

EFFECT OF HERBICIDES ON THE COURSE OF AMMONIFICATION IN SOIL

**Jan Kucharski, Małgorzata Baćmaga,
Jadwiga Wyszowska**

**Chair of Microbiology
University of Warmia and Mazury in Olsztyn**

Abstract

A laboratory experiment has been performed to determine the effect of soil pollution with the herbicides: Harpun 500 SC, Faworyt 300 SL, Akord 180 OF and Mocarz 75 WG on the course of ammonification. The soil material for the experiment consisted of loamy sand of pH 6.5.

The experiment comprised five replications. Soil samples in particular objects were polluted with the herbicides at rates corresponding to the dose recommended by the manufacturer: 0 – control, 1 – a dose recommended by the producer, and the rates 50-, 100-, 150- and 200-fold higher than the recommended dose. Next, nitrogen was introduced to soil in the form of L-aspartic acid, DL-alanine, L-arginine and urea in the amounts of 0 and 300 mg N kg⁻¹ soil. Having been thoroughly mixed with the additional substances, the soil was brought to moisture equal 60% capillary water capacity and incubated for 12, 24, 36 and 48 hours at 25°C.

The study has demonstrated that the course of ammonification depended on the type and rate of a herbicide added to soil, type of an organic compound undergoing ammonification and duration of the trial. L-arginine was ammonified most rapidly, while ammonification of L-aspartic acid lasted the longest. Among the tested herbicides, the strongest inhibitory effect on ammonification process was produced by Mocarz 75 WG, which continued to exert negative influence on mineralisation of organic substances for 36 hours. The other preparations did not have such a considerable effect on the quantities of ammonified nitrogen.

Key words: soil pollution, herbicides, ammonification, amino acids.

WPLYW HERBICYDÓW NA PRZEBIEG PROCESU AMONIFIKACJI W GLEBIE

Abstrakt

W doświadczeniu laboratoryjnym określono wpływ zanieczyszczenia gleby herbicydami: Harpun 500 SC, Faworyt 300 SL, Akord 180 OF i Mocarz 75 WG na przebieg procesu amonifikacji. Materiałem glebowym użytym w badaniach był piasek gliniasty o pH 6,5.

Doświadczenie przeprowadzono w pięciu powtórzeniach. Próbkę glebową w odpowiednich obiektach zanieczyszczono herbicydami w dawkach wyrażonych jako wielokrotność dawki zalecanej przez producenta: 0 – kontrola, 1 – dawka zalecana przez producenta, dawki 50-, 100-, 150- i 200-krotnie większa od zalecanej przez producenta. Następnie do gleby wprowadzono azot w postaci kwasu L-asparaginowego, DL-alaniny, L-argininy i mocznika w ilości 0 i 300 mg N·kg⁻¹ gleby. Po dokładnym wymieszaniu glebę doprowadzono do wilgotności 60% kapilarnej pojemności wodnej i w takim stanie inkubowano próbki glebowe przez 12, 24, 36 i 48 h w temp. 25°C.

Wykazano, że przebieg procesu amonifikacji determinowany był rodzajem i dawką herbicydu, rodzajem amonifikowanego związku organicznego i czasem trwania doświadczenia. Najszybciej amonifikacji ulegała L-arginina, natomiast najwolniej kwas L-asparaginowy. Największy inhibicyjny wpływ na proces amonifikacji spośród wszystkich testowanych środków chwastobójczych wywierał Mocarz 75 WG. Jego negatywne oddziaływanie na mineralizację związków organicznych utrzymywało się przez 36 h. Pozostałe preparaty nie wpływały tak jednoznacznie na ilość zamonifikowanego azotu.

Słowa kluczowe: zanieczyszczenie gleby, herbicydy, amonifikacja, aminokwasy.

INTRODUCTION

In farming, herbicides are used to control weeds and improve the quality of agricultural produce. However, these preparations can also limit the uptake of nitrogen by crops and microorganisms, as well as modify mineralisation of organic compounds present in soil (KARA et al. 2004). Transformation of these compounds could serve as a principal index when evaluating changes triggered by various xenobiotics, including plant protection chemicals (KOSTOV, CLEEMPUT 2001, PIETRIL, BROOKES 2008). The process of mineralisation of organic nitrogen forms is highly sensitive to soil pollution with biocides, which can therefore partly or completely contribute to the inhibition of ammonification. The effect produced by these substances depends mainly on applied doses and biodegradability of a given chemical (MARTENS, BREMNER 1994).

Mineralisation of organic compounds is one of the basic processes which condition the amounts of nitrogen easily available to plants and soil microorganisms (BONDE et al. 2001, CASTALDI et al. 2009, KUCHARSKI et al. 2004, QIAN, CAI 2007, WYSZKOWSKA et al. 2006). The rate of this process is dependent on the soil reaction (pH), temperature, soil moisture, content of organic substances and soil biological activity (BOTTOMLEY et al. 2004, BRIERLEY et al. 2001, KRAVE et al. 2002, KUCHARSKI 1997, SIERRA 2006, ZHU, CARREIRO 1999).

Nitrogen occurring in soil in the organic form is broken down to ammonia or NH_4^+ ions (BARABASZ et al. 2002). The decomposition of organic forms of nitrogen such as proteins, amides and amino acids is run by various microorganisms (BARABASZ et al. 2002, KOSTOV, CLEEMPUT 2001, LINA, BROOKES 1999), which are all highly sensitive to effects produced by plant protection chemicals (KUCHARSKI, WYSZKOWSKA 2008). During mineralisation, organic compounds can be decomposed by bacteria and fungi (BARABASZ et al. 2002, LINA, BROOKES 1999).

The objective of the present study has been to determine the effect of soil pollution with the newest generation herbicides, such as Harpun 500 SL, Faworyt 300 SL, Akord 180 OF and Mocarz 75 WG, on the course of ammonification.

MATERIAL AND METHODS

A laboratory experiment was established on soil material collected from the arable-humus soil horizon. Under natural conditions, this was proper brown soil classified as loamy sand according to its granulometric composition. The soil properties are presented in Table 1.

For the purpose of the experiment, 100 cm³ beakers were filled with 50 g air dried soil mass, sieved through a 1 mm mesh sieve. All the experimental objects had five replicates. Soil samples were polluted with the herbicides Harpun 500 SL, Faworyt 300 SL, Akord 180 OF and Mocarz 75 WG (factor I) in rates corresponding to multiples of the dose recommended by the manufacturer: 0 – control, 1 – recommended dose, 50- 100-, 150- and 200-fold higher than the recommended dose (factor II).

The active substances in the herbicides were: isoproturon – a compound belonging to the class of urea derivatives (500 g dm⁻³) in Harpun 500 SC, chlpyralid – a compound in the form of monoethanolamine salt (300 g dm⁻³) in Faworyt 300 SL, fenmedifan, desmedifan – compounds belonging to phenyl carbaminians (each in the amount of 60 g dm⁻³) and ethofumesate – a compound belonging to benzofuran derivatives in Akord 180 OF and trito-sulfuron – a compound belonging to sulfonylurea derivatives (25%) and dicamba – a benzoic acid derivative (50%) in Mocarz 75 WG.

Once the herbicides had been applied, nitrogen was introduced to soil in the form of the following organic compounds (factor III): L-aspartic acid, DL-alanine, L-arginine and urea, in the amounts of 0 and 300 mg N kg⁻¹ soil (factor IV). Whole portions of soil were thoroughly mixed and brought to the soil moisture level corresponding to 60% capillary water capacity. Soil samples were incubated in a thermostat at temperature of 25°C for 12, 24, 36 and 48 hours (factor V).

The concentration of mineral nitrogen forms was determined using an Orion 720A ionometer. Aqueous solution of 0.03 mol acetic acid was used for extraction of N-NH_4^+ and N-NO_3^- . The extractor to soil ratio was 10 : 1. Levels of ions were measured using ion-selective electrodes. NH_4^+ ions were determined with an ammonium electrode (type 9300BNWP) whereas NO_3^- ions were determined with a nitrate electrode (type 9300BN). The intensity of ammonification was determined at appropriate hours according to the concentrations of N-NH_4^+ and N-NO_3^- ions from the formula (WYSZKOWSKA 2002):

$$N = \left(\frac{N_1 - N_0}{D} \right) \cdot 100\%$$

where:

N – % ammonified nitrogen;

N_1 – content of N-NH_4^+ and N-NO_3^- in the analysed object, in mg;

N_0 – content of N-NH_4^+ and N-NO_3^- in the control object, in mg;

D – rate of nitrogen N kg^{-1} soil.

The results were verified statistically using Duncan's multiple range test. All statistical computations were run with the software Statistica (Statsoft, Inc. 2006).

Table 1

Some physicochemical properties of soil used in the experiment

Soil type	Granulometric composition			pH _{KCl}	Hh	S	Corg (g kg ⁻¹)
	soil grains diameter (mm)				mmol(+) kg ⁻¹ soil		
	1 - 0.1	0.1 - 0.02	< 0.02				
ls	49	37	14	6.5	8.25	78.00	6.30

ls – loamy sand, Corg – organic carbon content, Hh – hydrolytic acidity,

S – sum of exchangeable base cations

RESULTS AND DISCUSSION

The rate of ammonification of L-aspartic acid, DL-alanine, L-arginine and urea in the control samples was varied (Tables 2-5). After 48 hrs of soil incubation, urea was nearly completely ammonified, compared to the successful ammonification of 87% L-arginine, 47% DL-alanine and 42% L-aspartic acid. The compounds which proved to be the most easily ammonified (L-arginine and urea) were ammonified at 26 and 25%, respectively, after 12 hours of incubation. Conversion of nitrogen organic compounds depended on the type of a herbicide and its dose, type of ammonified organic compound and duration of soil incubation (Tables 2-5).

Table 2

Effect of soil pollution with the herbicide Harpun 500 SC
on amount of ammonified nitrogen, in %

Rate of herbicide*	Time of incubation in hours			
	12	24	36	48
L-aspartic acid				
0	6.50 ± 0.17	20.37 ± 0.86	29.41 ± 2.38	41.71 ± 4.12
1	5.09 ± 1.45	22.42 ± 1.61	24.55 ± 0.77	47.33 ± 1.09
50	5.23 ± 1.67	34.47 ± 1.12	37.13 ± 0.99	51.30 ± 3.69
100	4.31 ± 1.16	32.99 ± 1.11	36.43 ± 0.69	37.30 ± 3.81
150	4.55 ± 1.01	23.17 ± 3.96	25.46 ± 1.66	30.31 ± 2.22
200	5.79 ± 0.99	18.24 ± 3.86	22.53 ± 1.34	24.69 ± 3.15
<i>r</i>	-0.27	-0.18	-0.32	-0.87
DL-alanine				
0	2.50 ± 0.36	8.10 ± 0.43	27.16 ± 2.18	47.18 ± 1.01
1	1.02 ± 0.74	10.37 ± 1.00	27.57 ± 0.59	56.46 ± 2.20
50	1.13 ± 0.43	10.73 ± 1.19	26.41 ± 2.05	63.37 ± 1.19
100	0.63 ± 0.41	13.46 ± 0.83	19.38 ± 2.72	65.92 ± 4.80
150	1.49 ± 0.67	12.07 ± 0.56	16.01 ± 1.44	70.78 ± 2.09
200	2.21 ± 0.53	11.29 ± 0.56	15.52 ± 1.15	65.41 ± 2.55
<i>r</i>	0.12	0.61	-0.96	0.79
L-arginine				
0	26.07 ± 1.69	36.16 ± 1.82	40.84 ± 2.48	87.18 ± 5.92
1	19.37 ± 1.16	42.85 ± 3.42	43.31 ± 2.61	99.93 ± 1.85
50	17.57 ± 2.38	46.52 ± 4.46	46.60 ± 1.23	95.17 ± 0.78
100	17.97 ± 2.08	46.13 ± 0.86	48.90 ± 0.73	79.05 ± 5.56
150	18.23 ± 1.29	46.55 ± 1.48	51.94 ± 2.12	78.45 ± 4.58
200	18.77 ± 0.49	42.47 ± 0.61	51.85 ± 1.70	80.61 ± 3.14
<i>r</i>	-0.51	0.42	0.95	-0.76
Urea				
0	24.83 ± 1.14	56.29 ± 1.12	73.91 ± 1.14	98.31 ± 2.34
1	22.45 ± 0.90	54.58 ± 2.84	72.58 ± 0.99	61.38 ± 0.99
50	22.05 ± 2.47	55.12 ± 3.57	72.60 ± 2.85	61.50 ± 0.85
100	13.43 ± 2.92	53.93 ± 3.57	71.30 ± 0.87	58.46 ± 1.73
150	12.99 ± 1.51	54.02 ± 2.49	73.07 ± 2.78	58.52 ± 0.75
200	12.93 ± 0.96	53.80 ± 0.72	67.65 ± 1.75	56.20 ± 0.61
<i>r</i>	-0.92	-0.77	-0.74	-0.91
LDS _{0.01**}	$a - 0.55, b - 0.50, c - 0.45, a \cdot b - 1.23, a \cdot c - 1.10, b \cdot c - 1.01,$ $a \cdot b \cdot c - 2.46$			

** LSD for: *a* – rate of herbicide, *b* – date of analysis, *c* – type of organic substance;
r – correlation coefficient; *0 – control not polluted with herbicides, 1 – dose recommended
by manufacturer; rates higher than recommended: 50-, 100-, 150- and 200-fold

Table 3

Effect of soil pollution with the herbicide Faworyt 300 SL
on amount of ammonified nitrogen, in %

Rate of herbicide*	Time of incubation in hours			
	12	24	36	48
L-aspartic acid				
0	6.50 ± 0.17	20.37 ± 0.86	29.41 ± 2.38	41.71 ± 4.12
1	6.11 ± 1.14	20.26 ± 0.08	30.58 ± 0.90	49.09 ± 1.00
50	4.45 ± 0.55	19.67 ± 0.29	41.12 ± 0.41	51.16 ± 1.89
100	4.84 ± 1.74	18.56 ± 0.37	44.80 ± 0.43	59.58 ± 0.85
150	4.04 ± 0.57	18.34 ± 0.43	51.66 ± 0.77	79.35 ± 2.18
200	4.02 ± 0.79	14.17 ± 0.17	63.57 ± 0.87	90.88 ± 1.06
<i>r</i>	-0.87	-0.91	0.99	0.97
DL-alanine				
0	2.50 ± 0.36	8.10 ± 0.43	27.16 ± 2.18	47.18 ± 1.01
1	5.53 ± 2.18	13.23 ± 1.43	23.52 ± 3.16	74.15 ± 1.49
50	6.72 ± 1.18	13.74 ± 1.26	22.25 ± 2.39	75.37 ± 2.30
100	9.15 ± 1.24	15.78 ± 2.59	21.04 ± 3.23	86.68 ± 4.03
150	9.42 ± 0.49	17.74 ± 2.86	16.37 ± 1.81	92.45 ± 4.63
200	10.66 ± 2.04	14.84 ± 1.71	16.02 ± 2.50	93.48 ± 2.92
<i>r</i>	0.92	0.71	- 0.94	0.84
L-arginine				
0	26.07 ± 1.69	36.16 ± 1.82	40.84 ± 2.48	87.18 ± 5.92
1	23.05 ± 0.85	37.42 ± 4.07	44.33 ± 4.14	88.84 ± 2.73
50	22.55 ± 1.01	46.49 ± 6.53	42.09 ± 1.10	87.12 ± 1.15
100	21.30 ± 1.86	52.89 ± 1.10	47.09 ± 1.87	94.90 ± 5.10
150	21.41 ± 0.88	53.52 ± 7.35	49.62 ± 2.45	89.77 ± 0.89
200	16.91 ± 0.35	54.34 ± 6.46	50.88 ± 3.93	85.49 ± 2.68
<i>r</i>	-0.89	0.92	0.92	-0.02
Urea				
0	24.83 ± 1.14	56.29 ± 1.12	73.91 ± 1.14	98.31 ± 2.34
1	29.05 ± 2.39	73.12 ± 3.52	75.32 ± 0.88	93.15 ± 1.43
50	30.91 ± 2.21	72.84 ± 3.42	71.46 ± 0.91	97.94 ± 2.37
100	20.90 ± 5.29	46.92 ± 2.23	68.02 ± 2.78	99.19 ± 2.17
150	16.29 ± 1.17	42.58 ± 1.56	67.89 ± 1.83	88.43 ± 3.23
200	15.66 ± 0.58	40.40 ± 1.22	62.36 ± 1.65	81.28 ± 2.71
<i>r</i>	- 0.86	-0.81	-0.97	-0.76
LDS _{0.01**}	$a - 0.62, b - 0.57, c - 0.51, a \cdot b - 1.40, a \cdot c - 1.25, b \cdot c - 1.14,$ $a \cdot b \cdot c - 2.79$			

* for designations cf. Table 2

Table 4

Effect of soil pollution with the herbicide Akord 180 OF
on amount of ammonified nitrogen, in %

Rate of herbicide*	Time of incubation in hours			
	12	24	36	48
L-aspartic acid				
0	6.50 ± 0.17	20.37 ± 0.86	29.41 ± 2.38	41.71 ± 4.12
1	5.59 ± 0.44	21.60 ± 0.18	31.31 ± 3.55	41.14 ± 4.31
50	5.09 ± 0.43	21.14 ± 0.26	35.19 ± 4.28	43.66 ± 2.27
100	5.72 ± 0.42	21.04 ± 0.40	44.53 ± 2.14	44.77 ± 2.34
150	4.74 ± 0.72	22.35 ± 0.34	42.55 ± 2.18	44.81 ± 2.16
200	4.56 ± 0.25	22.24 ± 0.46	42.35 ± 3.35	44.87 ± 2.76
<i>r</i>	-0.80	0.75	0.87	0.89
DL-alanine				
0	2.50 ± 0.36	8.10 ± 0.43	27.16 ± 2.18	47.18 ± 1.01
1	2.37 ± 0.81	7.97 ± 0.84	26.43 ± 1.87	56.22 ± 3.36
50	2.48 ± 0.81	7.22 ± 0.27	26.12 ± 1.84	56.83 ± 1.74
100	1.04 ± 1.23	6.96 ± 0.39	24.50 ± 1.20	51.88 ± 3.19
150	1.23 ± 0.76	6.65 ± 0.24	23.91 ± 1.70	51.36 ± 3.71
200	1.35 ± 0.61	5.21 ± 0.38	22.09 ± 1.79	50.69 ± 1.63
<i>r</i>	-0.82	-0.96	-0.98	-0.21
L-arginine				
0	26.07 ± 1.69	36.16 ± 1.82	40.84 ± 2.48	87.18 ± 5.92
1	26.79 ± 3.21	41.53 ± 1.75	34.22 ± 2.81	95.32 ± 2.34
50	26.37 ± 1.67	43.36 ± 1.84	33.24 ± 6.00	97.17 ± 2.64
100	25.95 ± 1.51	46.79 ± 0.95	27.70 ± 2.50	97.60 ± 2.83
150	26.54 ± 0.51	43.89 ± 3.04	26.95 ± 4.03	98.27 ± 2.83
200	29.93 ± 0.81	39.66 ± 1.68	23.29 ± 2.68	99.79 ± 3.45
<i>r</i>	0.67	0.27	-0.93	0.75
Urea				
0	24.83 ± 1.14	56.29 ± 1.12	73.91 ± 1.14	98.31 ± 2.34
1	16.30 ± 1.06	60.38 ± 1.13	60.55 ± 3.72	95.70 ± 1.98
50	12.96 ± 0.67	59.95 ± 1.04	56.37 ± 2.23	93.46 ± 1.83
100	13.21 ± 0.70	61.52 ± 0.47	54.28 ± 3.86	91.30 ± 0.84
150	11.47 ± 0.92	80.04 ± 0.68	45.75 ± 2.65	97.09 ± 1.96
200	11.60 ± 0.57	79.96 ± 1.12	42.56 ± 2.87	97.48 ± 0.47
<i>r</i>	-0.73	0.91	-0.91	0.08
LDS _{0.01**}	$a - n.s., b - 0.50, c - 0.45, a \cdot b - 1.23, a \cdot c - 1.10, b \cdot c - 1.00,$ $a \cdot b \cdot c - 2.46$			

* for designations cf. Table 2

Table 5

Effect of soil pollution with the herbicide Mocarz 75 WG
on amount of ammonified nitrogen, in %

Rate of herbicide*	Time of incubation in hours			
	12	24	36	48
L-aspartic acid				
0	6.50 ± 0.17	20.37 ± 0.86	29.41 ± 2.38	41.71 ± 4.12
1	5.69 ± 0.32	15.82 ± 0.58	29.19 ± 0.79	28.53 ± 0.27
50	4.27 ± 0.56	12.98 ± 0.21	25.81 ± 0.98	22.82 ± 0.20
100	3.38 ± 0.41	12.25 ± 0.33	22.80 ± 1.44	17.59 ± 0.30
150	2.68 ± 0.58	11.80 ± 0.26	21.83 ± 0.50	15.84 ± 0.63
200	3.08 ± 0.25	10.02 ± 0.27	21.32 ± 0.97	13.64 ± 0.19
<i>r</i>	-0.90	-0.85	-0.96	-0.86
DL-alanine				
0	2.50 ± 0.36	8.10 ± 0.43	27.16 ± 2.18	47.18 ± 1.01
1	2.99 ± 0.43	9.12 ± 0.38	23.71 ± 1.65	43.70 ± 1.83
50	2.99 ± 0.34	6.26 ± 0.57	21.23 ± 2.91	35.74 ± 2.33
100	2.84 ± 0.47	5.90 ± 0.47	21.23 ± 3.40	34.13 ± 0.87
150	2.63 ± 0.26	4.53 ± 0.42	17.09 ± 2.54	41.27 ± 1.38
200	2.76 ± 0.56	2.96 ± 0.25	15.74 ± 2.18	44.74 ± 0.43
<i>r</i>	-0.37	-0.97	-0.94	-0.10
L-arginine				
0	26.07 ± 1.69	36.16 ± 1.82	40.84 ± 2.48	87.18 ± 5.92
1	39.98 ± 1.12	35.48 ± 2.79	33.47 ± 1.34	86.00 ± 2.27
50	46.53 ± 1.22	19.95 ± 1.64	30.21 ± 2.67	86.09 ± 1.96
100	49.00 ± 1.61	16.71 ± 1.08	26.54 ± 1.99	85.62 ± 1.85
150	51.02 ± 1.09	12.48 ± 1.01	23.46 ± 1.60	82.52 ± 3.32
200	55.57 ± 0.29	9.44 ± 0.47	19.52 ± 0.84	80.59 ± 2.08
<i>r</i>	0.85	-0.94	-0.94	-0.93
Urea				
0	24.83 ± 1.14	56.29 ± 1.12	73.91 ± 1.14	98.31 ± 2.34
1	21.83 ± 1.41	48.39 ± 5.61	69.86 ± 1.61	98.42 ± 3.05
50	22.79 ± 1.72	45.58 ± 1.96	68.61 ± 1.12	98.73 ± 2.32
100	22.49 ± 1.77	42.26 ± 1.33	67.61 ± 3.10	96.57 ± 2.15
150	20.50 ± 1.36	42.77 ± 2.30	57.15 ± 2.15	95.57 ± 2.33
200	18.05 ± 1.44	37.41 ± 2.11	53.87 ± 2.18	97.43 ± 0.68
<i>r</i>	-0.85	-0.88	-0.95	-0.70
LDS _{0.01**}	$a - 0.45, b - 0.41, c - 0.37, a \cdot b - 1.00, a \cdot c - 0.90, b \cdot c - 0.82,$ $a \cdot b \cdot c - 2.02$			

* for designations cf. Table 2

The course of ammonification process was modified by the herbicide Harpun 500 SC (Table 2). It had a negative effect on the transformation of urea. For 36 hrs it intensified transformations of L-arginine and, when applied at the lowest doses (technological and 50-fold higher), it continued to do so for 48 hrs. However, it had a variable influence on the ammonification of L-alanine. After 24 hrs of incubation, the soil containing this herbicide and DL-alanine was determined to possess more ammonia nitrogen than after 36 hrs, although at the end of the incubation (48 hrs) the ammonification of this compound accelerated again. Excessive quantities of Harpun 500 SC (applied at the rates 100- and 200-fold higher than recommended) depressed the rate of the ammonification of L-aspartic acid.

The rate of the ammonification process as shaped by Faworyt 300 SL varied between the dates of analyses (Table 3). The transformations of L-aspartic acid were initially inactivated by this herbicide, although after 36 and 48 hrs of incubation, Harpun was found to be stimulating the ammonification of this compound. The positive influence of this herbicide on decomposition of DL-alanine at 12 and 24 hrs disappeared at 36 h to reappear at 48 h. Faworyt 300 SL retarded the ammonification of L-arginine at 12 h, but accelerated it at 24 h and 36 h. When applied at the highest doses (over 100-fold higher than the recommended dose), this herbicide halted hydrolysis of urea for 36 hours, and when introduced to soil at the doses 150- and 200-fold higher than recommended, it had a negative effect on this process throughout the whole incubation period (48 h).

Another tested herbicide, Akrod 180 OF, had a weaker effect on ammonification (Table 4). It only had an inhibitory influence on the transformation of urea in soil and stimulated transformations of L-aspartic acid. When applied at the technological rate and 50-fold higher than recommended, it intensified decomposition of DL-alanine, while producing only a light effect on the ammonification of L-arginine. The rate of ammonification was changeable during the incubation. At 12 h it retarded the transformation of L-aspartic acid, DL-alanine and urea, but already at 24 h it stimulated the decomposition of all the organic compounds except DL-alanine. It continued to exert the negative influence on DL-alanine until 36 h of incubation. At that time it also negatively affected the transformations of L-arginine and urea, but had a positive effect on the ammonification of L-aspartic acid. After 48 hours, it reduced only the hydrolysis of urea, while stimulating the transformation of all the remaining organic compounds.

The herbicide Mocarz 75 WG also had a significant effect on ammonification (Table 5). In the soil containing the highest rate of this chemical, it was found out that ammonification was depressed by 51% for L-aspartic acid, 22% for DL-alanine, 18% for urea and 13% for L-arginine. This preparation inhibited transformations of all the tested organic compounds for 36 hours. After 48 hours, it ceased to have a negative influence on the ammonification of urea and had a much weaker adverse effect on decomposition of L-arginine. However, it continued to significantly inhibit the ammonification of L-aspartic acid and DL-alanine.

Owing to ammonification, nitrogen present in organic compounds is transformed to forms easily available to plants. Amino acids, which are a valuable source of organic nitrogen, can enhance the intensification of nitrogen ammonification (WYSZKOWSKA 2002). The literature (JONES et al. 2005, WYSZKOWSKA et al. 2006) suggests that mineralisation of amino acids in soil can occur very rapidly and be completed in the course of just a few hours. The authors' own studies have shown that introduction of herbicides to soil can retard the rate of ammonification. Nonetheless, urea and L-arginine were the two organic substances whose ammonification was the fastest.

Chemical pollutants entering soil destabilize ammonification, which has microbiological and biochemical consequences for soil environment (KUCHARSKI, WYSZKOWSKA 2008, WYSZKOWSKA et al. 2007).

The rate of ammonification is determined by several factors, but particularly by a type of organic compound, including amino acids. L-arginine, which belongs to alkaline amino acids, was most rapidly ammonified, whereas L-aspartic acid, which is an acidic amino acid, underwent ammonification most slowly. The chemical properties of L-aspartic acid may have led to acidification of soil and lower soil pH, which resulted in a slower rate of its mineralisation (MARTENS, BREMNER 1994, WYSZKOWSKA et al. 2006, WYSZKOWSKA ET al. 2007). Changing the conditions within soil environment, such as depressed soil reaction, is not indifferent to soil microorganisms, especially to ammonifying bacteria (BARABASZ et al. 2002).

CONCLUSIONS

1. All the tested herbicides had a significant effect on the course of ammonification.
2. The rate of ammonification depended on the type and dose of a herbicide, type of ammonified organic compound and time of incubation.
3. Among the tested organic compounds, i.e. L-aspartic acid, DL-alanine, L-arginine and urea, ammonification of urea and L-arginine was the fastest.
4. The strongest inhibitor of ammonification proved to be the herbicide Mocarz 75 WG, which for 36 hours retarded the transformation of all the tested organic nitrogen compounds, while the other preparations did not have such an unambiguous influence on ammonification.

REFERENCES

- BARABASZ W, ALBIŃSKA D., JASKOWSKA M., LIPIEC J. 2002. *Biological effects of mineral nitrogen fertilization on soil microorganisms*. Pol. J. Environ. Stud., 11(3): 193- 198.
- BONDE T. A., NIELSEN T. H., MILLER M., SORENSEN J. 2001. *Arginine ammonification assay as a rapid index of gross N mineralization in agricultural soils*. Biol. Fertil. Soils, 34: 179-184.

- BOTTOMLEY P.J., TAYLOR A.E., BOYLE S.A., MCMAHON S.K., RICH J.J., CROMACK JR. K., MYROLD D.D. 2004. *Responses of nitrification and ammonia-oxidizing bacteria to reciprocal transfers of soil between adjacent coniferous forest and meadow vegetation in the Cascade Mountains of Oregon*. Microb. Ecol., 48: 500-508.
- BRIERLEY E.D., WOOD M., SHAW P.J.A. 2001. *Influence of tree species and ground vegetation on nitrification in an acid forest soil*. Plant Soil, 229: 97-104.
- CASTALDI S., CARFORA A., FIORENTINO A., NATALE A., MESSERE A., MIGLIETTA F., COTRUFO M. F. 2009. *Inhibition of net nitrification activity in a Mediterranean woodland: possible role chemicals produced by Arbutus unedo*. Plant Soil, 315: 273-283.
- JONES D.L., KEMMITT S.J., WRIGHT D., CUTTLE S.P., BOL R., EDWARDS A.C. 2005. *Rapid intrinsic rates of amino acid biodegradation in soils are unaffected by agricultural management strategy*. Soil Biol. Biochem., 37: 1267-1275.
- KARA E.E., ARIL M., UYGUR V. 2004. *Effects of the herbicide topogard on soil respiration, nitrification and denitrification in potato-cultivated soils differing in pH hort communication*. Biol. Fertil. Soils, 39: 474-478.
- KOSTOV O., CLEEMPUT O. 2001. *Nitrogen transformation in copper-contaminated soils and effects of lime and compost application on soil resiliency*. Biol. Fertil. Soils., 33: 10-16.
- KRAVE A. S., STRAALEN N. M., VERSEVELD H. W. 2002. *Potential nitrification and factors influencing nitrification in pine forest and agricultural soils in Centarl Java, Indonesia*. Pedobiologia, 46: 573-594.
- KUCHARSKI J. 1997. *Relation between enzymes activity and soil fertility. Microbes in environment – appearance, activity and importance*. (Ed. W. BARABASZ). AR Kraków, 327-347.
- KUCHARSKI J., JASTRZĘBSKA E., WYSZKOWSKA J. 2004. *Effects of some oil products on course of ammonification and nitrification*. Acta Agr. Silv. ser. Agr., 42: 249-255.
- KUCHARSKI J., WYSZKOWSKA J. 2008. *Biological properties of soil contaminated with the herbicide Apyros 75 WG*. J. Elementol., 13(3): 357-371.
- LINA Q., BROOKES P.C. 1999. *Arginine ammonification as method to estimate soil microbial biomass and microbial community structure*. Soil Biol. Biochem., 31: 1985-1997.
- MARTENS D.A., BREMNER J.M. 1994. *Effects of preemergence and postemergence herbicides on urna hydrolysis and nitrification of urea nitrogen in soil*. Biol. Fertil. Soils, 17, 309-313.
- PIETRIL J.C.A., BROOKES P.C. 2008. *Nitrogen mineralisation along a pH gradient of a silty loam UK soil*. Soil Biol. Biochem., 40: 797-802.
- QIAN C., CAI Z. 2007. *Leaching of nitrogen from subtropical soils as affected by nitrification potential and base cations*. Plant Soil, 300: 197-205.
- SIERRA J. 2006. *A hot-spot approach applied to nitrification in tropical acid soils*. Soil Biol. Biochem., 38: 644-652.
- Statsoft. Inc. 2006. Statistica (data analysis software), version 6.0. www.statsoft.com.
- WYSZKOWSKA J. 2002. *Biological properties of soil contaminated with hexavalent chromium*. Wyd. UWM., Rozpr. i Monogr., 65: 1-134.
- WYSZKOWSKA J., BOROS E., KUCHARSKI J. 2007. *The effect of nickel on the ammonification process in soil*. Pol. J. Natur. Sc., 22(3): 383-394.
- WYSZKOWSKA J., KUCHARSKI M., KUCHARSKI J., BOROWIK A. 2006. *Transformations of aspartic acid and l-glutamic acid in the presence of nitrification inhibitors*. Acta Agr. Silv. ser. Agr., 49: 507-515.
- ZHU W., CARREIRO M. M. 1999. *Chemoautotrophic nitrification in forest soils along an urban-to-rural transect*. Soil Biol. Biochem., 31: 1091-1100.

