Communications in Soil Science and Plant Analysis, 37: 1571–1586, 2006

ISSN 0010-3624 print/1532-2416 online
DOI: 10.1080/00103620600710124

Nitrogen Fractions in Arable Soils in Relation to Nitrogen Mineralization and Plant Uptake

Mabel M. Bregliani
Rio Gallegos' Academic Unit, Austral Patagonia National University,
Rio Gallegos, Argentina

Erwin J. M. Temminghoff and Willem H. van Riemsdijk
Department of Soil Quality, Wageningen University,
Wageningen, The Netherlands

Ernesto S. Haggi
Rio Gallegos' Academic Unit, Austral Patagonia National University,
Rio Gallegos, Argentina

Abstract: Nitrogen (N) as a major constituent of all plants is one of the most important nutrients. Minimizing input of mineral nitrogen fertilizer is needed to avoid harm to the environment. Optimal input of mineral nitrogen should take the nitrogen supply of the soil into account. Many different soil tests have been proposed for determining soil nitrogen availability. In this article we present a new approach that is based on the measurement of nitrate, ammonium, and dissolved organic nitrogen (DON) in a 0.01 *M* $CaCl_2$ soil extract. Eighteen agricultural soils, differing widely in the availability of nitrogen were used, fertilized and unfertilized. It is shown that the nitrogen uptake by maize plants (*Zea Mays* L.) in both "N-fertilized" and "N-unfertilized" soils as measured in a pot experiment can be described with a simple model using the measured nitrogen fractions in the extract. The main source of nitrogen uptake by the plants is the mineralized organic nitrogen during the growing period.

Received 28 February 2005, Accepted 21 September 2005
Address correspondence to Erwin J. M. Temminghoff, Department of Soil Quality, Wageningen University, P.O. Box 8005, 6700 EC Wageningen, The Netherlands. E-mail: erwin.temminghoff@wur.nl

It is shown that the initial measured DON fraction is a good indicator of the nitrogen mineralized during plant growth.

Keywords: Arable soil, $CaCl_2$ extraction, DON, mineralization, mineral nitrogen, nitrogen fractions

INTRODUCTION

Among elements essential for plant growth, nitrogen (N) occupies a unique position because relatively high amounts are required by most agricultural crops for optimal yields (Black 1968; Brady 1990; Stevenson 1982; Stevenson and Cole 1999a, 1999b). Although some plants acquire N from the air by symbiotic N fixation, most plant species rely on uptake of nitrate (NO_3^-) and ammonium (NH_4^+) from the soil solution (Blackmer 2000). The main sources of N for these crops are mineralization of soil organic matter, inorganic fertilizers, and organic inputs. To improve the effectiveness of N management at the farm, it is important to be able to estimate how much N can be supplied by the soil during the growing season (Sogbedji et al. 2000). Soils differ widely with respect to the potential N supply. Because mineralization is a microbial process, actual N supply of a particular soil depends on factors such as temperature and moisture content of the soils, which are dependent on the weather. An estimation of the potential N supply can thus best be based on methods that are calibrated under standard conditions as measured in the lab (Dendooven, Murphy, and Powlson 2000).

A variety of chemical or biological soil tests have been proposed to predict the soil N availability to crops (Bundy and Meisinger 1994; Stanford 1982; Keeney 1982; Hergert 1987; Hood et al. 2000). The tests range in severity of extraction from drastic (e.g., total organic N) to intermediate (e.g., alkaline potassium permanganate) to mild (e.g., hot water and neutral salt extractions). For example, mineral N fraction in fresh soil samples after extraction with 1 M potassium chloride is often used for fertilizer recommendation (Lathwell, Dubey, and Fox 1972). The link between these chemical extractions and the mineralization that may occur over a certain time period for standard conditions is not clear. Recently it has been suggested that the organic N extracted with 0.01 M $CaCl_2$ might give a reasonable correlation with the potential N supply of soils (Nunan et al. 2001). This study investigates this idea further but also includes in the analysis the mineral N fractions as measured in the extract.

The extraction procedure based on 0.01 M calcium chloride can be used to measure not only the NO_3^- and NH_4^+ fractions but also additionally the dissolved organic N fraction (DON) (Houba, van der Lee, and Novozamsky 1997, Houba et al. 2000; Van Erp, Houba, and Van Beusichem 1998, 2001; Van Erp 2002). Nowadays researchers are increasingly taking into account the importance of the DON fraction. This fraction is regarded in soils as a significant N pool from which plant N uptake may occur (Murphy et al. 2000). Many studies have been published dealing with the interpretation of the DON and other N fractions as plant nutrients (e.g., Németh et al. 1979; Appel and Mengel 1990, 1992; Houba, Jázberényi, and Loch 1991; Yu, Northup, and Dahlgren 1994; Hood et al. 2000; Kempl et al. 2001; Nordin, Hogberg, and Nasholm 2001; Smolander, Kitunen, and Malkonen 2001; Verchot et al. 2001). However, there are many gaps in our understanding about the DON fraction with respect to N uptake by plants. The aim of this article is to investigate in how far the initial DON fraction measured in 0.01 *M* $CaCl_2$ extract can be used as a predictor for N mineralization as measured under standardized conditions for a series of 18 soils, which differ widely in N fertility. Besides this, we investigated how the various measured N fractions in the extract can be used to describe measured N uptake in a short-term growth experiment using maize as a test plant.

MATERIALS AND METHODS

Pot Experiment

Soils from 18 different agricultural locations in The Netherlands were selected to obtain a wide range in organic C and N content (resulting in different C/N ratios), clay content, and pH (Table 1).

The soils were air dried and passed through a 2-mm sieve, and a pot experiment was set up using 1.5 kg of each soil per pot. Three pot experiments were performed in a greenhouse in February–March (experiment 1) and June–July (experiments 2 and 3). During experiments 1 and 2, no N fertilizer ("minus N") was used, whereas during experiment 3, N fertilizer was used ("plus N"). The "minus N" treatment of experiments 1 and 2 were taken as an independent replicate.

Treatments with N fertilizer were at a rate of 100 mg of N per kg of soil in the form of NH_4NO_3. All pots were also fertilized with a nutrient solution of P and K (12.5 mg P and 30 mg K per pot) and 2.5 mL of a solution of trace elements (0.5 mg/L of boron, manganese, and zinc; 0.02 mg/L of copper; and 0.01 mg/L molybdenum) to assure that N was the growth-limiting factor. Five seeds of maize (*Zea Mays* L.) were sown per pot. Water was added to all pots to reach field capacity (60% of the water-holding capacity). The pots were watered daily by weighing to keep the moisture content constant at field capacity. During the February–March pot experiment, additional light was provided for 16 h per day, at an intensity of $48\,W\,m^{-2}$. After 60 days of growth, the shoot part was harvested, dried at 70°C for 24 h and weighed. The (visible) roots were removed from the soils by hand, washed, dried at 70°C, and weighed. All plant samples were milled and stored for analysis. The soils were air dried and sieved (<2 mm) for further analysis.

Table 1. Some physical and chemical characteristics of the 18 soils used in the pot experiment

Sample number	Clay fraction $<2\,\mu m$ (%)[a]	pH[b] ($CaCl_2$)	Organic carbon[c] ($g\,kg^{-1}$)	N total[d] ($g\,kg^{-1}$)	C/N
1	4	4.3	38	2.3	17
2	8	5.8	37	2.7	14
3	2	6.8	10	0.8	13
4	15	4.7	13	1.3	10
5	3	4.9	13	0.8	16
6	11	5.5	9	1.0	9
7	3	5.3	18	1.2	15
8	25	4.8	83	5.2	16
9	7	7.1	12	1.0	12
10	28	5.2	140	8.3	17
11	23	7.5	16	1.2	13
12	26	7.4	18	1.9	9
13	52	6.9	33	3.5	9
14	38	4.9	88	5.5	16
15	6	5.9	10	0.6	17
16	46	7.6	31	3.4	9
17	12	7.2	29	2.8	10
18	12	5.2	21	1.7	12

[a]Determination of the clay fraction by pipet method (Houba, van der Lee, and Novozamsky 1997).

[b]Determination of the pH in a 1 : 10 (w/V) suspension of soil in a solution of 0.01 *M* $CaCl_2$ (Houba et al. 2000).

[c]Determination of organic carbon by sulfochromic oxidation and spectrophotometry (Houba, van der Lee, and Novozamsky 1997).

[d]Determination of the total nitrogen by spectrophotometry (CFA) after digestion with concentrated sulphuric acid–salicylic acid–hydrogen peroxide and selenium (Novozamsky et al. 1983).

Incubation Experiment

Simultaneously with the pot experiment, an incubation experiment was set up using the same soils. Only soils were used that had not received N fertilizer in the lab.

The soils were incubated in bags made of special gas-permeable plastic (Audiothene 0.10 mm, Art. N°: a15100). The soils were wetted to field capacity and mixed. A bag was filled with 30 g of each soil (in duplicate) and sealed to minimize moisture changes. During the incubation, the bags were kept in the dark and left under the same temperature conditions of the pot experiment.

After 60 days of incubation (which is the same time as used for the plants to growth during the pot experiment), each bag of each soil was extracted with 300 mL of 0.01 *M* $CaCl_2$ (in duplicate) to measure the rate of mineralization.

Analytical Methods

The soils from the pot experiment and the incubation experiment were extracted with 0.01 M $CaCl_2$ at a 1 : 10 (w/v) soil–solution ratio at $20 \pm 2°C$ for 2 h (Houba et al. 2000). The concentrations of total dissolved N, NO_3^--N, and NH_4^+-N were measured spectrophotometrically in the extract using a Skalar segmented flow analyzer; dissolved organic carbon (DOC) was measured using an automatic carbon analyzer, model SK 12, Skalar, Breda, The Netherlands (Houba et al. 2000). The DON can be calculated by subtracting NO_3^--N and NH_4^+-N from the total dissolved N.

The dry plant samples were digested with H_2SO_4–Se–salicylic acid–H_2O_2 in duplicate (Novozamsky et al. 1983). In the digests, the total N content was measured spectrophotometrically by a Skalar segmented flow analyzer.

Statistical Analysis

The variables determined were analyzed using the statistical program SPSS for Windows 7.0 (1995). Data from replicate determinations were characterized by computing means, standard deviations, coefficient of variations, and linear regression. The duplicate analysis of each soil showed a coefficient of variation within 10% for NO_3^-, 9% for NH_4^+, and 4% for total dissolved N. Pairwise t-tests were performed for the means values of the data from the replicates of the pot experiment; no statistical significant difference was found at a significance level of 0.01.

Multiple linear regression analysis was employed to quantify the relationship between the different initial soil N fractions in $CaCl_2$ extract and N plant uptake.

RESULTS AND DISCUSSION

The maize plant (*Zea Mays* L.) grown in the various soils varied strongly in total dry matter yield (TDM: sum shoots and roots) and N uptake (Table 2). To facilitate the comparison, the same units, mg N per kg soil, are used for both the N plant uptake and the $CaCl_2$ concentration. To do so, the weight of soil employed in each pot was taken into account. The total dry matter varied from 6 to 41 g/kg soil, whereas the N uptake varied from 35 to 375 mg/kg soil. In all soils, the dry matter yield and N uptake were higher in the "plus N" treatment than in the "minus N" treatment, showing that N

Table 2. Yield of maize plants (shoots and roots, dry) and mean total nitrogen uptake by the plants (shoots and roots) for the A) soils without added nitrogen fertilizer, experiment 1 (February–March), and experiment 2 (June–July), and B) soils, which received additional nitrogen fertilizer, experiment 3 (June–July)

	A, minus N				B, plus N	
	TDM[a] ($g\,kg^{-1}$)		N plant uptake ($mg\,kg^{-1}$)		TDM[1] ($g\,kg^{-1}$)	N plant uptake ($mg\,kg^{-1}$)
Sample number	exp. 1	exp. 2	exp. 1	exp. 2	exp. 3	exp. 3
1	11	12	73	74	18	133
2	17	20	105	125	26	181
3	9	13	79	89	15	150
4	8	16	42	69	25	140
5	6	7	35	38	23	112
6	12	15	74	89	26	146
7	18	28	208	170	29	240
8	20	25	107	115	30	153
9	9	10	49	47	28	128
10	26	36	298	323	39	375
11	7	9	43	41	26	110
12	11	14	81	89	18	161
13	20	27	158	185	34	244
14	26	31	193	219	32	284
15	6	6	38	39	20	113
16	23	29	160	165	32	238
17	19	26	91	109	37	173
18	27	33	164	153	41	212

[a]TDM: Total dry matter of plant material in g per kg soil.

is a growth-limiting factor in the controls. There was no statistical difference between the yields as well as for N uptake obtained for the duplicates of the "minus N" pot experiment (exp. 1 and exp. 2) [$n = 18$, $t = 2.05$, $\alpha = 0.05$ (t critical $= 2.10$)].

The $CaCl_2$ contents of the different N species (N-NO_3^-, N-NH_4^+, and DON), as well as for DOC, varied strongly for the 18 soils (Table 3). The NO_3^- fraction for all soils varies from 6 to 125 $mg\,kg^{-1}$ NO_3^--N, NH_4^+ from 1 to 71 $mg\,kg^{-1}$ NH_4^+-N and DON varies from 2 to 36 mg $DON \cdot kg^{-1}$ and the total dissolved N from 8 to 201 $mg\,N\,kg^1$ soil. The DOC varies from 66 to 698 $mg\,kg^{-1}$. The large variation in the different N species makes the data set very suitable to test the effect of various N species in the extract on N uptake and the N mineralization. In general, the amounts of NH_4^+ in Dutch arable soils are limited because of relatively high rates of nitrification. However, at low soil pH, the rate of nitrification is lower, and higher amounts of NH_4^+ are possible. Also, shortly after manure or fertilizer application,

Table 3. Mean nitrogen concentration of various fractions, N-NO_3^-, N-NH_4^+, N_t, DON, and DOC contents in the soils without added N fertilizer as determined in the 0.01 *M* $CaCl_2$ soil extracts, expressed in mg N per kg soil

Sample number	N-NO_3^- ($mg\,kg^{-1}$)	N-NH_4^+ ($mg\,kg^{-1}$)	N_t ($mg\,kg^{-1}$)	DON ($mg\,kg^{-1}$)	DOC ($mg\,kg^{-1}$)	DOC/DON
1	6.8	4.6	16.7	5.3	131	25
2	39.0	4.7	50.7	7.0	158	22
3	43.1	5.7	53.3	4.5	78	17
4	1.6	2.3	11.1	7.2	178	25
5	10.3	2.5	15.1	2.3	66	29
6	45.2	2.2	53.2	5.8	106	18
7	125.4	71.1	201.2	4.7	76	16
8	8.2	5.7	26.5	12.6	361	29
9	6.1	2.2	11.8	3.5	81	23
10	96.0	11.2	143.4	36.2	698	19
11	3.3	1.1	7.8	3.4	121	36
12	32.7	2.4	42.4	7.3	174	24
13	27.1	2.5	46.0	16.4	415	25
14	54.8	6.1	79.6	18.7	596	32
15	4.9	2.1	10.6	3.6	70	19
16	21.2	0.8	36.8	14.8	394	27
17	9.2	3.0	20.3	8.1	226	28
18	101.7	3.8	123.3	17.8	194	11

higher amounts of NH_4^+ can be found. The DOC/DON ratio (Table 2) is higher than the C/N ratio in the soil in all cases except one (soil no. 18) (Table 1). The C/N ratio varied from 9 to 17, and the DOC/DON ratio varied from 11 to 36. In general, it is assumed that the mineralization of (soil) organic N is higher at lower C/N ratio. The lowest C/N ratio in the soil does not, however, correspond with the lowest DOC/DON ratio in the $CaCl_2$ extraction.

In Figure 1, the N uptake is given as a function of the N mineral (sum of NO_3^--N and NH_4^+-N). A relatively poor correlation is obtained between the initial concentration of the mineral N fraction and plant N uptake ($R^2 = 0.50$, $n = 54$, $\alpha = 0.05$). For nearly all soils, more N was taken up by the maize plants than the amount that would have been expected on the basis of the mineral N initially available in the soils. This indicates that the mineral N pool is replenished by N originating from the mineralization of organic matter during the growth experiment. Only for one soil (soil no. 7, a sandy soil with high amounts of NO_3^- and NH_4^+) was the initial amount of mineral N high enough to supply the maize plants completely during the duration of the experiment. Besides mineral N, N compounds, which can easily be mineralized during crop growth, also need to be taken into account (Murphy et al. 2000). Therefore, DON could be a good estimator

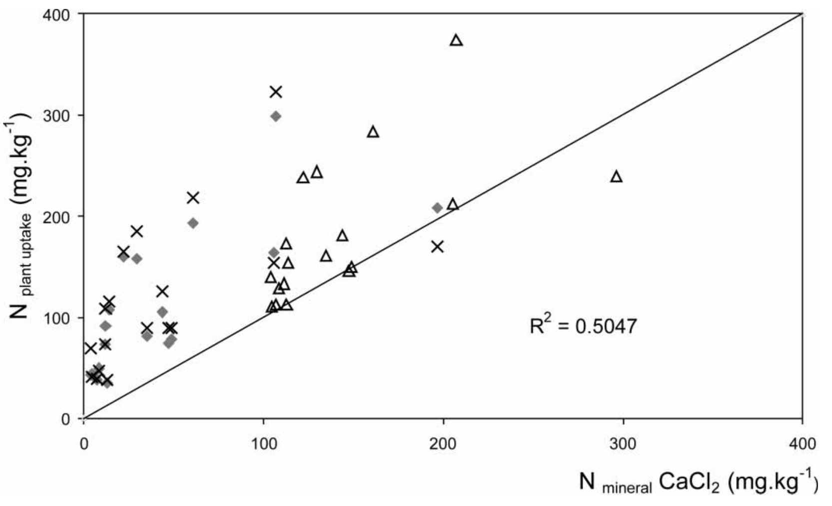

Figure 1. Relationship between the mineral nitrogen available in the soils measured in a 0.01 *M* $CaCl_2$ extract and the nitrogen uptake by the maize plants (units mg N per kg soil). "Minus N" soils experiment 1 (February–March) (◆) and experiment 2 (June–July) (×) and the "plus N" soils experiment 3 (△). Note: The same units are used for the concentration in the extract and the plant uptake (mg N per kg soil), dividing N content in plant tissues by pot soil weight.

(e.g., Németh et al. 1979; Appel and Mengel 1990, 1992; Houba, Jászberényi, and Loch 1991). To test the relative importance of the different measured N fractions as predictors for the availability of N to the plants, a multiple linear regression analysis was performed on the data obtained from the first "minus N" pot experiment soils using the SPSS statistical program. The dependent variable used was the N plant uptake, and as independent variables NO_3^--N, NH_4^+-N, and the DON initial concentrations measured in the $CaCl_2$ extract expressed in mg N per kg soil were used. The regression model is given by

$$N_{\text{plant uptake}} = 20.5 + 0.27[NO_3^- - N] + 1.68[NH_4^+ - N] + 6.88[DON] \quad (1)$$

The model explains 96% of the variability among N plant uptake. Using Eq. (1), we predicted the N plant uptake ($N_{\text{plant uptake}}$ model) for experiment 2 ("minus N") and 3 ("plus N") by using the NO_3^--N, NH_4^+-N, and the DON initial concentrations measured in the $CaCl_2$ extract. The calculated N uptake by the plant ($N_{\text{plant uptake}}$ model) is given as a function of the measured N ($N_{\text{plant uptake}}$ measured) in Figure 2. It shows that the N plant uptake can be described very well with the model and that the same regression model gives a good prediction not only for the unfertilized soils (exp. 2 "minus N": $R^2 = 0.91$) but also for the N fertilized soils (exp. 3 "plus N": $R^2 = 0.90$).

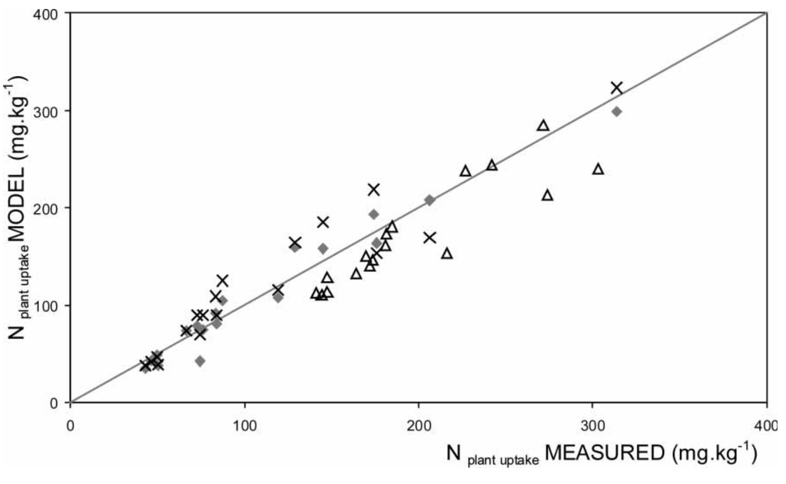

Figure 2. Nitrogen uptake by the plants calculated using the developed regression model [Eq. (1)] in relation with the same value measured in the growth experiment: "minus N" soils, experiment 1 (◆) ($R^2 = 0.96$) and experiment 2 (×) ($R^2 = 0.91$), and for experiment 3 the "plus N" (△) ($R^2 = 0.90$) soils. For the two last data sets combined, $R^2 = 0.90$.

By making use of Eq. (1), the contribution of NO_3^--N, NH_4-N, and the DON to the total N uptake by the plant can be calculated (Table 4). It is remarkable that NO_3^--N only explains a small fraction (<13%) of the $N_{plant\ uptake}$ for the unfertilized soils. The NH_4^+ concentration also contributes relatively little according to the model, ranging from 1 to 11% with the exception of one soil (soil 7: 56%), which has a very high initial NH_4^+-N (and NO_3-N) content. The most important fraction is DON, which contributes between 37 and 82% for the unfertilized soils with again the exception of soil 7 (only 19%). For the fertilized soils, the contribution of the measured nitrate concentration is quite similar to the unfertilized soils where the measured ammonium concentration in the extract becomes more important (30–60%), again with the exception of soil 7 (66%). The relative importance of DON to explain the measured uptake according to the regression model is in general very high; it explains from 19% of the uptake, in soils with a high concentration of mineral N, to a maximum of 79%, in soils with relatively low concentrations of mineral N in the extract. This is in accordance with Maci and Mengel (2001), who found that Electro Ultra Filtration organic N (EUF-N_{org}) was the main N source for *Lolium multiplirum*. For the "plus N" soils, the effect of DON was only slightly less important, explaining between 13 and 65% of the uptake. In the fertilized soils, there is a N source that the plants can use more easily. Kempl et al. (2001) developed an analogous model for the prediction of N fertilization of sugar beets using EUF extracts; however, they used only NH_4^+-N fraction and EUF-extractable

Table 4. Contribution of the different nitrogen fractions as present in the extract to the nitrogen plant uptake for A) exp. 2 "minus N" soils and B) exp. 3 "plus N" soils as calculated by the developed regression model [Eq. (1)]

	A, minus N soils				B, plus N soils			
	$N_{plant\ uptake}$ MODEL	Contribution to $N_{plant\ uptake}$			$N_{plant\ uptake}$ MODEL	Contribution to $N_{plant\ uptake}$		
Sample number	(mg/kg)	$N\text{-}NO_3^-$ (%)	$N\text{-}NH_4^+$ (%)	DON (%)	(mg/kg)	$N\text{-}NO_3^-$ (%)	$N\text{-}NH_4^+$ (%)	DON (%)
1	71	1.8	9.6	55.5	154	7.0	52.4	25.4
2	90	8.3	7.8	57.8	173	9.8	46.8	29.9
3	73	11.2	11.5	45.4	157	11.3	52.6	21.2
4	80	0.4	4.2	66.3	164	6.0	47.2	32.5
5	46	4.2	8.0	36.9	130	8.8	60.0	13.1
6	78	11.0	4.2	54.9	162	11.2	47.8	26.5
7	187	12.7	56.2	18.6	271	12.3	66.2	12.8
8	127	1.2	6.7	73.6	210	5.3	39.2	44.4
9	54	2.2	6.1	48.2	137	7.8	56.3	18.9
10	326	5.6	5.1	82.1	410	6.8	22.1	65.4
11	51	1.2	3.2	49.5	134	7.5	56.3	18.7
12	87	7.1	4.1	62.0	171	9.2	45.4	31.6
13	154	3.4	2.4	79.0	237	6.2	32.8	51.2
14	181	5.7	5.0	76.4	265	7.5	31.4	52.3
15	54	1.7	5.7	49.3	138	7.6	56.0	19.4
16	138	2.9	0.9	79.3	222	6.1	33.9	49.4
17	90	2.0	5.0	67.0	173	6.5	45.3	34.6
18	180	10.7	3.1	73.2	264	10.9	30.2	50.0

organic C instead of dissolved organic N. They found a relative low coefficient of determination of 0.56 for only NH_4^+-N, which was improved by incorporating EUF-extractable organic C into the model ($R^2 = 0.80$). So by using DON instead of EUF organic C, the plant N uptake model can be further improved. However, the uptake of N is still unclear. The extra N taken up by the plants, besides mineral N, can be from the mineralization of the soil organic N fraction and/or the uptake of DON. To study this in more detail, the data obtained from the incubation experiment was used (Table 5). In Figure 3, the N plant uptake is given as a function of the sum of the mineral N and the amount of N, which has been mineralized during the incubation experiment for 60 days. For all soils, the total N uptake by the maize plants (roots and shoot) can be fully explained by the sum of the mineral N and the mineralized amount of N during the incubation experiment. The results are very close to the 1 : 1 line. Only at high N contents (>250 mg/kg) was a lower N plant uptake found with respect to the available N in the soil. Probably above 250 mg/kg N the plants do not need all available N in the soil for optimal growth. The improvement by taking the mineral N and the mineralized amount of N during the incubation experiment is similar to the improvement by taking DON into account by using Eq. (1). This suggests that DON may be used as an indicator for the potential N mineralization during plant growth. In Figure 4, the relationship is given between N

Table 5. Mean nitrogen species (N-NO_3^-, N-NH_4^+, N_t, DON) and DOC concentrations in the $CaCl_2$ extracts for the incubation experiment after 60 days of incubation

Sample number	N_t (mg kg^{-1})	N-NO_3^- (mg kg^{-1})	N-NH_4^+ (mg kg^{-1})	DON (mg kg^{-1})	DOC (mg kg^{-1})	DOC/DON
1	76	68	1	7	38	5
2	122	117	1	5	43	9
3	104	98	1	5	45	9
4	76	66	2	8	37	5
5	32	26	1	5	33	7
6	75	71	1	3	28	9
7	194	185	2	7	35	5
8	135	119	1	15	73	5
9	46	45	1	0	22	—
10	469	390	0	79	191	2
11	43	39	1	3	47	16
12	97	93	0	4	57	14
13	156	149	0	7	97	14
14	272	253	1	18	142	8
15	39	35	1	4	22	6
16	79	71	2	6	106	18
17	83	81	0	2	49	25
18	201	188	2	11	58	5

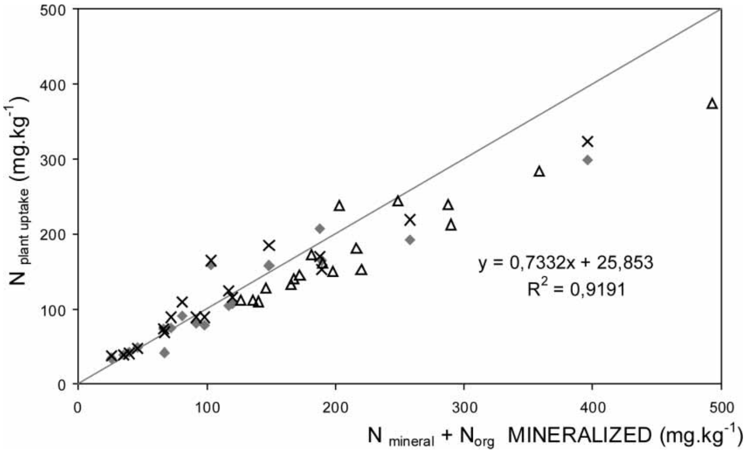

Figure 3. Relationship between the nitrogen uptake by the plants and the sum of the initial mineral nitrogen in the soils and the mineralized organic nitrogen after 60 days of incubation for the "minus N" soils, experiment 1 (◆) and experiment 2 (×), and the "plus N" soils, experiment 3 (△).

mineralized and the DON content in the extract of soils that received no additional N fertilizer in the lab. There is a good correlation between these two parameters ($R^2 = 0.87$), which shows that DON can indeed be used as a good estimator of the amount of N that is mineralized during a certain period under standardized conditions. The amount of N mineralized during the growth period (Table 5) is on average about eight times higher than the

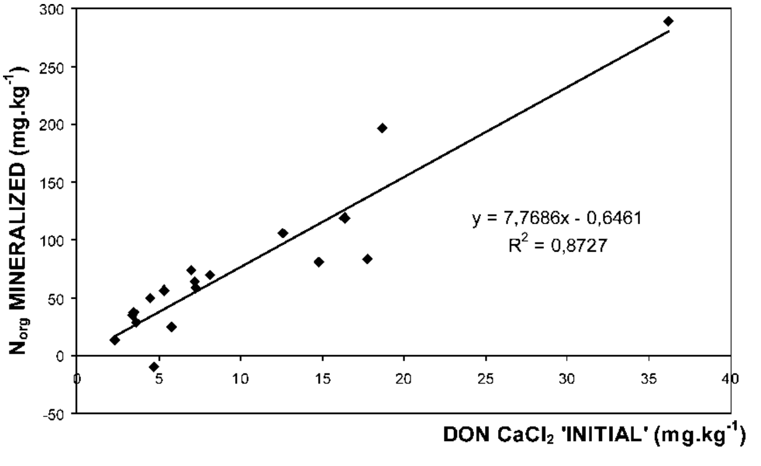

Figure 4. Relationship between the organic nitrogen mineralized during the incubation experiment and the dissolved organic nitrogen (DON) concentration measured in the initial extract.

initial amount of $CaCl_2$-extractable DON in the soil (Table 3). The DON content before and after incubation is rather constant. This strongly suggests that DON has been replenished during the growth period. The direct link between the amount mineralized in the incubation experiment, the amount taken up by the plant, and the measured DON concentration in the extract suggests that, at the conditions of our experiments, mineralization of N is the dominant mechanism and not direct uptake of DON. However, the initial DOC/DON ratio (Table 3) is much higher than at the end of the incubation experiment (Table 5), which suggest that the mineralization rate is changed during our experiment.

Although the model has been obtained with the data from a pot experiment that lasted for a time much shorter than a growing season (only 60 days), it is expected that the model is also useful for field conditions. However, more research is needed (e.g., effect of temperature and porewater content in the soil) on the relationship between DON and the rate of N mineralization to gain more knowledge on N availability and losses under field conditions.

CONCLUSIONS

The uptake of N by maize plants (roots and shoots), measured in a short-term pot experiment, is poorly correlated with the $N_{mineral}$ (NO_3^--N, N-NH_4^+-N) fraction in soil. Measuring various soluble N pools, NO_3^--N, NH_4^+-N, and DON in a single 0.01 *M* $CaCl_2$ extracts leads to much better insight in N availability than merely measuring total dissolved N or soluble mineral N. In most soils, the main source of the N uptake by plants is the mineralized organic N. For maize plants, the uptake of N can be adequately described by the proposed model using the NO_3^--N, NH_4^+-N, and DON concentrations measured in 0.01 *M* $CaCl_2$ extracts. Although in the fertilized soils the DON represents a smaller fraction of the N taken up by the plants, the model predicted the total N uptake by the maize plants rather well. The organic N mineralized during the growing period can be very well estimated using the initial DON concentration of the soil as measured in 0.01 *M* $CaCl_2$ extract.

ACKNOWLEDGMENTS

Special appreciation is expressed to V. J. G. Houba and I. Novozamsky for their assistance in this study. We thank E. Hoffland for valuable comments on the manuscript. Thanks to the Department of Soil Quality from the Wageningen University for the use of the greenhouses and the Central Laboratory, where the analytical determinations were performed. This work was developed with the financial support of a grant awarded in the context of the Netherlands Fellowships Programme of the Government of

The Netherlands, and partial support was obtained from the Science and Technology Secretary Austral Patagonia National University (SeCyT UNPA Argentina).

REFERENCES

Appel, T. and Mengel, K. (1990) Importance of organic nitrogen fractions in sandy soils, obtained by electro-ultrafiltration or $CaCl_2$ extraction, for nitrogen mineralization and nitrogen uptake of rape. *Biology and Fertility of Soils*, 10: 97–101.

Appel, T. and Mengel, K. (1992) Nitrogen uptake of cereals grown on sandy soils as related to nitrogen fertilizer application and soil nitrogen fractions obtained by electro-ultrafiltration (EUF) and $CaCl_2$ extraction. *European Journal of Agronomy*, 1: 1–9.

Black, C.A. (1968) Nitrogen. In *Soil–Plant Relationships*, 2nd ed.; Wiley: New York, 405–557.

Blackmer, A.M. (2000) Bioavailability of nitrogen. In *Handbook of Soil Science*. Sumner, M.E. (ed.); CRC Press: Boca Raton, Florida, D-3–D-17.

Brady, N.C. (1990) Nitrogen and sulfur economy of soils: Influence of nitrogen on plant development. In *The Nature and Properties of Soils*, 10th ed.; Maxwell Macmillan International Editions: New York, 315–349.

Bundy, L.G. and Meisinger, J.J. (1994) Nitrogen availability indices. In *Methods of Soil Analysis, Part 2: Microbiological and Biochemical Properties*; Page, A.L. (ed.); Soil Science Society of America: Madison, Wisconsin, 951–984.

Dendooven, L., Murphy, E., and Powlson, D.S. (2000) Failure to simulate C and N mineralization in soil using biomass C-to-N ratios as measured by the fumigation extraction method? *Soil Biology and Biochemistry 32 (5): 659–668.*

Hergert, G.W. (1987) Status of residual nitrate–nitrogen soil tests in the United States of America. In *Soil Testing: Sampling, Correlation, Calibration, and Interpretation*. Brown, J.R. (ed.); Special Publication 21; Soil Science Society of America: Madison, Wisconsin, 73–88.

Hood, R., Merckx, R., Jensen, E.S., Powlson, D., Matijevic, M., and Hardarson, G. (2000) Estimating crop N uptake from organic residues using a new approach to the N-15 isotope dilution technique. *Plant and Soil*, 223 (1–2): 33–44.

Houba, V.J.G., Jászberényi, I., and Loch, J. (1991) Application of 0.01 *M* $CaCl_2$ as a single extraction solution for evaluation of the nutritional status of Hungarian soils. *Debreceni Agrártudomáyi Egyetem Tumánnyos Közleményei. Tom.*, 30: 85–90.

Houba, V.J.G., Temminghoff, E.J.M., Gaikhorst, G.A., and van Vark, W. (2000) Soil analysis procedures using 0.01 *M* calcium chloride as extraction reagent. *Communications in Soil Science Plant Analysis*, 31 (9–10): 1299–1396.

Houba, V.J.G., van der Lee, J.J., and Novozamsky, I. (1997) *Soil Analysis Procedures, Other Procedures*. Soil and Plant Analysis, part 5B; Agricultural University: Wageningen, Netherlands.

Keeney, D.R. (1982) Nitrogen availability indices. In *Methods of Soil Analysis, part 2: Chemical Methods*, 2nd edn.; Page, A.L. (ed.); American Society of Agronomy: Madison, Wisconsin, 711–733.

Kempl, F., Steffens, D., Horn, D., and Schubert, S. (2001) Importance of EUF-extractable organic C for soil N mineralization and for N management of sugar beet. In *Plant Nutrition: Food Security and Sustainability of Agro-Ecosystems*;

Horst, W.J. et al. (eds); Kluwer Academic Publishers: Dordrecht, Netherlands, 750–751.

Lathwell, D.J., Dubey, H.D., and Fox, R.H. (1972) Nitrogen-supplying power of some tropical soils of Puerto Rico and methods for its evaluation. *Agronomy Journal*, 64: 763–766.

Maci, A. and Mengel, K. (2001) Net mineralization of organic N in soils related to EUF (electroultrafiltration) extractable organic N, organic C, nitrate and growth. In *Plant Nutrition: Food Security and Sustainability of Agro-Ecosystems*; Horst, W.J. et al. (eds); Kluwer Academic Publishers: Dordrecht, Netherlands, 748–749.

Murphy, D.V., Macdonald, A.J., Stockdale, E.A., Goulding, K.W.T., Fortune, S., Gaunt, J.L., Poulton, P.R., Wakefield, J.A., Webster, C.P., and Wilmer, W.S. (2000) Soluble organic nitrogen in agricultural soils. *Biology and Fertility of Soils*, 30: 374–387.

Németh, K., Makhdum, I.Q., Koch, K., and Beringer, H. (1979) Determination of categories of soil nitrogen by electro-ultrafiltration (EUF). *Plant and Soil*, 53: 445–453.

Nordin, A., Hogberg, P., and Nasholm, T. (2001) Soil nitrogen form and plant nitrogen uptake along a boreal forest productivity gradient. *Oecologia*, 129 (1): 125–132.

Novozamsky, I., Houba, V.J.G., van Eck, R., and van Vark, W. (1983) A novel digestion technique for multi-element plant analysis. *Communications in Soil Science and Plant Analysis*, 14: 239–249.

Nunan, N., Morgan, M.A., Brennan, D., and Herlihy, M. (2001) Organic matter extracted with 0.01 $CaCl_2$ or with 0.01 M $NaHCO_3$ as indices of N mineralisation and microbial biomass. *Biology and Fertility of Soils*, 34: 433–440.

Smolander, A., Kitunen, V., and Malkonen, E. (2001) Dissolved soil organic nitrogen and carbon in a Norway spruce stand and an adjacent clear-cut. *Biology and Fertility of Soils*, 33 (3): 190–196.

Sogbedji, J.M., Van Es, H.M., Yang, C.L., Geohring, L.D., and Magdoff, F.R. (2000) Nitrate leaching and nitrogen budget as affected by maize nitrogen rate and soil type. *Journal of Environmental Quality*, 29: 1813–1820.

Stanford, G. (1982) Assessment of soil nitrogen availability. In *Nitrogen in Agricultural Soils*; Stevenson, F.J. (ed.); Agronomy Monograph 22. American Society of Agronomy, Crop Science Society of America and Soil Science Society of America: Madison, Wisconsin, 651–688.

Stevenson, F.J. (1982) Origin and distribution of nitrogen in soils. In *Nitrogen in Agricultural Soils*. Stevenson, F.J. (ed.); American Society of Agronomy: Madison, Wisconsin, 1–42.

Stevenson, F.J. and Cole, M.A. (1999a) The nitrogen cycle in soil: Global and ecological aspects. In *Cycles of Soil*, 2nd ed.; Wiley: New York, 139–190.

Stevenson, F.J. and Cole, M.A. (1999b) The internal cycle of nitrogen in soil. In *Cycles of Soil*, 2nd ed.; Wiley: New York, 191–229.

Van Erp, P.J., Houba, V.J.G., and Van Beusichem, M.L. (1998) One hundredth molar calcium chloride extraction procedure, part I: A review of soil chemical, analytical, and plant nutritional aspects. *Communications in Soil Science and Plant Analysis*, 29: 1603–1623.

Van Erp, P.J., Houba, V.J.G., and Van Beusichem, M.L. (2001) Effect of drying temperature on amount of nutrient elements extracted with 0.01 M $CaCl_2$ soil extraction procedure. *Communications in Soil Science and Plant Analysis*, 32: 33–48.

Van Erp, P.J. (2002) The potentials of multi-nutrient soil extraction with 0.01 M $CaCl_2$ in nutrient management. PhD thesis, Wageningen University: Wageningen, Netherlands.

Verchot, L.V., Holmes, Z., Mulon, L., Groffman, P.M., and Lovett, G.M. (2001) Gross vs. net rates of N mineralization and nitrification as indicators of functional differences between forest types. *Soil Biology and Biochemistry*, 33 (14): 1889–1901.

Yu, Z.S., Northup, R.R., and Dahlgren, R.A. (1994) Determination of dissolved organic nitrogen using persulfate oxidation and conductimetric quantification of nitrate–nitrogen. *Communications in Soil Science and Plant Analysis*, 25 (19–20): 3161–3169.