12	0.13	0.24	0.01	0.12	0.24	0.53	0.70	0.05	0.10
Harp1 S98	0.79	0.40	0.55	0.59	0.84	1.13	0.39	0.56	0.32	0.26	0.23	0.13	0.16	0.28	0.02	0.13	0.28	0.41	0.52	0.02	0.12
Harp1 F98	0.80	0.41	0.55	0.62	0.88	1.06	0.38	0.63	0.26	0.24	0.25	0.15	0.15	0.26	0.02	0.12	0.26	0.58	0.68	0.05	0.13
Harp1 S99	0.76	0.48	0.75	0.76	1.04	0.86	0.40	0.73	0.32	0.29	0.25	0.15	0.14	0.24	0.01	0.14	0.23	0.92	1.14	0.05	0.11
Harp2 S97	0.82	0.40	0.60	0.70	1.02	1.06	0.39	0.53	0.30	0.28	0.24	0.14	0.15	0.26	0.01	0.12	0.20	0.57	0.69	0.03	0.10
Harp2 F97	0.73	0.39	0.56	0.68	0.96	0.83	0.35	0.51	0.26	0.24	0.22	0.12	0.13	0.22	0.01	0.10	0.21	0.50	0.57	0.04	0.12
Harp2 S98	0.80	0.39	0.60	0.70	1.00	0.96	0.38	0.54	0.27	0.25	0.23	0.13	0.13	0.23	0.01	0.10	0.18	0.75	0.91	0.04	0.09
Harp2 F98	0.85	0.43	0.63	0.77	1.06	0.95	0.37	0.55	0.25	0.25	0.22	0.12	0.13	0.22	0.01	0.11	0.16	0.74	1.03	0.04	0.08
Harp2 S99	0.86	0.41	0.57	0.70	1.03	1.17	0.43	0.60	0.34	0.33	0.25	0.15	0.16	0.29	0.02	0.09	0.23	0.74	0.64	0.05	0.14

† Clar1 and 2, Clarion 1 and 2; Web1, 2, 3 and 4 Webster 1, 2, 3 and 4; Oko, Okoboji; Harp1 and 2, Harps 1 and 2. GalX and GluX, galactosamine and glucosamine, respectively. S and F stand for spring and fall, respectively.

Table 3. Organic N composition for the 10 soils sampled from May 1997 to May 1999.†

Sample‡	Total							Percentage
	AA	AA-N	AS	AS-N	NH_4^+–N	ON	Soil N	ON/Total N
				g kg^{-1} soil				
Zenor S97	2.47	0.34	0.45	0.03	0.50	0.87	1.00	87.0
Zenor F97	3.15	0.44	0.52	0.04	0.31	0.79	0.90	87.8
Zenor S98	1.62	0.25	0.33	0.03	0.46	0.74	0.81	91.3
Zenor F98	3.22	0.49	0.57	0.04	0.41	0.94	1.20	78.3
Zenor S99	3.66	0.48	0.59	0.05	0.45	0.98	1.05	93.3
Clar1 S97	4.12	0.51	0.93	0.07	0.13	0.71	0.76	93.4
Clar1 F97	3.62	0.46	0.92	0.07	0.11	0.64	0.75	85.3
Clar1 S98	3.66	0.47	0.98	0.08	0.23	0.76	0.82	92.7
Clar1 F98	3.96	0.51	0.93	0.07	0.35	0.93	1.00	93.0
Clar1 S99	4.47	0.58	1.04	0.08	0.36	1.02	1.01	101
Clar2 S97	4.04	0.52	0.69	0.05	0.34	0.91	1.02	89.2
Clar2 F97	2.33	0.30	0.46	0.04	0.42	0.76	0.92	82.6
Clar2 S98	1.69	0.23	0.27	0.02	0.38	0.63	0.72	87.5
Clar2 F98	2.10	0.27	0.36	0.03	0.44	0.74	0.81	91.3
Clar2 S99	3.54	0.46	0.64	0.05	0.36	0.87	1.00	87.0
Web1 S97	6.56	0.85	1.36	0.11	0.52	1.48	1.60	92.5
Web1 F97	4.96	0.62	1.14	0.09	0.39	1.10	1.21	90.9
Web1 S98	4.87	0.61	1.10	0.09	0.53	1.23	1.45	84.8
Web1 F98	5.78	0.76	1.19	0.09	0.41	1.26	1.60	78.8
Web1 S99	5.53	0.70	1.21	0.10	0.53	1.33	1.50	88.7
Web2 S97	7.94	1.04	1.51	0.12	0.69	1.85	2.14	86.4
Web2 F97	6.95	0.91	1.17	0.10	0.65	1.66	1.98	83.8
Web2 S98	4.90	0.68	0.76	0.06	0.79	1.59	1.90	83.7
Web2 F98	8.12	1.07	1.58	0.12	0.67	1.86	2.10	88.7
Web2 S99	7.58	1.00	1.46	0.11	0.44	1.55	1.70	91.5
Web3 S97	7.48	0.99	1.55	0.12	0.74	1.85	1.89	97.8
Web3 F97	5.21	0.70	1.21	0.09	0.83	1.62	1.82	88.7
Web3 S98	6.24	0.84	1.23	0.10	0.49	1.43	1.60	89.2
Web3 F98	7.54	1.03	1.37	0.11	0.50	1.64	1.80	91.3
Web3 S99	6.44	0.86	1.42	0.11	0.43	1.40	1.52	92.1
Web4 S97	9.66	1.28	1.61	0.13	0.69	2.10	2.29	91.8
Web4 F97	8.92	1.17	1.61	0.13	0.51	1.81	2.18	83.0
Web4 S98	9.02	1.21	1.40	0.11	0.66	1.98	2.41	82.7
Web4 F98	10.2	1.27	1.69	0.13	0.69	2.19	2.50	87.6
Web4 S99	9.09	1.22	1.57	0.12	0.83	2.17	2.43	89.3
Oko S97	9.62	1.25	1.89	0.15	0.95	2.30	3.00	80.0
Oko F97	11.0	1.47	2.20	0.17	0.66	2.27	2.85	75.1
Oko S98	10.2	1.37	1.96	0.15	0.73	2.25	3.02	76.4
Oko F98	7.98	1.10	1.34	0.10	1.05	2.26	2.95	87.2
Oko S99	11.0	1.47	2.19	0.17	0.94	2.57	2.95	85.0
Harp1 S97	8.47	1.10	1.72	0.14	0.84	2.07	2.65	78.3
Harp1 F97	7.60	0.98	1.64	0.13	0.81	1.92	2.45	78.0
Harp1 S98	8.59	1.11	1.70	0.13	0.78	2.02	2.40	84.1
Harp1 F98	8.87	1.14	1.83	0.14	0.92	2.20	3.00	73.5
Harp1 S99	9.05	1.17	1.73	0.14	0.93	2.25	2.90	77.4
Harp2 S97	8.30	1.07	1.60	0.13	0.97	2.18	3.41	63.8
Harp2 F97	7.80	1.02	1.47	0.11	0.97	2.12	3.21	66.0
Harp2 S98	8.00	1.08	1.43	0.11	0.91	2.10	3.25	64.7
Harp2 F98	8.31	1.11	1.50	0.12	0.91	2.14	3.41	62.6
Harp2 S99	9.60	1.23	1.79	0.14	0.99	2.36	3.23	73.6

† ON, organic N (measured as total of AA-N, AS-N and NH_4^+-N).
‡ Clar1 and 2, Clarion 1 and 2; Web1, 2, 3 and 4 Webster 1, 2, 3 and 4; Oko, Okoboji; Harp1 and 2, Harps 1 and 2. GalX and GluX, galactosamine and glucosamine, respectively. S and F stand for spring and fall, respectively.

soils that occupy higher landscape positions (Kaspar et al., 2004) had lower corn yields in 1997 (average 6.6% lower) due to the limited moisture compared with 1999 weather conditions. Figure 2 shows the regression analysis between organic C content and total N content and corn yield for 1997. The general trend noted in Table 4 of increased corn yields with increased soil C and N content (Table 1) are also evident in Fig. 2.

The relationships between corn yields (Mg ha^{-1}) in 1997 (all 10 soils) and 1999 (8 soils) and the respective May (spring) soil AA-N concentrations (g m^{-2} for the top 15 cm) are shown in Fig. 3. The AA-N values graphed represent the AAs with the highest percentage AA concentration based on the data from Table 2 and Fig. 1. The regression line for the 1997 data (limited spring precipitation) resulted in a distinct linear trend between the

soil AAs and the resulting corn yields. The regression correlation values (r^2) between the 1997 corn yields (Mg ha^{-1}) and AA-N concentrations ranged from 0.84 for the total AA-N concentration (g m^{-2}, 15 cm) to 0.59 for the glutamic acid and aspartic acid (aspartic acid data were not presented as the regression results were similar to glutamic acid) AA-N (g m^{-2}, 15 cm). Similar values for the y intercept (corn yields) and the change in yield per unit AA (slope of regression line) presented in Fig. 3 for the different AAs again suggests a uniform nature of the AAs contribution to the resulting corn yields. Results of the regression analysis suggest that the AA-N contributes to the large pool of soil N that influences the total N pool (native + fertilizer N) that affects corn yield (Power et al., 1986). Although the statistical relationships between spring soil organic C and total N contents and fall
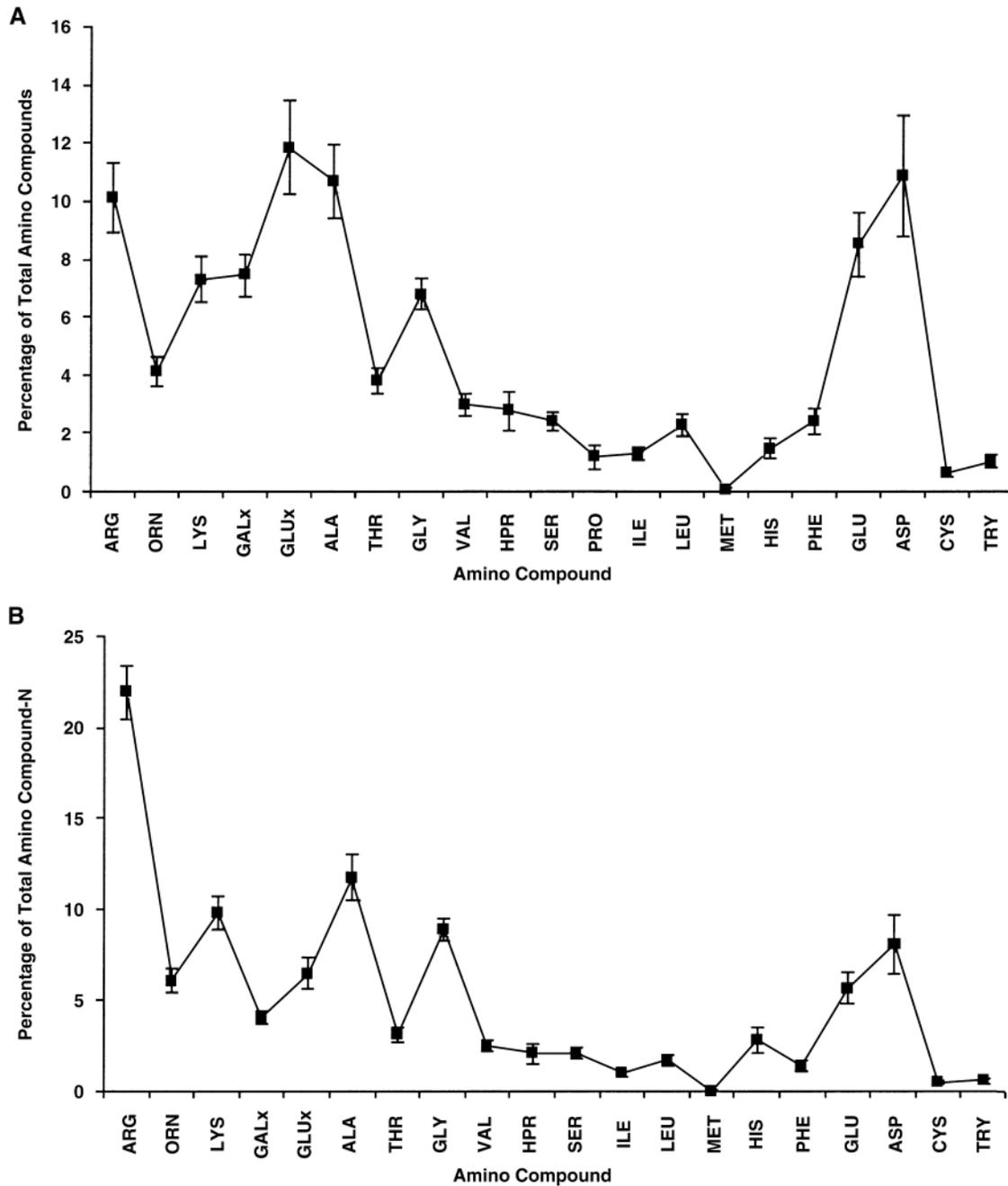
Fig. 1. For each amino acid and amino sugar detected, the average (n = 50) percentage ± standard deviation of (A) the total amino acid and amino sugar concentration and (B) the total amino acid and amino sugar-N concentration.

corn yields show strong trends for increased yield with higher C and N contents (Fig. 2), the relationships noted for the 1997 AA-N and corn yield relationship were better predictors of yield response than those using organic C or total N (Fig. 2).

The 1999 growing season was marked by a period of extensive spring precipitation events with prolonged water ponding that resulted in no corn yield from the two Harps soils and a reduced yield from the Okoboji soil (Table 4). The yields of the two lowest yielding soils (Zenor and Clarion 1), both at the highest topographic field location, changed little in 1999 from 1997, while the five remaining

soils (excluding the Okoboji and Harps 1 and 2) had 32% higher yields in 1999 than in 1997. The regression analysis trends from the 1997 data, which included yields from all 10 soils, were not evident in the 1999 data (regression data not presented).

Mulvaney et al. (2001) and Khan et al. (2001) suggested that the soil AS-N pool was an index to differentiate soils that responded to N fertilizers from those soils that did not. Plants contain low concentrations of GalX and GluX (Sharon, 1974; Martens and Frankenberger, 1990) and the presence of the AS in microorganisms, especially fungi, is the premise for discussion of these

Table 4. Corn yields in 1997 and 1999 and soybean yields in 1996 and 1998.

Soil	Soybean 1996	Corn 1997	Soybean 1998	Corn 1999
		Mg ha^{-1}		
Zenor	1.09	4.50	1.10	3.92
Clarion 1	1.68	5.39	1.77	5.87
Clarion 2	2.92	7.77	3.28	10.6
Webster 1	3.65	9.61	3.42	11.1
Webster 2	3.38	10.6	3.44	11.3
Webster 3	3.28	9.98	3.63	10.8
Webster 4	3.52	11.1	3.40	12.0
Okoboji	3.01	11.2	1.89	9.5
Harps 1	0	10.0	0	0
Harps 2	0	11.1	0	0

ASs as an indicator of microbial activity in soil (Parsons, 1981). In this study, the strong correlations between the ASs GalX and GluX with the 1997 corn yields and similar *y* intercept values (Fig. 4; 1.59– 2.80) compared with the other AAs (Fig. 3; 2.18–3.08) suggests that microbial activity and mineralization of soil ON were important factors that affect corn yields.

The AAs measured in the spring and fall can provide a measure of soil ON contribution to corn yields. Isotopic research has suggested a great contribution of soil N to corn grain production (Power et al., 1986), but little nonisotope data exist to determine the contribution of fertilizer N to corn yields. From the statistical regressions, the *y* intercept for the total AA-N graph was used to approximate the contribution of the N fertilizer applied to corn yields when soil ON = 0. Using the *y* intercept of the 1997 total AA regression line (Fig. 3) suggests that the fall fertilizer N application (185 kg N)

in 1997 accounted for 2.18 Mg corn ha^{-1}. If the 2.18 Mg corn ha^{-1} assumption was subtracted from the total corn yields (Table 4), then from 40% (Zenor) to 81% (Webster 3, Okoboji, and Harps 2) of the 1997 corn yield were contributed by the soil AA concentration.

This fertilizer N contribution assumption appears to be valid as the nonisotope values calculated from the regression analysis are comparable to previous work using ^{15}N labeled fertilizers for soils with comparable fertility levels. Reddy and Reddy (1993) reported fertilizer ^{15}N plant uptake of 53% (47% from native soil N) for a North Carolina soil with an organic C and total N contents similar to the Zenor soil. Similarly, Sanchez and Blackmer (1988) reported that studies on an Iowa Webster soil (12 km south of field site) showed 29% plant uptake from ^{15}N fertilizer (i.e., 71% from native soil N).

The regression analysis of the corn yields vs. the AA concentrations (Fig. 3) and ASs and ornithine (Fig. 4) consistently showed two groups or clusters of soils. The low-yielding soils (3, 1997; 2, 1999) at the bottom left of the graphs (40–60% of yield from soil ON) and the high-yielding soils on the top right portion of the graph (77–81% of yield from soil N) suggests that soils in this field exhibit degrees of responsiveness to N fertilizers. The increased precipitation in 1999 resulted in a higher yield AA and AS separation than noted in 1997, yet the two low-yielding soils did not increase yield with the increased precipitation. Several explanations for this cluster pattern may be possible. The lowest yielding soils may have exhibited water deficiencies even with the higher precipitation due to low soil water-holding capacity during the 1999 growing season. A second possibility was that the N needed for optimizing corn yields on these sandy, low organic matter soils was not available even during wetter years that would favor additional N mineralization. The higher yielding soils may be providing adequate N for crop needs and would not be as responsive to N inputs as the low corn yielding soils.

Currently, soil tests that measure spring NO$_3^-$ concentrations are considered the best option for identifying soils requiring additional N fertilizers (Bundy and Meisinger, 1994). But inherent limitations with the preplant or presidedress NO$_3^-$ tests have resulted in frequent failure to predict N needs due to the transient nature of mineral soil N (Mulvaney et al., 2001). Methodology for analysis of soil AS concentrations by chemical extractions has been promoted as a means to measure potential N fertilizer needed for corn growth and optimum yields (Khan et al., 2001).

To fully understand a soil's N fertilizer requirement to support optimum plant growth, the potential amount of N mineralized during the growing season must be estimated. The data in Table 5 show the spring and fall ON concentrations for a corn production year (1997) and a soybean production year (1998). The fall 1997 sampling showed a decrease compared to the May 1997 sampling that averaged 367 kg N ha^{-1} (top 15 cm) following corn production (value was calculated from the weight of the top 15 cm of soil as kg ha^{-1} multiplied by the amount of ON as g kg^{-1} soil). Net ON mineralization accounted for an average 9.5% of the total ON during
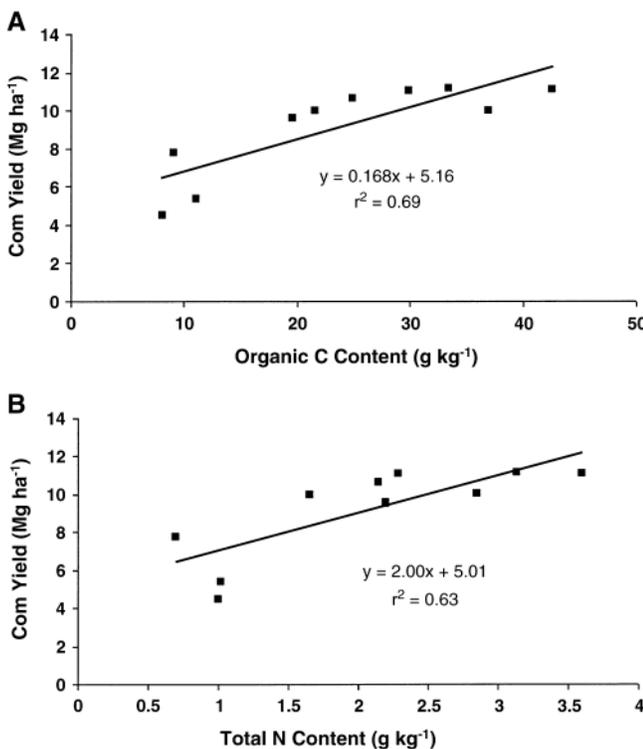
Fig. 2. Relationships between (A) May 1997 soil organic C content and fall corn yields and (B) May 1997 total N content and fall corn yields.
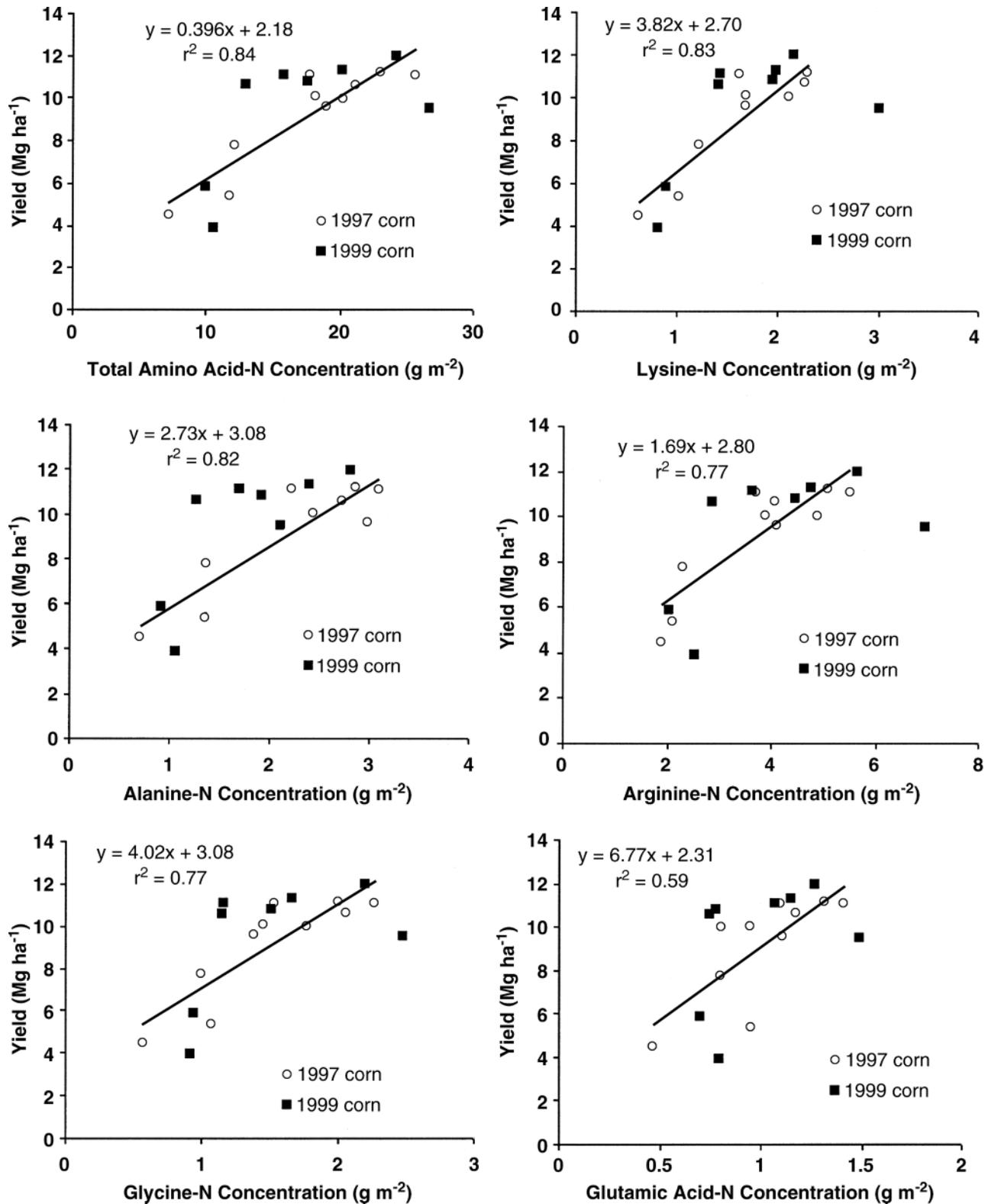
Fig. 3. Relationship of selected amino acid-N concentrations measured in May 1997 (circle) and May 1999 (square) and corn yields for the respective year. The regression equations are for the 1997 values and do not include the 1999 values.

the 1997 growing season (Table 5). This was offset by a 320 kg N ha^{-1} (top 15 cm) enrichment (8.8%) in ON following soybean in 1998. The data in Table 3 show the same pattern of decreased soil total N following corn

growth and enrichment following soybean. The measured changes in total N content during the corn year (1997) also showed a similar average decrease of 334 kg N ha^{-1}. Total N content increased during soybean growth
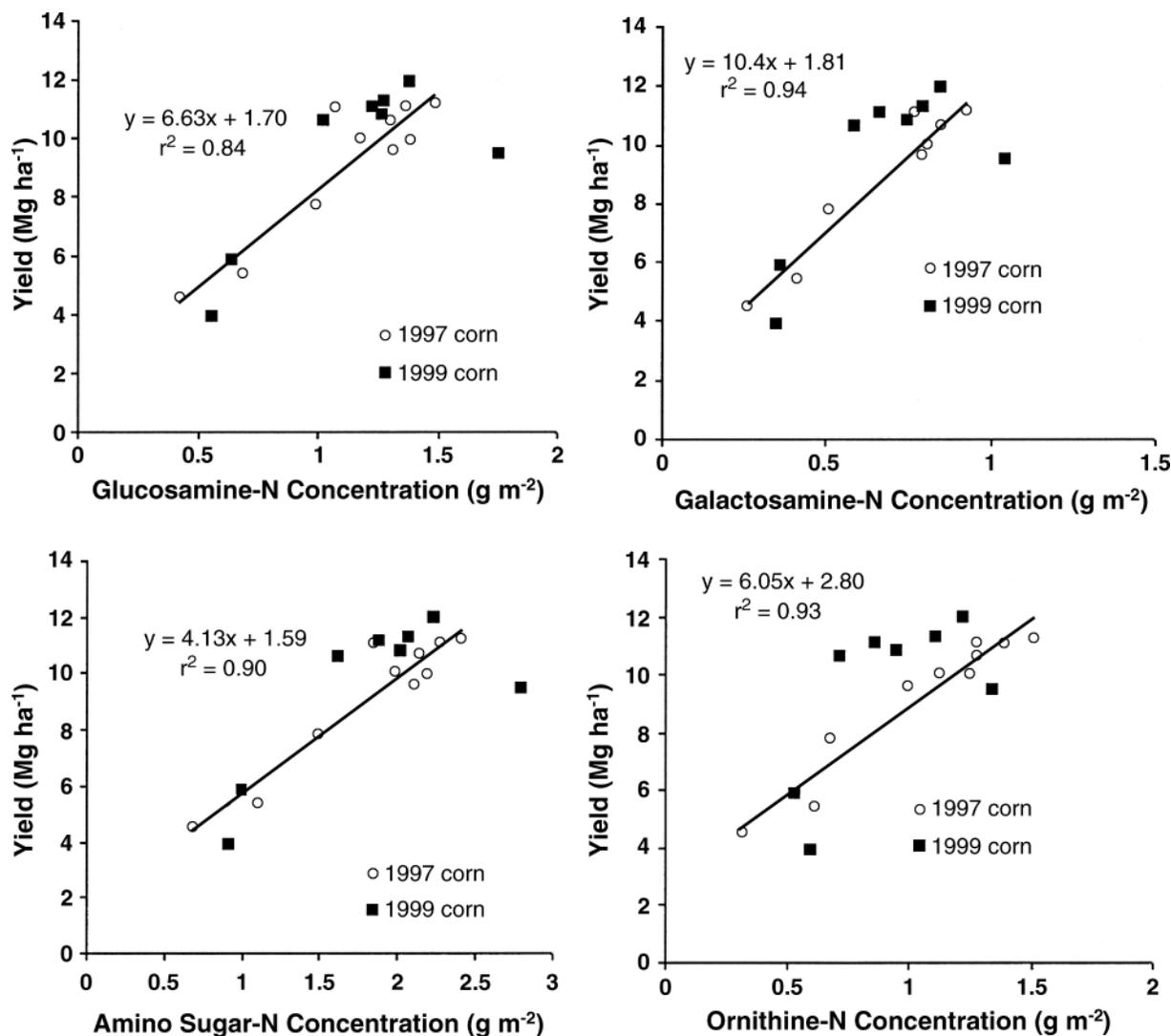
Fig. 4. Relationship of individual and total amino sugar-N and ornithine-N concentrations measured in May 1997 (circle) and May 1999 (square) and corn yields for the respective year. The regression equation are for the 1997 values and do not include the 1999 values.

(1998) by 444 kg N ha^{-1} (Table 3). The increased average total N content (444 kg N ha^{-1}) was inflated by the Harps 1 soil (1346 kg N ha^{-1}) that had no 1998 soybean yield, perhaps due to anaerobic N immobilization or variability

Table 5. Depletion of soil organic N (ON) following corn growth and enrichment of soil organic N following soybean growth (15-cm depth).

Soil	Corn			Soybean		
	May 1997	September 1997	Depleted	May 1998	September 1998	Enriched
	kg ON-N ha^{-1}					
Zenor	1951	1771	179	1659	2108	449
Clarion 1	1592	1435	157	1704	2086	382
Clarion 2	2041	1704	337	1413	1660	247
Webster 1	3327	2463	864	2750	2822	72
Webster 2	4149	3722	426	3566	4171	605
Webster 3	4147	3633	514	3202	3686	484
Webster 4	4715	4059	656	4451	4910	459
Okoboji	5158	5091	67	5046	5058	12
Harps 1	4653	4306	347	4531	4944	413
Harps 2	4880	4754	126	4719	4780	81
Average			367			320

in dry combustion analysis (values were calculated from the weight of the top 15 cm of soil as kg ha^{-1} multiplied by the amount of total N as g kg^{-1} soil). The data suggest that potential immobilization factors influencing total N content would limit the use of changes in total N content as a measure of plant-available N.

Martens and Loeffelmann (2003) reported that the quantity of soil AAs in the spring of 1997 and 1999 were significantly related to soybean yield in 1996 and 1998 ($r = 0.89$, $P < 0.01$). The ON enrichment following soybean was noted shortly after harvest, suggesting the soybean plants were indeed responsible for the increase. The enrichment may be due to root exudation as noted by Mayer et al. (2003). It may also point to one reason why a corn–soybean rotation on high-clay, poorly drained soils under no-tillage management yields as well as a tilled corn–soybean rotation, while continuous no-till corn on the same soils almost always yields less than with tillage (van Doren et al., 1976; Kanwar et al., 1997).

Our results show that some soils within a field have greater potential for mineralizing soil N for plant growth

suggesting that measuring ON concentrations may provide information for optimizing fertilizer N rates. The present concept of using a soil N test as a means of identifying N needs has been based on the "either or" principle of N needs for corn. Either the soil will support an optimum corn yield without N fertilizer or additional N fertilizer is needed. This concept is flawed, because regardless of soil N content in the spring, midwestern soils have the potential to experience prolonged cool and wet conditions. This can result in low soil N mineralization rates (Rice and Smith, 1983), and increased immobilization of plant-available N (Martens, 2001). A deficiency of plant-available N early in the season could decrease yields as important agronomic crops such as corn, sorghum [*Sorghum bicolor* (L.) Moench], and wheat (*Triticum aestivum* L.) genetically express the potential number and size of kernels during the emergence of the growing point from soil (Vanderlip, 1979; Johnston and Fowler, 1991a, 1991b; Dou et al., 1995). A deficiency in soil mineral N during this differentiation period can result in a season long drag on potential yields (Sweeney, 1993).

The season long yield drag noted for early season soil N deficiency appears to be due to several initial plant physiological and developmental processes involved in the uptake of soil NO_3^- or NH_4^+ and the subsequent nitrate-induced transcription that increases lateral root growth (Zhang and Forde, 1998). Furthermore, genetic microarray studies have revealed a number of regulatory and metabolic genes in plants (*Arabidopsis* as study plant) that are N-regulated (Wang et al., 2000). Thus, when plant-available N is limited, restricted activation of regulatory genes (C/N-sensing mechanisms) will result in slow root growth (less nutrient uptake) and low photosynthesis rates (slow growth and development) due to N limitations on metabolism and transport of the C skeletons from source to sink tissues. Experimentally, Evanylo (1991) showed that 32 kg N ha^{-1} must be available by the fifth leaf stage to prevent N deprivation and yield potential reduction.

The exact contribution of the ON content to the yield potential for an individual soil that is part of a soil mosaic across an agricultural field may prove difficult if not impossible to separate from the other factors contributing to yield such as climate, organic C content, soil texture, water holding capacity, etc. Nitrogen use efficiency results have shown that the vast majority of N assimilated by corn plants is from soil N. This report suggests that the cycling of a soil N pool composed of AA and AS contribute to the soil N assimilated by corn. Soil ON content enrichment was noted in the fall following soybean growth suggesting that the soil N is supplemented during soybean growth. Soil tests that would provide estimates of spring soil ON pools for corn growth and yields would be useful to help reduce fertilizer use in excess of plant needs. This spring ON hypothesis is supported by the data that seven out of the 10 study sites in the 16-ha field studied would supply adequate N for optimum growth with smaller N fertilizer additions than currently applied. Although a large net seasonal N mineralization was noted by the AA and AS analysis in these seven soils, maximum corn yields may still require an early season N addition near planting to compensate for potential periods of low soil N availability.

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