

## Changes in the soil's biological and chemical properties due to the land use

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**Abstract:** Increasing the soil productivity is challenged by the increasing biotic threat to plants and microorganisms, by the resistance to agrochemicals, and by the declining soil health. Soil management strategy is, therefore, aimed at erosion prevention and the minimisation of soil organic matter losses. A key factor in an agroecosystem is the appropriate biological stability. It is essential not only at present, but also for further sustainable agriculture. This study was based on the hypothesis that afforestation and conversion from arable land to permanent grassland improves the organic matter status and biological stability in the agroecosystem. The experiment was conducted from 2014 to 2018 in the Uhřetice bio-corridor (Kroměříž region, the Czech Republic). Haplic Luvisol has been investigated for its basic biological and chemical properties after the arable land was converted to a natural vegetation system. The afforested segment (F), permanent grassland segment (G), and arable land segment (A) have been sampled in the upper soil horizon (0–0.30 m). Standard analytical methods were applied for the determination of the basic soil properties. A principal component analysis and factor analysis were used for interpreting the connection between the parameters of the soil organic carbon, the humic substances, the humic acids, and the fulvic acids, the agrochemical properties of the soil (the pH, the content of the nitrogen, phosphorus and potassium, etc.), and the soil biological properties (basal soil respiration (BSR), the ratios of the N/BSR, NG/BSR, etc.). After five years of investigation, the differences in the studied parameters were evident. The factor analysis and multivariate exploratory techniques showed that the soil properties were grouped based on the management into three different categories – F, G and A. The different land use directly influenced the quality and stability of the humic substances, basal soil respiration, and carbon and nitrogen utilisation. In comparison to the arable land, the forest and grassland were considered to have a higher accumulation potential of carbon and nitrogen. A negative correlation between the soil basal respiration ( $r = -0.95$ ); total nitrogen ( $r = -0.93$ ); total organic carbon ( $C_{ox}$ ) content ( $r = -0.82$ ); and partial Ca ( $r = -0.82$ ) was found. A positive correlation ( $r = 0.80$ ) between the humic substances (C-HS) and soil reaction (pH) was determined.

**Keywords:** biological and chemical soil properties; Haplic Luvisol; multivariate statistical techniques

Land use conversion and intensive agriculture exploitation frequently results in significant humus loss via soil erosion, degradation and depletion. European agricultural policy identifies erosion and loss of soil organic matter as the most serious threat for agricultural lands (EU Thematic Strategy for Soil

Protection, EC 2012). All agricultural practices are recommended to be associated with the appropriate conservation policy (Doni et al. 2014; Plaza-Bonilla et al. 2015; Lal 2016). In the Czech Republic, more than 54% of the agricultural land is threatened by soil degradation (Šarapatka & Bednář 2015). According

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to Pražan and Dumbrovský (2011), and Skalský and Vopravil (2014), one of the most important measures on how to prevent the erosion of agricultural soils and stabilise the agroecosystem is the construction of bio-corridors and bio-centres. These stabilising systems represent the improvement of ecological conditions in the environment. A bio-corridor is defined as an ecologically important segment of landscape connecting two bio-centres. The main aim is to improve the contact, migration, and security of living organisms. Its functionality is given by parameters (length and width), and by the structure of the plant species. Different parts (segments) of the bio-corridors are usually afforested, or covered by permanent grassland. Many studies focus on the relationship between the biodiversity and ecosystem function (Loreau et al. 2001; Cardinale et al. 2012; Zhang et al. 2017). Biodiversity appears to affect the ecosystem stability in many different ways. Nannipieri et al. (2003) and Philippot et al. (2013) documented that soil microorganisms are extremely diverse and governed by the most ecologically relevant biochemical processes. They showed that the plant input, the soil heterogeneity, the spatial and temporal heterogeneity of the microorganism's population, and the climatic conditions are the main factors influencing the decomposition rate.

The main aim of this study was to evaluate the microbial activity, humic substances content and stability, and changes in the soil chemical properties within the different bio-corridor segments (arable soil, permanent grassland, and forest). Using multivariate exploratory techniques (principal component analysis and factor analysis), we were able to show differences between the forest, grassland, and arable land.

## MATERIAL AND METHODS

The field experiment is located in the Uhřice bio-corridor (Kroměříž region, the Czech Republic). The average annual air temperature is 8.5 °C. The average annual precipitation is 650 mm (Květoň 2001). The bio-corridor was constructed in 2014 with the aim to reduce water and wind erosion. The second reason was to connect two different bio-corridors in the Uhřice cadastre. The studied bio-corridor was divided into the afforested segment F1 (3 436 m<sup>2</sup>); segment F2 (3 898 m<sup>2</sup>); segment F3 (5 052 m<sup>2</sup>). The permanent grassland (G) was between the F1, F2, and F3 segments. The plan of planting trees was as follows: – alder (*Alnus glutinosa*); hornbeam (*Carpinus*

*betulus*); maple (*Acer platanoides*); elm (*Ulmus laevis*); bird cherry (*Prunus padus*); wild pear (*Pyrus pyraster*); and dogwood (*Cornus sanguinea*). All the trees were two years old and planted 1 × 1 m apart according to the bio-corridor project. None of the tree species was dominant. The selected trees species were very close to the natural flora found in this region. Monitoring the biological and chemical soil properties in all of the bio-corridor segments (A = arable land; G = permanent grassland; F = forest) was carried out over the five years (2014–2018). The soil sampling was carried out twice a year (in March and in October) at a depth of 0–0.30 m. The samples for the determination of the chemical properties were received as follows: thirty samples were taken at random in every part of the bio-corridor and bulked to form a single sample (A, G, F1, F2, and F3). All the average samples were dried and sieved through a 2 mm sieve. The instructions for the microbiological sampling and chemical analysis follows the methodology of Zbíral (2016). The basal soil respiration was determined as follows: 50 g of the soil sample was sieved through a 5 mm sieve, the greater part of the soil skeleton fraction, impurities and residues of vegetable or animal material were removed, and stored in a fridge (temperature of 5 °C) for 3 weeks according to the methodology. The basal soil respiration (BSR) was determined at the same moisture as the soil samples. The substrate induced respiration was measured after the glucose addition (G; 2 ml of 25% glucose), ammonium sulphate solution addition (N; 2 ml of a 1 : 1 solution of ammonium sulphate and water), and glucose + ammonium sulphate solution addition (G + N; 2 ml of 25% glucose and 2 ml of the ammonium sulphate solution). A Vaisala GMT 222 device was used (Vaisala Corporation, Finland). The ratios of N/BSR; NG/BSR; G/BSR; G/N were evaluated according to Novák and Apfelfhaler (1964) and Strálsková et al. (2001). The soil samples used for total organic carbon (C<sub>ox</sub>) determination were sieved through 2 mm sieve. Furthermore, we followed the instructions for the oxidimetric determination of the carbon given by Nelson and Sommers (1996). The humus content was calculated by multiplying the C<sub>ox</sub> content by the coefficient of 1.724. The total nitrogen (Nt) was determined according to the Kjeldahl method. The C/N ratio was assessed. The fractional composition of the humic substances (HS), and humic acids (HA) and fulvic acids (FA) ratio (HA/FA) followed the methodology proposed by Kononova (1963). The instructions for the chemical analysis

followed methodology applied by Zbírál (2016) and Pospíšilová et al. (2016). The soil reaction (1 : 2.5 suspension in water and 1 M KCl) was determined by a potentiometric method using a Hanna pH meter (HI 98120, Hanna Instruments, USA). Mehlich-III was applied for the available nutrients content determination. Ten average samples from each variant were analysed for all the studied parameters.

The statistical analysis, including the graphical outputs, was carried out using Statistica (Ver. 13, 2018). For the statistical data processing and evaluation, we applied: exploratory data analysis (EDA), analysis of variance (ANOVA), Tukey's test (HSD test), Fisher's LSD test (LSD test), principal component analysis (PCA) and factor analysis (Meloun & Militký 2011). The PCA was used for interpreting the parameters of the soil organic matter ( $C_{ox}$ , HS, HA, FA, HA/FA ratio), the agrochemical properties of the soil (the pH, the content of the nitrogen, phosphorus, magnesium, calcium, and potassium) and the soil biological properties (BSR, the ratios of N/BSR, NG/BSR). The selected measured characteristics were used as predictors (factors); they were chosen on the basis of an eigenvalue graph. Variables with an impaired assumption of normality were converted using logarithmic transformation. As part of step 1, the PCA was carried out with all the variables to compute the most important variables. Step 2 involved selecting the active and supplementary variables for better interpretation. In the case of a lower number of samples, this stepwise analysis significantly improves the outcome of the PCA. The PCA was used for calculating a component's weight for the investigated variables (Meloun & Militký 2011). Based on the correlations and contributions in the convincing factors, each of the characteristics was subsequently assessed for its relevance explaining the multidimensional dependencies (correlations) in the factorial plane. The factor analysis analysed the internal contexts and relationships (correlations) and revealed the basic structure of the source data matrix. The factor analysis also identified the factors and then assigned to each factor a content meaning (physical or chemical) (Meloun & Militký 2011). The statistical significance was assessed at a significance level of 0.05.

## RESULTS

The Haplic Luvisol was loamy textured (40% of clay particles), with a weak acid reaction ( $pH(KCl) = 5.7–6$ ),

and moderate humus content (1.36–1.60%). The humus quality evaluated by the HA/FA ratio was in the middle (HA/FA about 1). The content of the available macronutrients (Ca, P, K, Mg) determined according to the Mehlich III method was high. The calcium content varied from 2 500–3 200 mg/kg and the magnesium content varied from 220–300 mg/kg. The phosphorus content varied from 100–185 mg/kg and the potassium content varied from 200–300 mg/kg. The evaluation was performed according to Zbírál (2016) and Pospíšilová et al. (2016). The differences in the  $pH(KCl)$ ,  $C_{ox}$ , Nt, C/N, C-HS, HA/FA, P, K, Ca, Mg, BSR, N/BSR, G/BSR, NG/BSR in the selected segments of the bio-corridor were evaluated using the PCA and factor analysis (Figures 5 and 6). The average values of the exchangeable soil reaction were 5.84 (A), and 5.84 (F). The highest value of the  $pH(KCl)$  ratio was 6.02 (G). The differences in the exchangeable soil reaction were statistically significant ( $P = 0.05$ ). The average values of the total carbon ( $C_{ox}$ ) varied from 1.39% (A) to 1.54% (G and F). The carbon input in the forest and grassland was significantly higher compared with the arable land (Figure 1). The total nitrogen (Nt) was 0.29% in the grassland, followed by the forest (0.26%) and arable land (0.20%). The results of the carbon and nitrogen content were statistically different in the studied segments (A, F, G). The obtained results indicated that the best conditions for the soil microorganisms ( $C/N = 5.37$ ) were in the grassland, followed by the forest and arable land (Figure 1). The highest amount of the HS was in the forest and represented 0.43 mg/kg. The HS content was 0.39 mg/kg in the grassland and 0.34 mg/kg in the arable land (Figure 2). Increasing both the HA and FA is statistically significant in the G and F segments ( $P = 0.05$ ). An HA/FA ratio above 1 documents the formation of young HA and FA and the increasing HS total amount in the studied segments (G) and (F). The available nutrients are presented in Figure 3. The content of P, K, Mg (spring/winter seasonal comparison) was lower in the forest (F) compared with the arable land (A) and grassland (G). The calcium content was significantly higher in the grassland (G). The BSR was very low in the arable land ( $< 0.25 \pm 0.01$  mg  $CO_2/h$  per 0.1 kg of soil). Higher values were reached in the grassland ( $0.30 \pm 0.01$  mg  $CO_2/h$  per 0.1 kg of soil) and forest soil ( $0.30 \pm 0.01$  mg  $CO_2/h$  per 0.1 kg of soil) – Figure 4. The calculated N/BSR ratio (= the ratio of the respiration after the nitrogen addition and the basal soil respiration) indicates the physiological utilisation

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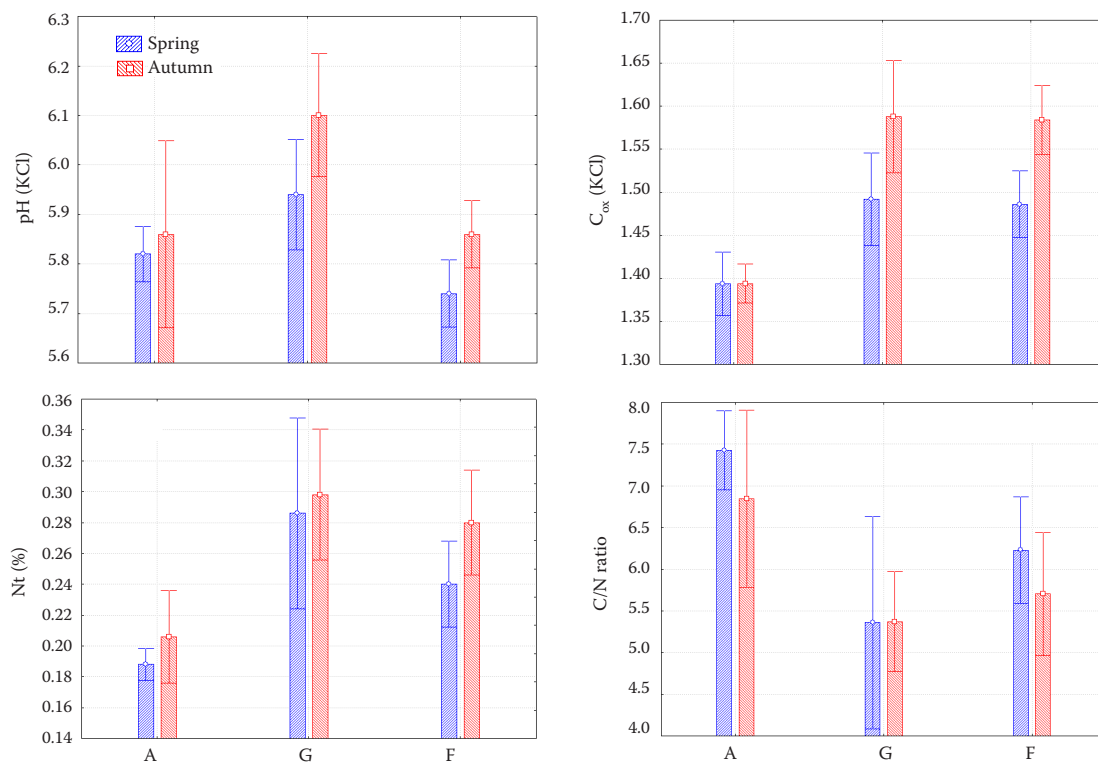


Figure 1. The exchangeable soil reaction (pH(KCl)), total organic carbon ( $C_{ox}$ ), total nitrogen (Nt) and C/N ratio during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F)

of the soil nitrogen. Lower N/BSR values show the lower availability of nitrogen for the microorganisms. If there is enough nitrogen, the addition of nitrogen does not increase the respiration rate and the N/BSR ratio is very close to the value of 1. The N/BSR ratio in the grassland (G) was less than 1, which means enough nitrogen for the soil biota. In this case, the addition of nitrogen caused a decrease

in the respiration intensity. The N/BSR values in the (A) and (F) segments were higher than 1, which means a low physiological utilisation of the nitrogen, and its addition increased the respiration in the (A) and (F) segments. In this case, the lack of available nitrogen was observed in the upper part of the soil profile (Figure 4). The G/N ratio characterises the respiration activity after the glucose (simply a usable

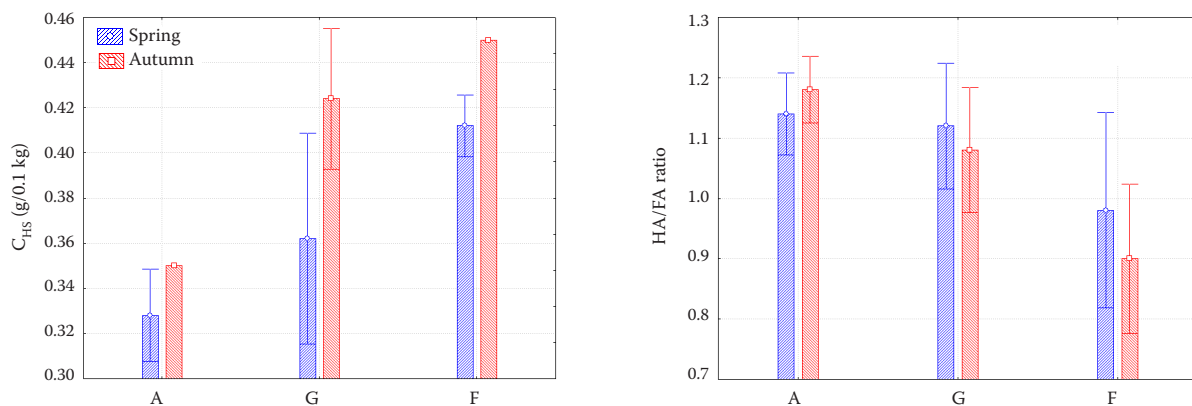


Figure 2. The fractional composition of the humic substances during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F)

$C_{HS}$  – carbon of humic substances; HA – humic acids; FA – fulvic acids

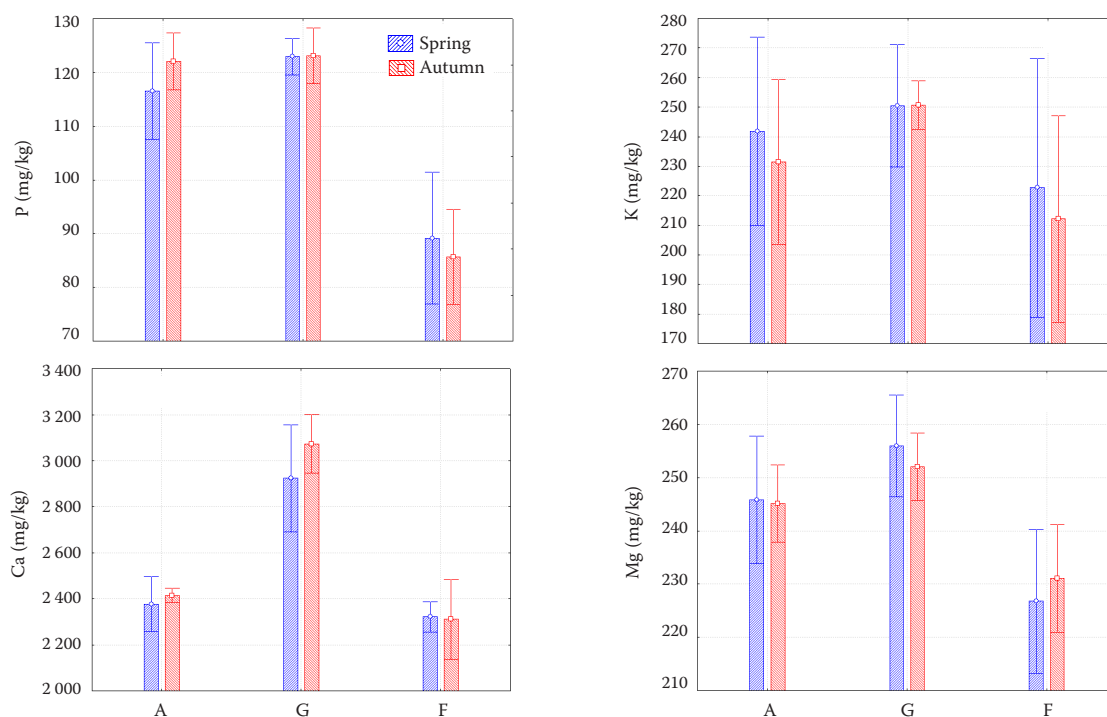


Figure 3. The available soil macronutrients (P, K, Ca, Mg) during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F)

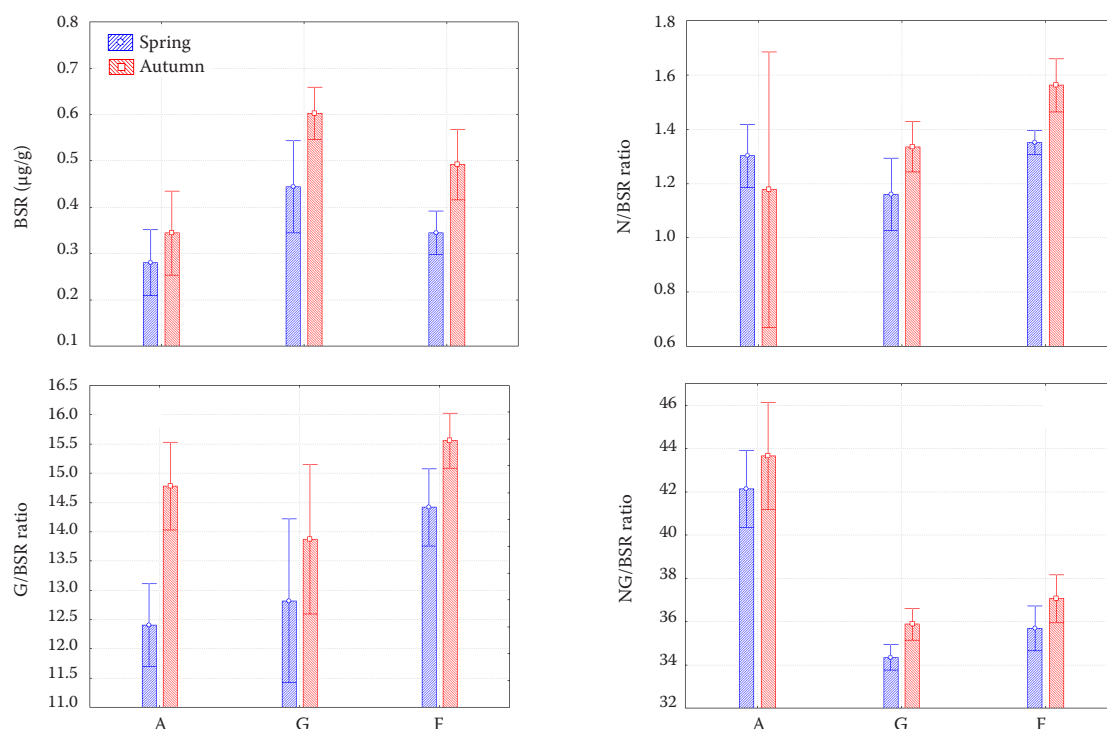


Figure 4. The soil's biological properties during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F).

BSR – basal soil respiration; N/BSR – respiration after the nitrogen (N) addition versus the basal soil respiration; G/BSR – respiration after the glucose (G) addition versus the basal soil respiration; NG/BSR – respiration after the nitrogen and glucose (NG) addition versus the basal soil respiration



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organic substance) and nitrogen addition (simply a usable nitrogen substance), thus, the connection between the physiologically available carbon and nitrogen was evaluated. If the G/N ratio was more than 5, the soil microorganisms were better supplied with organic substances (carbon) than with nitrogen. Values lower than 5 showed the opposite situation (the soil microorganisms were better supplied by nitrogen than by the organic carbon). The obtained G/N ratio was very high (from 6.5 to 18.7) in all the studied segments, which can be interpreted as the soil microorganisms were better supplied with carbon and worse with nitrogen (Figure 4). The stability of the organic matter was assessed by the NG/BSR ratio (the addition of nitrogen and glucose versus the basal soil respiration). Higher values in

the NG/BSR ratio indicated that the organic compounds are not available for the microorganisms. It should also be mentioned that the application of high doses of ammonium sulphate for a long time could have a side effect and can cause soil acidification. Regarding the stability of the HS (Figure 4), the results showed a lower HS stability in the G segment compared with the forest (F) and arable land (A). The principal component analysis was used to group the results of the studied period of 2014–2018 according to the relationship to the analysed variables, the spatial soil variability, and the type of land use. The principal components PC1 (49.84%) and PC2 (38.69%) accounted for 88% of the total variance (Figure 5). The principal component PC1 documents the negative loadings on the soil basal respiration

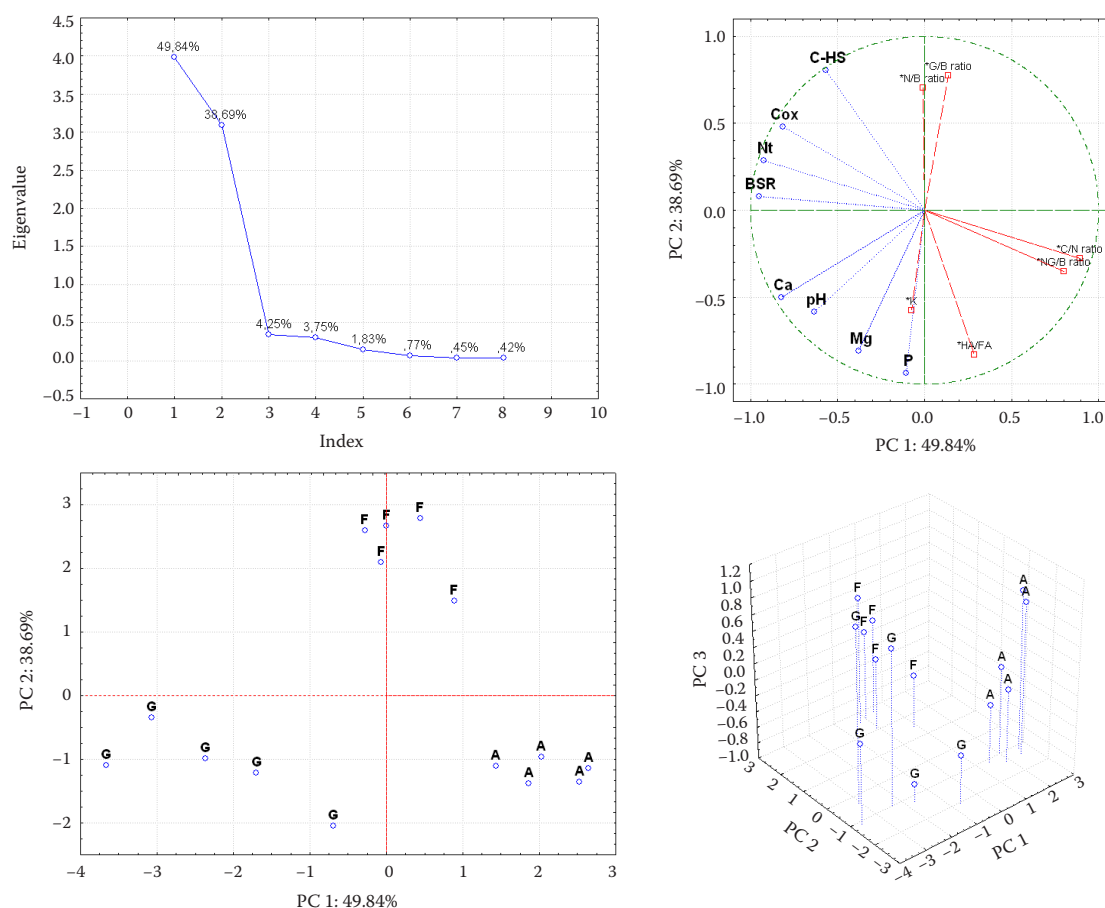


Figure 5. The principal component analysis (PCA) of the studied parameters of the soil organic matter, the agrochemical properties of the soil and the soil biological properties during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F)

$C_{ox}$  – total organic carbon; C-H S – content of carbon of the humic substances; HA – humic acids; FA – fulvic acids; C/N ratio – total carbon and nitrogen ratio; Nt – total nitrogen; BSR – basal soil respiration; G/BSR, N/BSR, NG/BSR ratios – respiration after the glucose (G) and nitrogen (N) addition versus the basal soil respiration; NG/BSR – respiration after the nitrogen and glucose (NG) addition versus the basal soil respiration

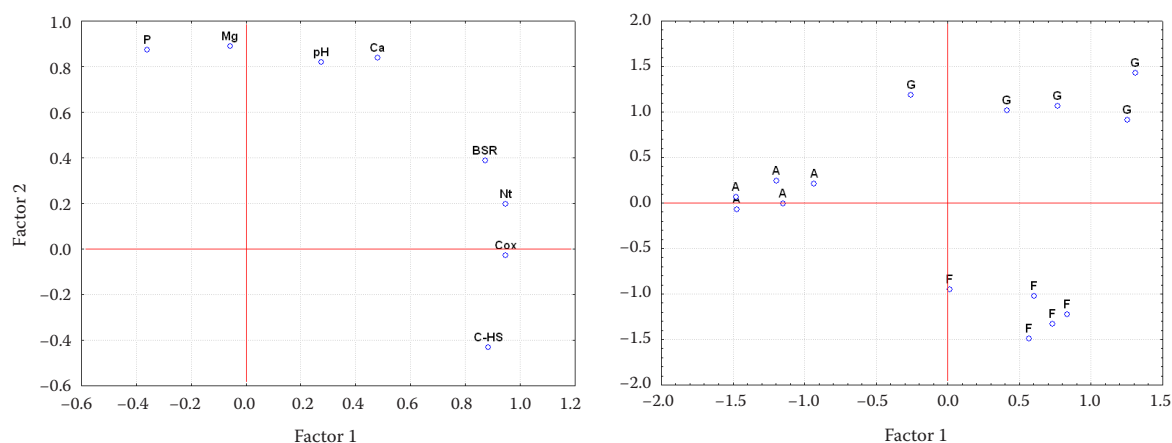


Figure 6. The factor analysis of the studied parameters of the soil organic matter, the agrochemical properties and the biological properties during the studied period (2014–2018) in the arable land (A), permanent grassland (G) and forest soil (F)

$C_{ox}$  – total organic carbon content; C-HS – carbon of the humic substances; Nt – total nitrogen; BSR – basal soil respiration

( $r = -0.95$ ); the total nitrogen ( $r = -0.93$ ); the  $C_{ox}$  content ( $r = -0.82$ ); and the partial Ca ( $r = -0.82$ ). The principal component PC2 shows the negative loadings on the P ( $r = -0.94$ ); the Mg ( $r = -0.80$ ); the positive loadings C-HS ( $r = 0.80$ ); and the soil reaction (pH). On the PCA diagram (Figure 5), the samples are grouped into three different groups according to the land use type (A – arable land; G – grassland; F – forest) in the bottom left quadrant. The different position of A, G, and F contributes to the variables of the soil environment indicating differences in the basal soil respiration, the total Nt,  $C_{ox}$  and calcium content (Figure 5). Factor 1 in the Factor Analysis (Figure 6) shows the relationships between the humic substances, pH and available nutrient content.

The communality represents the proportion of the variability of the attributes expressed by the factors involved. It is similar to the  $R^2$  value we get when explaining the original characters by the regression of selected factors (Sena et al. 2002; Shukla et al. 2006; Meloun & Militký 2011). From the contribution of Factor 1 and 2 to the communality (Table 1), it is clear how the communality acquires high values (more than 0.7) and, thus, the values of the attributes are precisely considered by the proposed factor model.

## DISCUSSION

Similar to our results, Blonska et al. (2017) stressed that the type of vegetation is an important factor of

Table 1. The factor weights and contributions of the selected factors to the communality for each parameter

Parameter	Factor weights		Factor contribution		
	factor 1	factor 2	factor 1	factor 2	communality
pH (KCl)	0.2748	0.8202	0.0755	0.7482	0.6544
$C_{ox}$ (%)	0.9477	-0.0297	0.8982	0.8991	0.8754
Nt (%)	0.9483	0.1956	0.8992	0.9374	0.9341
C-HS (g/0.1kg)	0.8849	-0.4313	0.7831	0.9690	0.9449
BSR ( $\mu\text{g/g}$ )	0.8731	0.3879	0.7623	0.9127	0.8943
P (mg/kg)	-0.3596	0.8729	0.1293	0.8912	0.9198
Ca (mg/kg)	0.4796	0.8369	0.2300	0.9304	0.9414
Mg (mg/kg)	-0.0571	0.8896	0.0033	0.7946	0.7580

BSR – basal soil respiration; Nt – total nitrogen;  $C_{ox}$  – total organic carbon; C-HS – carbon in the humic substances; factor 1 characterised the parameters of the soil organic matter ( $C_{ox}$ , Nt, C-HS and BSR); factor 2 characterised the soil reaction and nutrients content (pH, P, Mg, Ca); this is given in the factor weights (values higher than 0.80)

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the soil microbial activity. Reeves (1997) documented that besides the vegetation and plant diversity, the climatic conditions are also important as well. Němeček et al. (1990) quoted the average BSR value for similar soils was up to 1.38 mg CO<sub>2</sub>/h per 0.1 kg. Sánka et al. (2002, 2018) showed that the BSR in arable land is about 0.29 mg CO<sub>2</sub>/h per 0.1 kg of soil. Our results corresponded more with the general values given by Sánka et al. (2002, 2018). The N/BSR ratio in Luvisols is about 1.07, according to Němeček et al. (1990), this matched our results. Moreover, the respiration tests showed that the least stable were the HS in the grassland (G), followed by the forest (F) and arable land (A). In accordance with Stockman et al. (2013) and Adak et al. (2014), we suppose that if the soil organic matter contains a high portion of easily decomposable materials, the microbial activity correlates to this fraction. Therefore, by inputting an easily decomposable biomass, we can regulate the microbial activity. If the plant input is low, as it was in our case in segment (A), a shortage of the easily decomposable organic materials quickly appeared there. The obtained results also showed that there are statically significant differences in carbon and nitrogen stock in the soil due to the different amounts and quality of the plant input. The multivariate exploratory techniques recognised three different categories – forest (F), permanent grassland (G) and arable land (A) according to the variables of the soil environment. The last indicated differences in the basal soil respiration, Nt, C<sub>ox</sub> and calcium content. The factor analysis was useful in examining the relationships and correlations between the studied parameters. The hypothesis was confirmed that it is possible to modify the soil properties in a relatively short period. This should bring new insights into the link between the land use, the plant input, the microbial activity, the chemical soil parameters and the stability of humic substances.

## CONCLUSION

The biological and chemical soil properties were directly influenced by the different land use. In spite of the short period of time (5 years), the carbon stock, stability of the HS and the basal soil respiration varied in the arable land (A), grassland (G) and forest (F). The afforested part and permanent grassland in the studied bio-corridor had higher accumulation potential compared with the arable land. On the other side, the stability of the humic

substances was higher in the arable land and they became more inaccessible to microbial degradation. An organic input and liming were advised to improve this situation. The factor analysis and multivariate exploratory techniques grouped the studied segments into three different categories – forest (F), permanent grassland (G) and arable land (A).

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