

Dissolved organic carbon and dissolved nitrogen in soil under different fertilization treatments

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ABSTRACT

The objective of the study was to evaluate the effects of long-term fertilization of a sandy soil with differentiated doses of cattle slurry as well as its after-effect action on the possibilities of migration of dissolved organic carbon (DOC) and dissolved nitrogen (DNt) down to deeper layers of the soil profile. DOC and DNt were extracted with borate buffer and 0.004M CaCl₂ solution. Evaluation of effects of cattle slurry on the content of DOC and DNt was done in comparison with mineral fertilization. It was shown that the use of cattle slurry in the doses of 100 and 200 m³/ha caused a significant increase of labile organic matter in the 0–25 and 25–50 cm layers of soil. As compared with mineral fertilization the application of slurry increased also the amounts of extracted DNt, but only in the surface layer. The DNt content in the deeper soil horizons did not depend on the kind of fertilization. Concentrations of DOC and DNt in the extracts depended not only on their content in soil but it was also modified substantially by the extractant used.

Keywords: cattle slurry; dissolved organic carbon; dissolved nitrogen; washing out

Soil organic matter consists of organic material at different phases of decomposition and of macromolecular compounds of very specific properties. It is part soluble in water or in salt solutions of pH ~ 7 (dissolved organic matter) is the most mobile. It may contain carbohydrates, proteins, fats, hydrocarbons and their derivatives, fractions of humic acids of low molecular weight as well as other numerous simple organic compounds.

Quantification of soluble organic matter, i.e. fraction soluble in water or in salt solutions of reaction close to neutral is a complex process depending on a number of factors, connected first of all with the extraction procedure. The amount of soluble carbon assayed in extracts depends among others on the kind of extractant, time of extraction, sample moisture, the soil-extractant ratio as well as method of organic carbon assessing in the extracts (Zsolnay and Gorlitz 1994).

The labile organic matter fraction expressed as so called dissolved organic carbon (DOC) plays an important role in the soil environment: (1) it can be leached to deeper horizons of the soil profile and ground waters; an important process especially in sandy soils; (2) it can be transported in erosion

processes; (3) its components, especially low-molecular organic compounds and low-molecular humus fraction, may play the role of stimulators or inhibitors of the plant growth and plant development; (4) it can be an energetic material easily accessible by microorganisms.

The content of organic matter soluble in water in arable soils depends on agrotechnical factors, first of all on the kind of organic material introduced into soil (Reinersten et al. 1984, Hadas et al. 1993, McKenney et al. 1993, Černý et al. 2003). According to Asmar et al. (1994) the addition of fresh organic material to soil always increases the amount of water-soluble organic matter.

As a consequence of more and more popular non-bedding animal production manure as a fertilizer has been gradually replaced by slurry. Mineral and organic fertilization capacity of slurry is in stake for its agricultural application. That is why slurry is being compared both with manure and mineral fertilizers. In general fertilization with slurry increases yields of the cultivated plants, concentration of organic carbon and total nitrogen in soils (Einland 1981, Mackowiak 1994, Aoyama and Kumakura 2001). However, there

are a number of controversies connected first of all with the possibilities of migration of carbon mobile compounds released during mineralisation of organic material down the soil profile. Another important factor connected with the use of slurry are sometimes really big losses of nitrogen resulting from its leaching down the soil profile, even to ground waters (Mazur and Budzynska 1994, Periago et al. 2000).

The objective of this paper was to evaluate the effect of long-term fertilization of light soil with differentiated doses of cattle slurry and its sequential activity as well as possibilities of migration of labile carbon and nitrogen compounds down the soil profile. The evaluation of fertilization of cattle slurry on the content of dissolved organic carbon (DOC) and DNt in soil was done in comparison with mineral fertilization alone. The amount of DOC as well as that extracted with borate buffer of pH 7 and DNt were compared with labile forms of carbon and nitrogen obtained after extraction with 0.004M CaCl₂.

MATERIAL AND METHODS

The experiment was carried out on isolated (concrete barriers) plots of 1 m² filled in upper layer (1 m) with soil material of loamy sand grain size and clay at the bottom. In 1973–1989 cattle slurry was used at the calculation rates of 25 [G(25)], 50 [G(50)], 100 [G(100)] and 200 [G(200)] m³

per ha yearly – variants abbreviated as G1, or as a double rate every two years – variants abbreviated as G2. Every variant was repeated using slurry with extra nitrogen added as mineral fertilizers (NH₄NO₃, variants abbreviated as G1+N; G2+N). In order to make comparisons between the slurry and mineral fertilizers activity four parallel variants of mineral fertilisation were also included into the experiment (NPK1–NPK4) – Table 1. In this case the doses corresponded with 50% of the components being introduced into soil with cattle slurry. Average composition of the slurry was (in % of the fresh weight): N – 0.31%, P₂O₅ – 0.16%, K₂O – 0.41%, CaO – 0.23%, dry matter – 9.1%. There were 4 replications for every experimental variant.

The after effect was studied after stopping fertilisation with cattle slurry in 1990 and using corrected mineral fertilisation only, on all the plots up to 1999. An optional crop rotation system was used over the whole experimental period.

In 1973–1989 (17 years) the following plants were cultivated on the plots: potato, winter wheat, rye for green chop, corn for grains, oats, fodder beet, corn for grain, corn for silage, winter rape, sugar beet, spring barley, corn for grain, spring wheat, corn for grain, fodder cabbage, corn for grain, spring barley and potato. After the use of slurry, in 1990–1999 (10 years) the following plants were cultivated: sugar beet, corn for grain, oats, corn for grain (for two years), corn for silage (for two years), mustard for green chop, and corn for grain

Table 1. Experimental system

Treatment	Single dose of slurry (m ³ /ha)	Treatment	Single dose of slurry (m ³ /ha)
NPK1	–	NPK2	–
G1(25)	25	G1(50)	50
G1+N(25)	25	G1+N(50)	50
G2(25)	50	G2(50)	100
	each two years		each two years
G2+N(25)	50	G2+N(50)	100
	each two years		each two years
NPK3	–	NPK4	–
G1(100)	100	G1(200)	200
G1+N(100)	100	G1+N(200)	200
G2(100)	200	G2(200)	400
	each two years		each two years
G2+N(100)	200	G2+N(200)	400
	each two years		each two years

for the last two years (Maćkowiak 2000). After the harvest plant residues were left behind. All the cultivations measures were done by hand.

Soil samples were air dried and sieved ($\phi < 2$ mm). Organic matter content (TOC) in soil samples of the 0–25 cm, 25–50 cm, 50–75 cm and 75–100 cm layers was assayed with the Primacs^{SC} analyser (Skalar, Breda, the Netherlands), while total nitrogen content Nt was determined with the standard Kjeldahl method.

Dissolved organic carbon (DOC) and dissolved nitrogen (DNt) concentrations were measured in solutions obtained after extraction with borate buffer (pH = 7). The extraction was done using the soil sample to extractant ratio of 1:10 over 1 h. DOC and DNt content were assayed with the TOCN Formacs autoanalyser (Skalar, Breda, Holland).

The results were analysed using variation analysis with Tukey test at the significance level of $\alpha = 0.05$, for multi-factor experiments with ANALWAR software. The system of factors was as follows:

Factor I – year (sampling time: 1989, 1999)

Factor II – fertilizer dose (25, 50, 100, 200), for instance 25 indicates the variant of mineral fertilization NPK1 and all variants with the slurry used in the dose 25 m³/ha/year [G1(25), G1+N(25), G2(25) and G2+N(25)]

Factor III – kind of fertilization (NPK, G1, G1+N, G2, G2+N)

DOC and DNt contents determined by extraction with borate buffer were compared with their contents determined by extraction with 0.004M CaCl₂ solution. The dependencies among the fea-

tures under study were determined by correlation coefficients and regression equations with the use of STATISTICA software.

RESULTS AND DISCUSSION

Long-term fertilization with cattle slurry (1973–1989 – first step of the experiment) with doses ranging 50–200 m³/ha/year caused an increase of soil resources of organic matter. The introduction of slurry in the doses of 100 and 200 m³/ha/year produced a clear surplus of TOC existing even 10 years after the treatment, while fertilization with 50 m³ slurry per ha maintained humus content at the level of the starting data (7.9 g/kg). Experimental fertilization did not change carbon content in the soil layers deeper than 50 cm (Figure 1).

The application of slurry requires a close observation of changes in nitrogen concentration because of numerous opinions indicating that the major part of nitrogen introduced with the slurry which is not used by the plants undergoes leaching and/or is released to the atmosphere. Only a small portion of nitrogen remains in the soil (Mazur and Budzynska 1994). In our experiment fertilization with slurry increased by 27 to 108% total nitrogen content in the surface soil layer. Earlier fertilization with slurry in doses of 50–200 m³/ha/year resulted in the after effect observed as an increase of nitrogen concentration ranging 29–105% as compared with this element content in soil fertilized only with NPK. Nitrogen

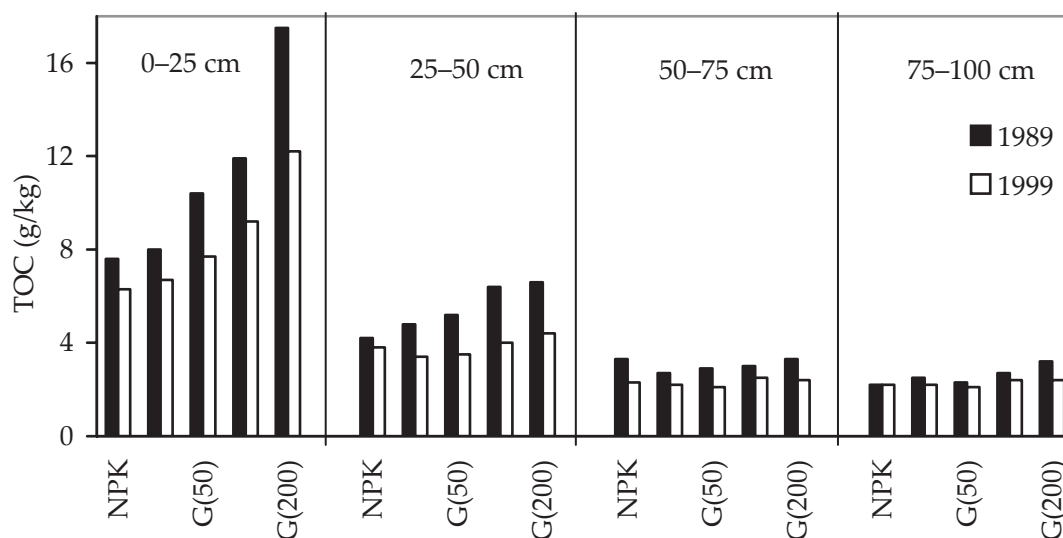


Figure 1. Average TOC content in soil fertilized only with minerals (NPK) and slurry doses of 25, 50, 100 and 200 m³/ha [G(25), G(50), G(100) and G(200), respectively]; 1989, 1999 – years of sampling

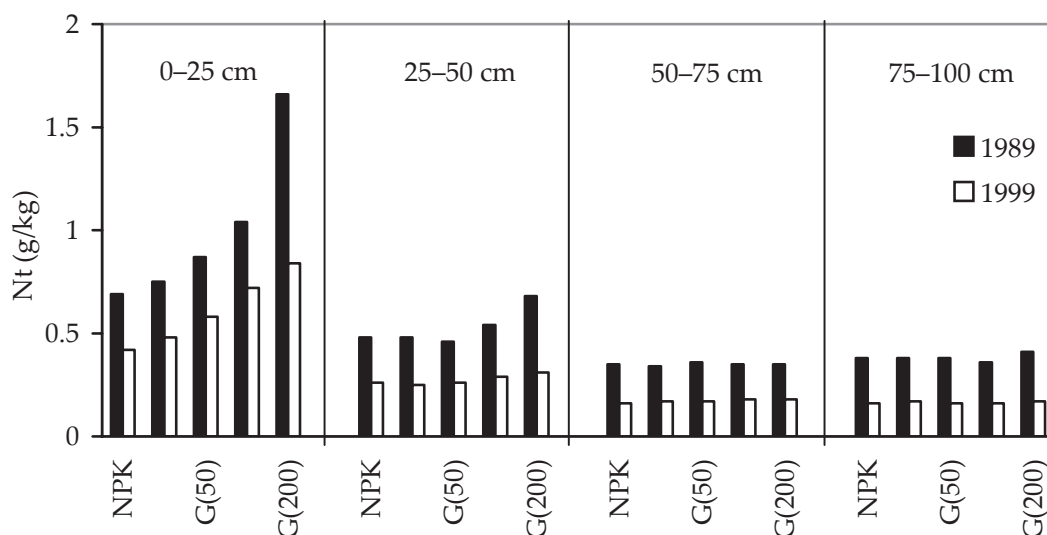


Figure 2. Average Nt content in soil fertilized only with minerals (NPK) and slurry doses of 25, 50, 100 and 200 m³/ha [G(25), G(50), G(100) and G(200), respectively]; 1989, 1999 – years of sampling

content in the 50–100 cm soil layers did not depend on the kind and dose of fertilizers (Figure 2).

Dissolved organic carbon (DOC)

The content of DOC extracted with borate buffer of pH = 7 in soil samples taken from the 0–25 and 25–50 cm soil layers significantly depended on the kind and amount of the fertilizer used. However, it was not related to the mode of slurry application (Table 2).

In 1989, fertilization of soil with slurry in the doses of 100 and 200 m³/ha significantly increased DOC content in the layers 0–25 and 25–50 cm as compared with fertilization with NPK (Table 3).

As compared with the data of 1989, surface soil sampled in 1999 from the plots fertilized earlier with cattle slurry and soil samples fertilized only with NPK contained less DOC (on average by 32% and 34%, respectively). The highest concentrations of DOC extracted with borate buffer were found in soil samples fertilized with slurry doses of 100 and 200 m³/ha. In 1999 the highest concentration of DOC in the 25–50 cm layer was shown in case of soil with earlier fertilization with slurry dose of 200 m³/ha. No effect of lower doses of slurry (less than 200 m³/ha) on dissolved organic matter was observed (Table 3).

Observations recorded for DOC content in soil layers lower than 50 cm were different. Statistical analysis did not show effects neither of fertilizers nor of the way of slurry application on the content of the most labile fraction of organic matter in

those horizons. Moreover, DOC concentration in soils taken from the 75–100 cm layer did not depend on the dose of fertiliser, too (Table 2). In the case of the 50–75 cm layer the highest contents of DOC were found in soil samples on which both slurry and mineral fertilizers were used in their highest doses. In 1999 DOC content in that soil layer decreased on average by about 26% as compared with its concentration noted in 1989 (Table 2).

Slurry application usually brings about threats of movements of labile carbon compounds toward deeper layers of the soil profile or even to ground waters. As compared with soil fertilized only with NPK the soil fertilized with 100 and 200 m³ of slurry per ha showed a significant increase of DOC but only in the layer 25–50 cm, while application of the highest dose of 200 m³/ha resulted in higher content of DOC in that part of soil even after 10 years from the completed use. The effect of enrichment of lower soil layers with extractable organic carbon did not depend on the kind of fertilization.

DOC content assessed in the surface soil in 1989 comprised on average 2.4% of TOC (Table 2). No effects of kind of fertilization and fertilizer dose on the percentage of this organic matter fraction in the 0–25 cm layer. A higher DOC share was found in soil organic matter in the 25–50 cm soil layer, while the percentage of TOC in this layer depended on the fertilizer dose reaching the highest values in the variants NPK4 and G(200). Over the experimental years the amount of DOC found in the 50–75 cm soil layer comprised on average

Table 2. Results of variance analysis of DOC content (mg/kg) in soil and DOC content as percent of TOC

Factor	Soil layer								
	0–25 cm		25–50 cm		50–75 cm		75–100 cm		
DOC (mg/kg)									
Factor I year	1989	261	<i>LSD</i> = 18.3	233	<i>LSD</i> = 11.9	172	<i>LSD</i> = 6.1	142	<i>LSD</i> = 7.7
	1999	175		140		127		110	
Factor II dose of fertilizers	25	169	<i>LSD</i> = 35.3	160	<i>LSD</i> = 21.6	131	<i>LSD</i> = 11.7	125	<i>LSD</i> = 14.9
	50	197		161		140		116	
	100	222		190		148		127	
	200	283		235		181		136	
Factor III mode of fertilization	NPK	167	<i>LSD</i> = 42.3	157	<i>LSD</i> = 25.9	141	ns	130	ns
	G1	232		197		148		133	
	G1+N	228		195		151		122	
	G2	229		193		155		125	
	G2+N	234		191		153		127	
Interactions									
I/II; II/I	36.6; 49.9			ns	13.6; 19.8			ns	
I/III; III/I	ns			ns	ns			17.3; 25.3	
II/III; III/II	ns			ns	ns			33.3; 35.7	
DOC content as percent of TOC									
Factor I year	1989	2.4	<i>LSD</i> = 0.16	4.3	<i>LSD</i> = 0.26	5.8	ns	5.7	<i>LSD</i> = 0.62
	1999	2.2		3.7		5.5		4.9	
Factor II dose of fertilizers	25	2.3	ns	4.0	<i>LSD</i> = 0.50	5.3	ns	5.4	ns
	50	2.3		3.8		5.5		5.4	
	100	2.3		3.8		5.3		5.3	
	200	2.2		4.5		6.4		5.4	
Factor III mode of fertilization	NPK	2.4	ns	3.9	ns	5.2	ns	5.7	ns
	G1	2.2		4.1		5.9		5.7	
	G1+N	2.1		4.0		5.6		5.0	
	G2	2.3		4.1		5.7		5.1	
	G2+N	2.3		3.9		6.0		5.1	

5.7% TOC. The percentage of DOC in the bulk of organic carbon determined in the 75–100 cm layer was similar to that observed for this carbon fraction in the 50–75 cm layer (Table 2).

Some controversies can arise because of more than doubled percentage of the DOC fraction in soil layers below 50 cm as compared with the share of the same fraction in the surface soil. The observation suggested a migration of dissolved organic matter occurring in the surface soil down the soil profile. However, a higher percentage of DOC in the

50–100 cm soil layer was not the result of the way of fertilization used and it did not depend of doses of fertilizers. Therefore, with some approximation it can be stated that organic matter of deeper soil layers is genetically richer in DOM fractions, what is a consequence of a high mobility of dissolved organic matter and/or a lower tendency of its mineralization. However, there is no doubt that because of fertilization and crop rotation certain enrichment in dissolved organic matter occurred in the 25–50 cm soil layer (Tables 2 and 3).

Table 3. Average DOC content (mg/kg) in soil layers (cm) after two study periods

Treatment	1989				1999			
	0–25	25–50	50–75	75–100	0–25	25–50	50–75	75–100
NPK1	180	181	143	121	145	115	115	105
NPK2	215	170	156	129	140	124	117	84
NPK3	206	185	150	131	112	128	128	91
NPK4	210	215	162	168	137	138	157	135
G(25)*	184	206	159	139	158	120	103	117
G(50)*	235	209	167	133	169	120	114	103
G(100)*	270	254	172	149	206	143	128	112
G(200)*	411	308	209	150	212	193	162	113

*average of four fertilization combinations (G1, G1+N, G2 and G2+N)

While analysing samples collected from long-term field experiments Debska (2004), Debska and Gonet (2002) showed that under fixed fertilization and crop rotation conditions the content of DOC in TOC is about the same depending directly on total organic matter concentration in soil. In our experiment the percentage of DOC in total organic carbon in surface soil ranged 2.2–2.4% and did not depend on any of the analysed factors. Therefore, the observed relations seem to corroborate with earlier findings.

Dissolved total nitrogen (DNt)

Nitrogen in slurry occurs in organic and mineral compounds. Among nitrogen organic ones one should mention proteins, aminoacids, urea, hippuric acid, ureic acid, etc. Inorganic nitrogen occurs mainly as ammonium (NH_4^+) and nitrate (V) (NO_3^-) nitrogen. About 50% of total nitrogen found in slurry is present in water-soluble forms, that is crucial for its high availability to plants as well as possibilities for penetration to lower soil layers and ground waters (Mackowiak 1994).

The content of total nitrogen extracted with borate buffer from the 0–25 cm layer depended on the kind and dose of the fertilizer used (Table 4). In 1989 after 17 years of fertilization with slurry, the highest DNt were found in soil fertilized with the dose 200 m³/ha. Also the highest NPK dose caused in soil an increase of total extractable nitrogen. After completing the second experimental step (1999) a higher DNt content were noted on plots fertilized with 100 and 200 m³ slurry/ha (Table 5).

Concentration of nitrogen in extracts obtained from samples taken from the 25–50 cm layer de-

pended exclusively on the dose of fertilized and was by about 79% higher in 1989 than in 1999 (Table 4). In 1989 the highest concentration of DNt was observed in soil samples with the highest doses of both slurry and NPK. After the second step of the experiment (1999) the concentration of DNt was still higher in soil after the treatment with high doses of the fertilizers.

Soil samples treated with the highest slurry dose collected in 1989 from the 50–75 and 75–100 cm layers contained the highest content of DNt (Table 5).

The percentage of DNt in the 0–25 cm layer sampled after the first step of the experiment (1989) was significantly higher as compared with corresponding samples collected after the second step in 1999. However, no univocal effect of fertilizer dose on the percentage of DNt fraction in the surface soil was noted. In the soil layers lower 25 cm the highest DNt percentage was observed after application of 200 m³ slurry/ha and the highest NPK dose.

The analysis of the DNt content expressed as absolute values and as percentage of the total nitrogen revealed that slurry doses strongly surpassing nutritional needs of plants (200 m³/ha, understood as a discarding dose) can cause an enrichment of deeper soil horizons with dissolvable nitrogen forms, similarly to overstated mineral fertilization.

The effect of extractant on the content of DOC and DNt

As can be found in the literature, the amount of extracted organic matter depends a great deal

Table 4. Results of variance analysis of DNt content (mg/kg) in soil and DNt content as percent of Nt

Factor	Soil layer								
	0–25 cm		25–50 cm		50–75 cm		75–100 cm		
DNt (mg/kg)									
Factor I year	1989	72	<i>LSD</i> = 4.7	43	<i>LSD</i> = 4.7	26	<i>LSD</i> = 3.0	30	<i>LSD</i> = 4.3
	1999	36		24		13		11	
Factor II dose of fertilizers	25	47	<i>LSD</i> = 9.0	29	<i>LSD</i> = 9.0	17	<i>LSD</i> = 5.8	16	<i>LSD</i> = 8.2
	50	42		27		16		15	
	100	53		33		18		20	
	200	73		46		27		29	
Factor III mode of fertilization	NPK	49		35		18		18	
	G1	53		36		22		20	
	G1+N	56	<i>LSD</i> = 10.9	35	ns	20	ns	21	ns
	G2	50		32		19		20	
	G2+N	61		31		20		23	
Interactions									
I/II; II/I	ns		9.3; 12.7		6.0; 8.2		8.5; 11.6		
I/III; III/I	10.5; 15.4		ns		ns		ns		
II/III; III/II	ns		ns		ns		ns		
DNt content as percent of Nt									
Factor I year	1989	7.8	<i>LSD</i> = 0.69	8.2	ns	7.6	ns	7.7	<i>LSD</i> = 1.01
	1999	5.9		8.7		7.6		6.6	
Factor II dose of fertilizers	25	7.7	<i>LSD</i> = 1.32	8.0	<i>LSD</i> = 1.60	6.6	<i>LSD</i> = 1.53	6.2	<i>LSD</i> = 1.94
	50	6.1		7.8		6.3		6.0	
	100	6.7		8.3		7.3		7.2	
	200	6.9		9.6		10.4		9.2	
Factor III mode of fertilization	NPK	8.6		9.4		7.5		6.6	
	G1	6.1		8.7		8.1		7.5	
	G1+N	6.2	<i>LSD</i> = 1.59	8.0	ns	7.4	ns	6.9	ns
	G2	6.3		8.1		7.7		7.1	
	G2+N	7.0		7.9		7.6		7.5	
Interactions									
I/II. II/I	1.37; 1.87		ns		ns		2.02; 2.75		
I/III. III/I	1.54; 2.25		ns		1.77; 2.59		ns		
II/III. III/II	ns		ns		ns		ns		

on the kind and concentration of the extractant used (David et al. 1989, Zsolnay and Gorlitz 1994). In order to limit extraction of clay fraction and to get close with ionic strength to the one of pore water solutions of salts of divalent cations (for instance CaSO_4 , NaNO_3 , KCl , CaCl_2) of low

concentration are applied for DOM extraction. The use of distilled water favors extraction of clay fraction, what in turn makes direct measurements (chromatography, spectrometry) of the solutions containing DOM difficult. Gonet et al. (2002) showed that after using borate buffer of pH = 7

Table 5. Average DNt content (mg/kg) in soil layers (cm) after two study periods

Treatment	1989				1999			
	0–25	25–50	50–75	75–100	0–25	25–50	50–75	75–100
NPK1	71	43	27	20	22	20	9,9	10
NPK2	69	40	18	14	27	22	12	8.7
NPK3	68	47	22	38	18	24	16	8.8
NPK4	85	60	23	33	32	24	16	13
G(25)*	64	36	23	23	30	21	11	11
G(50)*	56	32	22	22	25	20	11	11
G(100)*	69	38	23	29	42	26	13	9.6
G(200)*	97	64	41	44	56	30	17	13

*average of four fertilization combinations (G1, G1+N, G2 and G2+N)

solutions containing amounts of DOC similar to its content in water extracts could be obtained. Such extracts carry much lower amount of clay fraction. However, calcium chloride is the compound most often used in DOM extractions.

The content of DOM and DNt of soil samples fertilized with NPK and slurry in solutions obtained after extraction with 0.004 M CaCl₂ (Debska 2004) was on average by 57 and 22% lower than the content of these fractions in borate buffer extracts, respectively. The relationship between amounts of DOC and DNt assessed in borate buffer and their concentration in 0.004M CaCl₂ solutions is described by the equations below:

$$\text{DOC (CaCl}_2) = 0.548 \cdot \text{DOC (buffer)} - 1.96$$

$$r = 0.926$$

$$\text{DNt (CaCl}_2) = 0.856 \cdot \text{DNt (buffer)} - 0.194$$

$$r = 0.979$$

The amount of dissolved organic carbon extracted and the content of nitrogen in the extracts, independently from the extractant used, significantly correlated with concentration of organic carbon and Nt, respectively:

– for borate buffer:

$$\text{DOC (mg/kg)} = 14.7 \cdot \text{TOC (g/kg)} + 98.6$$

$$r = 0.798$$

$$\text{DNt (mg/kg)} = 62.5 \cdot \text{Nt (g/kg)} + 4.70$$

$$r = 0.880$$

– for 0.004M CaCl₂:

$$\text{DOC (mg/kg)} = 9.55 \cdot \text{TOC (g/kg)} + 27.0$$

$$r = 0.876$$

$$\text{DNt (mg/kg)} = 54.6 \cdot \text{Nt (g/kg)} + 1.58$$

$$r = 0.873$$

While assaying dissolved organic matter in soils it is necessary to consider the fact that its absolute amount depends on the kind (properties) of the

extractant. Our results also indicate a possibility for parallel assessing the content of dissolved organic carbon and nitrogen in the extracts. The DNt parameter appears to be an important index of the quality of soil organic matter, especially because of potential losses of nitrogen connected both with its volatilisation and leaching down the soil profile.

On the basis of experimental results it can be concluded:

Under the conditions of fertilization and crop rotation of the experiment a significant increase of the mobile organic matter fraction was observed after the use of 100 and 200 m³ slurry per ha as compared with its content in soil after application only of NPK. Soil treatment with 200 m³ slurry per ha resulted in an increased concentration of this carbon fraction maintained as long as after 10 years.

DOC concentration in soil layers lower than 50 cm did not depend on the kind of fertilizer (slurry, NPK).

A comparison of the results of application of slurry and NPK indicated an increase of DNt concentration in the surface soil. DNt content in the 25–100 cm soil layer depended only on the dose of the fertilizer used. Therefore it can be assumed that fertilization with slurry is a decisive factor for the migration of organic carbon compounds down the soil profile, while compared with mineral fertilization did not increase the leaching of nitrogen compounds.

The assessed amount of DOC and DNt in the extracts depends on the properties of the extractant used. Concentrations of DOC and DNt found after the extraction with of borate buffer of pH = 7 were higher as compared with extraction with 0.004M CaCl₂.

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