Microbiological characterization of land set-aside before and after Roundup desiccation

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ABSTRACT

Luvic chernozem (set-aside from 1996) was evaluated. The first period, before Roundup desiccation (2002–2003), was characterized by black, spontaneous and controlled fallows (mowed with the removal of plant biomass or mulched); the following period (2003–2004) by black fallow with repeated Roundup applications; and the last period (2004–2006) involved re-grassing by a mowed *Lolium multiflorum* Lam. monoculture. The characterization included microbial biomass, available organic carbon, basal respiration, metabolic quotient, biomass-specific available organic carbon, arylsulfatase activity, soil organic matter carbon and total nitrogen. Mulching of pure cultures of grasses and legumes contributed to a high soil organic matter accumulation. Repeated Roundup desiccation caused a strong (highly significant) decrease of arylsulfatase activity (–28%), however highly significant increase of microbial biomass (+69%) and nitrate-nitrogen (+86%) were determined. The subsequent re-grassing compensated the changes described. The soil biological properties were best preserved on mulched fallow with *Lotus corniculatus* L. and *Festuca pratensis* L., also in regard to contamination with weeds.

Keywords: fallow; microbial biomass; metabolic quotient; respiration; arylsulfatase

Desiccation can play a very important role in the changes of soil biological properties. Reinecke et al. (2002) studied the impact of different herbicides on biological activity in several vineyard soils, with and without cover crops. A comparison of microbial activity after treatment with paraquat, simazine, glyphosate and a glyphosate-terbutylazine mixture showed (with the exception of simazine) that herbicide application had a positive influence on it. Any possible negative effects of applied herbicides (Hiller et al. 2009) are mainly due to chemical and textural parameters of treated soil. Roundup desiccation had a retardation effect on mineralization of C_{org}, N_{org} and S_{org} (Nováková and Voříšek 2006). Total soil organic carbon content and its qualitative characteristics, in particular, are the most important factors affecting sorption-desorption of herbicides in soil (Hiller et al. 2009). Chernozems of Central Europe have been used for cereals production for 7000 years without substantial decreases in fertility (Kunzova and Hejcman 2009). The quoted paper confirms extraordinary stability and certainty in achieved yields of cereals in Chernozems. Therefore their 'settling to rest' (set-aside) occurs less frequently than in less stable soils.

The article responds to the question how to maintain or improve the biological properties of luvic chernozem, when was transformed into a land set-aside for many years. Different variations of vegetation cover treating, mowing or mulching, followed by all-over black (= Roundup) fallow and re-grassing by a mowed Italian ryegrass monoculture (*Lolium multiflorum* Lam.), were evaluated. A variant is recommended, in which the biological properties of luvic chernozem were preserved at the best.

Supported by the Ministry of Education, Youth and Sports of the Czech Republic, Project No. MSM 6046070901.

Soils set-aside can be mulched, cut, desiccated, sown with pure cultures, sown with mixtures, maintained as spontaneous or black barren. The aim of the article is to describe the changes in the biological parameters under different soil management. Using chemical and biological parameters, the article compares which of the ways mentioned are more and which are less suitable.

The submitted publication presents a continuation of findings published in Růžková et al. (2008) and provides new conclusions.

MATERIAL AND METHODS

We participated in the experiment established by the Department of Forage Crops and Grass Management in the year 1996 for the study of land set-aside. The experimental field is located in the western part of Prague, on the campus of the Czech University of Life Sciences. Altitude at the site is 281 m, longitude 14°22'E, latitude 50°08'N. The average annual precipitation is 431 mm and the mean annual temperature is 9.3°C (Meteorological Station Prague-Karlov, measurements of 1961–2000). The soil is loamy luvic chernozem developed on carbonate loess with a 200 mm thick layer of arable top-soil. The proportion of sand (ISO 11277) is 20.5 ± 2.8%, the proportion of clay 26.8 ± 2.2% and the proportion of silt 52.7 ± 4.2% in the arable layer. The initial soil chemical properties before the start of the experiment were: A400/A600 4.59, C_{org} 1.45%, total Kjeldahl nitrogen 0.19%, pH (H_2O) 7.55.

The experimental field (0.22 ha) was formerly used in arable system until 1995.

(1) In 1996 it was changed into a land set-aside and divided into 45 experimental plots (15 m × 3 m). In rows of 125 mm, without cover crops, there were sown a pure culture of legumes, a pure culture of grasses, and mixtures of grasses and legumes:

Four legumes: white clover *Trifolium repens* L. cv Hájek (12 kg/ha), black medick *Medicago lupulina* L. cv. Ekola (20 kg/ha), birdsfoot trefoil *Lotus corniculatus* L. cv. Lotar (15 kg/ha) and alfalfa *Medicago media* Pers. cv. Mediana (18 kg/ha)

Four grasses: mountain brome *Bromus marginatus* Nees ex Steud. cv Tacit (50 kg/ha), false oat-grass *Arrhenatherum elatius* (L.) P.Beauv. ex J.S. et K.B. Presl cv Median (50 kg/ha), meadow fescue Festuca pratensis L. cv. Otava (30 kg/ha) and orchard grass *Dactylis polygama* Horv. (syn. *D. aschersoniana* Graben.) cv. Tosca (18 kg/ha)

Four mixtures: crownvetch *Coronilla varia* L. cv. Eroza + mountain brome *Bromus marginatus* Nees ex Steud. cv. Tacit (10 + 20 kg/ha), birdsfoot trefoil *Lotus corniculatus* L. cv. Lotar + meadow fescue *Festuca pratensis* L. cv. Otava (6 + 20 kg/ha), white clover *Trifolium repens* L. cv. Hájek + orchard grass *Dactylis polygama* Horv. cv. Tosca (6 + 15 kg/ha), black medick *Medicago lupulina* L. cv. Ekola + false oat-grass *Arrhenatherum elatius* (L.) P.Beauv. ex J.S. et K.B.Presl cv. Median (10 + 20 kg/ha)

The scheme of variants (4 variants of pure cultures of grasses in 3 replication; 4 variants of pure cultures of legumes in 3 replications; 4 variants of their mixtures in 3 replications; black fallow in 3 replication; spontaneous fallow in 6 replications) is presented in the Table 1.

(2) The whole experimental field was repeatedly desiccated by the glyphosate herbicide Roundup Biaktiv (5 l/ha):

In July 2003 – seven-year old herbaceous cover desiccation;

In June 2004 – weeds desiccation;

In August 2004 – pre-sowing application.

(3) The entire area was sown with Italian ryegrass *Lolium multiflorum* L. cv. Lolita (60 kg/ha) in September 2004.

Soil sampling included: period before Roundup desiccation (2002–2003) was characterized as

Table 1. Scheme of controlled, spontaneous and black fallows on luvic Chernozem set-aside (1997–2003)

Mulched part by mulching machine Enduro, i.e. finely crushed and left to decompose naturally	one time per year	pure culture of grasses (4 variants)[1] pure culture of legumes (4 variants)[2] grass-legume mixtures (4 variants)[3]	spontaneous fallow (6 replications)
	two time per year		
Mowed part	three time per year with plant biomass removing	pure culture of grasses (4 variants)[1] pure culture of legumes (4 variants)[2] grass-legume mixtures (4 variants)[3]	black (Roundup) fallow (3 replications)

[1]mountain brome (50 kg/ha), false oat-grass (50 kg/ha), meadow fescue (30 kg/ha) and orchard grass (18 kg/ha); [2]white clover (12 kg/ha), black medick (20 kg/ha), birdsfoot trefoil (15 kg/ha) and alfalfa (18 kg/ha); [3]crownvetch + mountain brome (10 + 20 kg/ha), birdsfoot trefoil + meadow fescue (6 + 20 kg/ha), white clover + orchard grass (6 + 15 kg/ha) and black medick + false oat-grass (10 + 20 kg/ha)

mowing and mulching fallows (controlled, black, spontaneous); period after desiccation (2003–2004) was black fallow with repeated Roundup applications and period after re-grassing (2004–2006) was a mowed monoculture of Italian ryegrass *Lolium multiflorum* Lam. cv. Lolita (60 kg/ha).

Soil samples were taken as a mixture of 10 subsamples from the profile (0–200 mm) using an Eijkelkamp sampler every year three times in spring (April, May, June), one time in summer (July) and three times in autumn (September, October, November). To eliminate the edge effect, only the central area of each experimental plot was used for soil samples collection. Soil samples were transported in a cooling box (temperature 6–12°C), sieved (mesh 2 mm) and stored in a refrigerator (4–6°C) for two weeks. Before biological analysis all soil samples were pre-incubated in laboratory over night at room temperature (22 ± 2°C).

The list of methods used for soil sample analysis is as follows:

Texture – sand, silt, and clay content was determined by the pipette method (ISO 11277);

pH (H_2O) – 15 ml of deionized water (DW) and 3 g of a field-moist soil sample were shaken (1 h) and pH was determined with an electrode by Hanna;

Total Kjeldahl nitrogen (TKN) – European Standard EN 13342;

Soil organic carbon (C_{org}) – colorimetric determination of a field-moist soil sample at 600 nm (Sims and Haby 1971, Růžek et al. 2009);

Humus quality (A400/A600) – determined using the ratio of absorbances of a field-moist soil sodium pyrophosphate extract at wavelengths of 400 and 600 nm (Růžek et al. 2009);

Microbial biomass carbon (MBC) – determined by the re-hydration technique (RHD); colorimetric C determination at 590 nm (Blagodatskiy et al. 1987, Růžek et al. 2009);

Available organic carbon (C-K_2SO_4) – extracted by 0.5 mol/l K_2SO_4 from field-moist soil; colorimetric C determination at 590 nm (Vance et al. 1987, Růžek et al. 2009);

Basal respiration (BR) – 50 g field-moist soil were incubated with 2 ml distilled water for 20 h at 29°C; interpherometric CO_2 detection (Novák and Apfelthaler 1964);

Control nitrification (N-NO_3^-) – 5 g field-moist soil with 1 ml distilled water were incubated 7 days at 29°C; N-NO_3^- was detected by automatic flow analyzer (CFA) SAN Plus system type; Skalar Analytical, Breda, the Netherlands; (ISO/TS 14256-1 2003);

Arylsulfatase activity (ARS) – 1 g field-moist soil (three repetitions and control) was incubated with 4 ml acetate buffer and 1 ml p-nitrophenylsulphate for 1 h at 37°C (control without 1 ml *p*-nitrophenylsulphate); before filtration 1 ml *p*-nitrophenylsulphate was added to the control and 25 ml distilled water were amended to both variants; 6 ml filtrate were taken and mixtured with 4 ml 0.5 mol/L NaOH; colorimetric determination at 420 nm (Tabatabai and Bremner 1970, Nováková and Voříšek 2006).

The following characteristics were calculated:

Biomass-specific available organic carbon (C-K_2SO_4/MBC) (Hofman and Dušek 2003);

Metabolic quotient: BR/MBC = C respirated (mg/h) calculating on 1 g MBC.

Statistical evaluation was performed using one-way ANOVA (Statgraphic Centurion XV). Scheffe's test was used to evaluate the differences between variants and periods.

RESULTS AND DISCUSSION

Characteristics of the studied luvic chernozem are presented in the Tables 2 and 3. Table 2 covers averages from 4, 8 and 16 sampling terms from main three periods of experiment (November 2002 to October 2006) and Table 3 is giving more precise information about averages (4 sampling terms) from the first period of experiment (November 2002–June 2003), respectively. Statistical evaluations by Scheffe's test were processed always considering the smallest group of values.

Soil organic matter (SOM) mineralization – basal respiration (BR)

The average values of basal respiration were from 1.08 to 1.70 mg C/kg DM/h (Table 2), which is typical for grass-covered luvic chernozems. Černohlávková et al. (2009) presented 1.67 ± 0.12 mg C/kg DM/h for sandy grassland Cambisol (C_{org} 5.76%; MBC 1270 mg C/kg DM). However, 55% of the experimental plots showed respiration values in the upper margin of this interval in the third period (data not published). Roundup desiccation provided a statistically insignificant increase of basal respiration from 1.08 to 1.25 mg C/kg DM/h (Table 2), and re-grassing led to a significant increase of the average value to 1.70 mg C/kg DM/h (Table 2). However, 7 months after the re-grassing respiration was even 2.67 mg C per kg

Table 2. Chemical and biological soil characteristics in main three periods of experiment with luvic Chernozem set-aside

Parameters	Before Roundup desiccation November 2002–June 2003 averages from 4 samplings	After Roundup desiccation July 2003–August 2004 averages from 8 samplings	After Italian ryegrass grassing September 2004–October 2006 averages from 16 samplings
DM[1] (%)	84.08[a]	84.77[a]	84.45[a]
C_{org} (%)	1.55[a]	1.93[b]	1.86[b]
TKN[2] (%)	0.16[a]	0.20[b]	0.20[b]
Ratio C_{org}/TKN	9.7[a]	9.7[a]	9.3[a]
pH (H_2O)	7.6[a]	7.6[a]	7.6[a]
C-K_2SO_4[3]	41.1[b]	48.8[c]	35.8[a]
N-NO_3^- [4]	1.53[a]	2.84[b]	1.09[a]
A 400/A 600[5]	4.86[a]	4.39[b]	4.07[c]
MBC[6]	452.4[a]	765.8[c]	593.1[b]
((C-K_2SO_4/MBC) × 100)[7]	9.08[b]	6.37[a]	6.04[a]
ARS[8]	341.4[b]	245.1[a]	372.5[b]
BR[9]	1.08[a]	1.25[a]	1.70[b]
qCO_2 = (BR/MBC) × 1000)[10]	2.4[b]	1.6[a]	2.9[c]

[1]dry mass of soil gravimetrically; 5 g at 105°C/24 h; [2]total Kjeldahl nitrogen (European Standard EN 13342); [3]0.5 mol/l K_2SO_4 extracted C (mg/kg DM); [4]1 mol/l KCl extractable N-NO_3^- (mg/kg DM/day); [5]humus quality was determined using the ratio of absorbance of a field-moist soil sodium pyrophosphate extract at the wavelengths of 400 and 600 nm; [6]microbial biomass carbon, re-hydration technique (mg C/kg DM; field-moist soil sterilization at 64°C in forced-air ventilation oven, 24 h; K = 0.25); [7]biomass specific available organic carbon; [8]arylsulfatase activity (mg PNP/kg DM/h); [9]basal respiration (mg C/kg DM/h); [10]metabolic quotient (mg C/g MBC/h); [a, b, c]different characters indicate a significant difference (ANOVA; Scheffe's Test)

DM/h (April 2005) and the average BR value of the last three collections was 1.36 mg C/kg DM/h (July–October 2006) – data not presented.

In general, the average respiration value was lowered by test plots where either black (= Roundup) fallow, mulched mixtures, or mowed pure cultures of grasses were maintained. On the other hand, a spontaneous fallow, mowed pure cultures of legumes and mulched pure cultures of grasses, as well as pure cultures of legumes, increased the average. A highly significant increase in CO_2 production after grassing of black (= Roundup) fallow by *Lolium multiflorum* Lam. is unfavorable. Maintenance of set-aside soil which increases greenhouse gas concentration in the atmosphere is not desirable. A twice-yearly mowed Italian ryegrass monoculture with plant biomass removal in 2005–2006 increased the mineralization of SOM more than mulching did (Table 3). A mulched grass-legume mixture with birdsfoot trefoil and meadow fescue best preserved the biological properties of luvic chernozem, according to our results, and also in regard to contamination with weeds (Brant et al. 2000, Svobodová et al. 2004).

Control nitrification (N-NO_3^-)

In our experiment we have determined (Table 2) a significant N-NO_3^- increase after desiccation in the soil because of decreased immobilization nitrates by the plants.

Arylsulfatase activity (ARS)

Roundup desiccation strongly (with high significance) decreased ARS activity (Table 2). The ARS values in topsoil under mowed pure cultures of grasses were lower. The highest values of ARS occurred on plots with mulched pure cultures of legumes (Table 3). Similarly green manure was proposed as a soil conserving and more bioresource-efficient alternative to bare fallow by Biederbeck et al. (2005) in a comparable 6-year study focused on a partial fallow with four annual legumes. This is confirmed by comparison with tilled fallow-wheat (*Triticum aestivum* L.) with increasing parameters: 170% for MBC, 191% for microbial biomass nitrogen, 202% for dehy-

Table 3. Chemical and biological soil characteristics of mowed and mulched plots before desiccation (sampled from November 2002 to June 2003)

Parameter	7 years mowed (1997–2003)			7 years mulched (1997–2003)		
	grasses[1]	legumes[2]	their mixtures	grasses	legumes	their mixtures
DM[3] (%)	84.84[a]	85.23[a]	85.25[a]	84.01[a]	83.73[a]	84.31[a]
C_{org} (%)	1.58[ab]	1.44[a]	1.47[a]	1.80[c]	1.83[c]	1.77[bc]
TKN[4] (%)	0.17[ab]	0.16[a]	0.15[a]	0.19[c]	0.20[c]	0.18[bc]
Ratio C_{org}/TKN	9.3[ab]	9.1[a]	9.8[b]	9.5[ab]	9.2[a b]	9.8[ab]
pH (H_2O)	7.6[ab]	7.7[b]	7.6[a b]	7.5[a]	7.7[b]	7.7[b]
C-K_2SO_4[5]	33.9[a]	42.0[ab]	37.7[a b]	35.5[a]	47.1[b]	44.3[b]
N-NO_3^-[6]	1.76[a]	1.96[a]	1.61[a]	1.99[a]	2.51[a]	2.48[a]
A 400/A 600[7]	4.48[a]	4.71[a]	4.56[a]	4.49[a]	4.54[a]	4.60[a]
MBC[8]	483.5[a]	499.7[ab]	507.2[ab]	535.1[ab]	603.7[b]	553.8[ab]
((C-K_2SO_4/MBC) × 100)[9]	7.01[a]	8.41[a]	7.43[a]	6.63[a]	7.80[a]	8.00[a]
ARS[10]	266.3[a]	319.6[ab]	341.9[ab]	315.0[ab]	383.8[b]	352.2[ab]
BR[11]	1.20[a]	1.22[a]	1.27[a]	1.37[a]	1.33[a]	1.30[a]
qCO_2 = (BR/MBC) × 1000)[12]	2.5[a]	2.4[a]	2.5[a]	2.6[a]	2.2[a]	2.3[a]

[1]pure culture of grasses: mountain brome (50 kg/ha), false oat-grass (50 kg/ha), meadow fescue (30 kg/ha) and orchardgrass (18 kg/ha); [2]pure culture of legumes: white clover (12 kg/ha), black medick (20 kg/ha), birdsfoot trefoil (15 kg/ha) and alfalfa (18 kg/ha); [3]dry mass of soil gravimetrically; 5 g at 105°C/24 h; [4]total Kjeldahl nitrogen (European Standard EN 13342); [5]0.5 mol/l K_2SO_4 extracted C (mg/kg DM); [6]1 mol/l KCl extractable N-NO_3^- (mg/kg DM/day); [7]humus quality was determined using the ratio of absorbances of a field-moist soil sodium pyrophosphate extract at wavelengths of 400 and 600 nm; [8]microbial biomass carbon, re-hydration technique (mg C/kg DM; field-moist soil sterilization at 64°C in forced-air ventilation oven, 24 h; K = 0.25); [9]biomass specific available organic carbon; [10]arylsulfatase activity (mg PNP/kg DM/h); [11]basal respiration (mg C/kg DM/h); [12]metabolic quotient (mg C/g MBC/h); [a, b, c] different characters indicate a significant difference (ANOVA; Scheffe's Test) at the whole line

drogenase, 171% for phosphatase, and 287% for arylsulfatase activity.

Microbial biomass carbon (MBC)

The RHD technique which was used for determination of MBC in luvic chernozem set-aside in the profile of 0–200 mm, reached values that ranged within 452.4–765.8 mg C/kg DM (Table 2). Surprisingly, the highest values were typical for the second period following Roundup desiccation (Table 2), and the lowest values were for the first period with various mowed or mulched herbaceous cover. There were highly significant differences among all three periods. Mulching and the presence of legumes in herbaceous cover significantly increased MBC values (Table 3), which is confirmed with significantly highest value before desiccation 603.7 mg C/kg DM. Dying plants upon desiccał tion support C immobilization in the microbial cells and this leads to the highest average values (765.8 mg C/kg DM; Table 2) in the course of the entire experiment. Ecosystem stabilization upon grassing is gradual. The experiment was terminated 25 months after grassing, while ecosystem stabilization was still going on. This was confirmed by the average MBC values (560.2 mg C/kg DM; data not presented) of the last three samplings.

Ratio ((BR/MBC) × 1000)

Metabolic quotient (qCO_2) is a very common indicator of the metabolic status of soil microbial associations. Araujo et al. (2008) used qCO_2 in a study designed to evaluate conventional and organic systems. They showed that qCO_2 was greater in conventional than in organic systems. These results indicate that organic practices rapidly improve soil microbial characteristics and slowly increase soil organic C. According to our experience, very low values (under 1.5 mg C/g MBC/h) originated in low mineralization activity. In our study, qCO_2 strongly (with high significance) declined following Roundup desiccation from 2.4 to

1.6 mg C/g MBC/h, and strongly (with high significance) moved to a level of 2.9 mg C/g MBC/h after re-grassing of Roundup fallow by a twice-yearly mowed Italian ryegrass monoculture (Table 2). The metabolic quotient obviously signaled unfavorable soil maintenance with higher efflux of CO_2 to the atmosphere. According to our results, a mulched grass-legume mixture with birdsfoot trefoil and meadow fescue is a better option.

Biomass-specific available organic carbon

The ratio, ((C-K_2SO_4/MBC) × 100), is another criterion for the evaluation of microbial associations in soil (Hofman and Dušek 2003). This ratio maps a C-K_2SO_4 accumulation out of the microbial cells against carbon immobilization into microbial cells. In other words, it maps the ability of soil microbial communities to utilize their own extracellular metabolites. A complex microbial community (one that is not exposed to stress) uses these trophically valuable and unstable C-K_2SO_4 substances and thereby maintains their low level (Růžek et al. 2009). In this study, the ratio reached its highest values (9.08; Table 2) in the first period before Roundup desiccation. After Roundup desiccation biomass-specific available organic carbon strongly (with high significance) plunged to 6.37 in connection with a strong increase of MBC (over 69%). However, in the last period, after re-grassing by a twice-yearly mowed Italian ryegrass monoculture, ratio decreased even more to a standard level of 6.04, which signals the most favorable status since the year 1997. This ratio decreased weakly mowed or mulched pure cultures of grasses in contrast to other variants (Tables 2 and 3). This contradictory result in comparison with metabolic quotient, which is increasing, follows from the fact that for decreasn ing values of biomass-specific available organic carbon C-K_2SO_4 values are of the key importance. During stabilization of ecosystem C-K_2SO_4 values are declining faster than basal respiration, which is crucial for metabolic quotient.

Carbon of soil organic matter (C_{org}) and total Kjeldahl nitrogen (TKN)

Soil organic matter is a potential source of energy for soil microbial communities. Its soil content is a stable soil property, which shows year-on-year non-measurable variation without human interference. In this study, the level was typical for grassed luvic chernozems (C_{org}: 1.55–1.93%; TKN: 0.16–0.20%). Růžek et al. (2006) cited that typical values for Chernozems in the Czech Republic are C_{org}: 1.52 ± 0.28 and TKN: 0.18 ± 0.03. After the first Roundup desiccation (July 10, 2003) of a very weedy seven-year-old herbaceous cover, a considerable amount of root mass remained in the soil profile of 0–200 mm. Due to an extremely dry period (August–November 2003), this root matter was very slowly mineralized and immobilized into a microbial biomass, and affected the monitored parameters, including C_{org} and TKN (Table 3). On the other hand, the ratio C_{org}/TKN after Roundup desiccation (Table 2) was not influenced and remained in the range 9.1–9.8 (Table 3), which is typical for luvic chernozems. Růžek et al. (2006) cited as a typical value 8.7 ± 1.7.

Desiccation and re-grassing had no effect on the dry mass of soil and pH value (Table 2).

Available organic carbon (C-K_2SO_4)

A significant increase was measured upon desiccation, followed with a significant reduction upon re-grassing on the lowest values for the entire period (1997–2006) (Table 2).

Humus quality (A 400/A 600)

A significant decrease of the values continued during the course of the whole experiment, i.e. the quality of extractable humic substances was improving (Table 2).

Among the general conclusions of this article it should be emphasized that mulching of pure cultures of grasses or legumes on land set-asides contributes to a highly significant soil organic matter accumulation, and this is a potential source of CO_2 for atmosphere. Mulched grass-legume mixtures are a much better way. Repeated Roundup desiccation resulted in a strong decrease of metabolic quotient (–29%) and arylsulfatase activity (–28%) and conversely to significantly increasing microbial biomass carbon (+69%) and nitrate nitrogen (+86%). Following re-grassing compensated the changes shown. We recommend for practise mulched grass-legume mixture (meadow fescue – *Festuca pratensis* Huds. and birdsfoot trefoil – *Lotus corniculatus* L.; 20 + 6 kg per ha) that presents also the best option, with regard to contamination with weeds.

Acknowledgements

The authors thank Dr. Jakub Hofman (RECETOX; Masaryk University Brno, Czech Republic) for pre-review, correction and valuable consultation.

REFERENCES

Araujo A.S.F., Santos V.B., Monteiro R.T.R. (2008): Responses of soil microbial biomass and activity for practices of organic and conventional farming systems in Piaui state, Brazil. European Journal of Soil Biology, *44*: 225–230.

Biederbeck V.O., Zentner R.P., Campbell C.A. (2005): Soil microbial populations and activities as influenced by legume green fallow in a semiarid climate. Soil Biology and Biochemistry, *37*: 1775–1784.

Blagodatskiy S.A., Blagodatskaya Y.V., Gorbenko A.Y., Panikov N.S. (1987): A rehydration method of determining the biomass of microorganisms in soil. Soviet Soil Science, *19*: 119–126.

Brant V., Šantrůček J., Svobodová M. (2000): The effect of different ways and frequency of harvesting of set aside arable land on the weed infestation. Zeitschrift fur Pflanzenkrankheiten und Pflanzenschutz-Journal of Plant Diseases and Protection, Special Issue *7*: 105–112.

Černohlávková J., Jarkovský J., Hofman J. (2009): Effects of fungicides mancozeb and dinocap on carbon and nitrogen mineralization in soils. Ecotoxicology and Environmental Safety, *72*: 80–85.

EN 13342 (2000): Characterization of sludges. Determination of Kjeldahl nitrogen. European Committee for Standardization, Brussels, Belgium.

Hiller E., Cernansky S., Krascsenits Z., Milicka J. (2009): Effect of soil and sediment composition on acetochlor sorption and desorption. Environmental Science and Pollution Research, *16*: 546–554.

Hofman J., Dušek L. (2003): Biochemical analysis of soil organic matter and microbial biomass composition – a pilot study. European Journal of Soil Biology, *39*: 217–224.

ISO 11277 (1998): Soil quality – determination of particle size distribution in mineral soil material. Method by sieving and sedimentation following removal of soluble salt organic matter and carbonates. International Organization for Standardization, Geneve, Switzerland, 30.

ISO/TS 14256-1 (2003): Soil quality – determination of nitrate, nitrite and ammonium in field-moist soils by extraction with potassium chloride solution – Part 1: Manual method. International Organization for Standardization. Geneve, Switzerland.

Kunzová E., Hejcman M. (2009): Yield development of winter wheat over 50 years of FYM, N, P and K fertilizer application on black earth soil in the Czech Republic. Field Crops Research, *111*: 226–234.

Novák B., Apfelthaler R. (1964): A contribution to method of determining respiration as an indicator of microbiological processes in soil. Rostlinná Výroba, *10*: 145–150. (In Czech)

Nováková M., Voříšek K. (2006): Microbiological parameters of soil set aside before and after desiccation. Plant, Soil and Environment, *52*: 97–104.

Reinecke A.J., Helling B., Louw K., Fourie J., Reinecke S.A. (2002): The impact of different herbicides and cover crops on soil biological activity in vineyards in the Western Cape, South Africa. Pedobiologia, *46*: 475–484.

Růžek L., Růžková M., Voříšek K., Kubát J., Friedlová M., Mikanová O. (2009): Chemical and microbiological characterization of Cambisols, Luvisols and Stagnosols. Plant, Soil and Environment, *55*: 231–237.

Růžek L., Voříšek K., Nováková M., Strnadová S. (2006): Microbial, chemical and textural parameters of main soil taxonomical units of Czech Republic. Plant, Soil and Environment, *52*: 29–35.

Růžková M., Růžek L., Voříšek K. (2008): Soil biological activity of mulching and cut/harvested land set aside. Plant, Soil and Environment, *54*: 204–211.

Sims J.R., Haby V.A. (1971): Simplified colorimetric determination of soil organic matter. Soil Science, *112*: 137–141.

Svobodová M., Šantrůček J., Urbanec J. (2004): Succession changes of temporary grass stands on set-aside land. Plant, Soil and Environment, *50*: 108–115.

Tabatabai M.A., Bremner J.M. (1970): Arylsulfatase activity of soils. Soil Science Society of America Journal, *34*: 225–229.

Vance E.D., Brookes P.C., Jenkinson D.S. (1987): An extraction method for measuring soil microbial biomass C. Soil Biology and Biochemistry, *19*: 703–707.

Received on August 17, 2010

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