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Effect of gap size and forest type on mineral nitrogen forms under different soil properties

Aleš Kučera¹ · Ladislav Holik² · Elena Muñoz Cerro³ · Jan Petříček¹

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Abstract Gaps play a key role in forest ecosystem development and result from either natural processes or targeted forest management activities. The aim of this study was to investigate the interrelationships of soil properties in each of three forest types and two treatments, and to identify factors that influence levels of soil mineral nitrogen forms. The relation between mineral nitrogen and factors of soil parameters and stand type (European beech, Norway spruce, mixed stand) categories were investigated. The spruce forest type stored significant nitrogen in both mineral forms of nitrogen. Moreover, there was a significant linear dependence between $N-NO_3^{-1}$ (nitrate anion) concentrations and cation exchange capacity (CEC) parameters such as base cation contents (S-CEC) and potential ureolytic activities (UreasePot), as well as between N-NH4⁺ (ammonium cation) concentrations and both hydrolytic acidities (Ha-CEC) and ureolytic activities.

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Ladislav Holik holik@vurv.cz

- ¹ Department of Geology and Pedology, Mendel University in Brno, Zemědělská 3, 61300 Brno, Czech Republic
- ² Crop Research Institute, Drnovská 507/73, 161 06 Praha 6 Ruzyně, Czech Republic
- ³ Institute for Research in Sustainable Forest Management, University of Valladolid, Palencia, Castille and León, Spain

The dependence of $N-NO_3^-$ concentrations on S-CEC contents and UreasePot was negative, especially in adjacent stand. The dependence of $N-NH_4^+$ concentrations on Ha-CEC and UreasePot was week in the beech and mixed forest types while it was significantly positive in the spruce forest type.

Keywords $NH_4^+ \cdot NO_3^- \cdot Urease \cdot Protease \cdot European beech \cdot Norway spruce$

Introduction

Canopy gaps play key roles in forest ecosystem development and result from either natural processes or targeted forest management activities. They significantly affect habitat conditions (D'Oliveira and Ribas 2011; Pang et al. 2016) such as nutrient cycling dynamics, thermal flow, site moisture (Li et al. 2017) and light conditions (He et al. 2012; Hniličková et al. 2016). These conditions affect soil microbial activities, and gaps contribute in shaping specific microclimate environments, subsequent biological activities, biochemical processes, and energy cycling within the ecosystem (Ritter and Bjørnlund 2005).

Nitrogen availability depends largely on organic matter decomposition by micro biota (Holik et al. 2016) and the conversion of organic nitrogen to its inorganic compounds (Ritter and Bjørnlund 2005). Soil microbes are subjected to microclimate characteristics and are specific for various woody species, i.e., Norway spruce (*Picea abies* (L.) H. Karst.) and European beech (*Fagus sylvatica* L.). They are also subjected to a broad range of soil properties (Setiawan et al. 2016).

This study examines the forms and contents of soil mineral nitrogen in gaps and the relation between mineral

nitrogen and soil parameters and stand types (European beech, Norway spruce, mixed stand). It is focused on the internal system of gaps which is edaphically determined.

In terms of internally determined relationships within a forest ecosystem, the following questions have been raised: (1) How does forest type, gap size and position of sampling (centre/parental), considered as categorical variables: For-Type (beech/spruce/mix), gap size (small/big) and Position (centre/parental) affect soil parameters, including physicalchemical parameters such as pH, cation exchange capacity (CEC); chemical parameters such as organic carbon as oxidative (Cox), total nitrogen (Nt), C/N ratio, nitrate and ammonium nitrogen (N–NO₃⁻, N–NH₄⁺, respectively); and, biochemical parameters such as protease, urease (both native and potential ureolytic activity-UreaseNat and UreasePot, respectively), catalase activity, microbial carbon (C_{mic}) ?; (2) What are the relationships among soil parameters in the context of categorical variables that focus on forms of mineral nitrogen?; and, (3) Which factors from the group of categorical variables and soil parameters have the largest influence on mineral nitrogen contents in terms of its individual forms (nitrate and ammonium nitrogen)?

Materials and methods

Area description and field work

The research plots (Table 1) were established in the Training Forest Enterprise Masaryk Forest Křtiny (TFE), which is an organizational part of Mendel University in Brno in winter 2013/14. Natural conditions are a slightly undulating topography and altitudes between 520 and 570 m.a.s.l. with an annual precipitation of approximately 610 mm and annual mean temperature of 7.5 °C. The plots are on the border of the Moravian Karst and transition into the lower plateau of the Cretaceous sediments, the so called Rudice Beds.

Plots were numbered from Gap 1 to Gap 6 and situated in three mature stands (95–105 years) of different forest types including beech (ForType-Beech) (Gaps 1 + 2; European beech 100%); mixed (ForType-Mixed) (Gaps 3 + 4; European beech 50%, Norway spruce 50%); and spruce (ForType-Spruce) (Gaps 5 + 6; Norway spruce 100%).

Soil surveys and sampling were performed in autumn 2015. Soil profiles were described for each forest type by determining the soil taxonomic unit and humus form (Table 1). The six gaps (gap size-big/B, and small gaps gap size-small/S) were sampled in the centre of the plots (Position-Centre/C) and in neighbouring forests (Position-Parental/P). In each position, four representative mixed samples were collected, (each sample approximately 500 g from three sites), from the organomineral A horizon. The samples were passed through a 2 mm sieve and stored in PET bags at 5 °C.

 Table 1
 Natural conditions by ForType, gap size, woody species percentage, geology (bedrock, topsoil) and pedology (humus form, taxo-nomical unit, A horizon thickness)

Gap no.	Coord (N/E) (WGS 84)	ForType/gap size (m ²)	Species composition ^a (%)	Bedrock	Topsoil/A hor. thickness	Humus form ^b /taxonomical unit ^c
1	49°19.03118' 16°43.66622'	Beech/big (1.291)	Beech 90 spruce + larch 10	Mixed (limestone, terra fusca)	Loess loam/ 4.5 cm	Dysmull/ST-al- crn.slp.can
2	49°19.02657′ 16°43.62308′	Beech/small (315)	Beech 90 spruce + larch 10			
3	49°18.89250′ 16°43.53232′	Mixed/big (1.256)	Spruce 50, beech 30, larch + fir 20	Mixed (limestone, terra fusca)	Loess loam/ 3 cm	Dysmoder/LV- go.le.cr.can-df.slp
4	49°18.90320′ 16°43.50110′	Mixed/small (310)	Spruce 50, beech 30, larch + fir 20			
5	49°19.07415′ 16°42.54572′	Spruce/big (1.251)	Spruce 90, beech + larch 10	Mixed (kaolinized, quartzose sandstone, loess admixture)	Loess loam/ 5 cm	Mor/ST-skn-slp.rp
6	49°19.08453′ 16°42.52463′	Spruce/small (302)	Spruce 90, beech + larch 10			

^aEuropean beech (*Fagus sylvatica* L.); Norway spruce (*Picea abies* (L.) Karsten); silver fir (*Abies alba* Mill.); European larch (*Larix decidua* Mill.)

^bBrêthes et al. (1995)

^cIUSS Working Group WRB (2015)

Laboratory analyses

Ammonium and nitrate nitrogen levels were determined according to Kučera et al. (2013), i.e., N expressed in terms of the relevant form of mineral nitrogen. Soil pH was measured in suspensions of soil: water and soil: 1 M KCl at a ratio of 1:2.5 (Zbiral and Honsa 2010). Hydrolytic acidity (Ha-CEC) and base cation content (S-CEC) were assessed in sodium acetate and hydrochloric acid, respectively (Lityński et al. 1976), and used to count base saturation (BS-CEC) from S-CEC and cation exchange capacity (T-CEC). Organic carbon was assessed as oxidative carbon (Cox) by sulphochromic oxidation (Zbiral and Honsa 2010). Total nitrogen (Nt) was determined according to Zbiral and Honsa (2010). Soil catalase activity was measured manometrically using O₂ production (Gömöryova et al. 2009); soil protease activity was measured spectrophotometrically based on casein hydrolysis of the substrate, and the amount of L-tyrosine produced was measured (Rejsek et al. 2008). Urease activity was determined spectrophotometrically according to Kandeler and Gerber (1988); the amount of released ammonium nitrogen was determined after the soil samples were incubated with urea. Protease activity was measured as native; urease activity was measured as both potential and native. The methodology for protease and urease activity was adjusted (Rejsek et al. 2008), specifically, demineralised water was added instead of a buffer. The determined protease and urease activity was so-called native; enzyme activity was limited by soil pH instead of by the pH of the added buffer. The determination of microbial biomass carbon was performed using a fumigant extraction method. In the presence of strong sulphuric acid, the organic matter is oxidized and Cr(VI) is reduced to Cr(III). The loss of Cr(VI) was determined spectrophotometrically (Zbiral and Honsa 2010).

Statistical analysis

The statistical analysis was performed in R software, version 3.2.3 (R Foundation for Statistical Computing). In the regression triplet, (data, model, method), the data were observed using boxplots and an ordination method of data projection within the 'vegan' package, version 2.3-5 (Oksanen et al. 2016). To observe the next relations among variables, correlation analysis used the 'pairs' function.

Linear regression was performed using a generalized linear model (GLM) with a 'gamma' error distribution and a natural logarithm link function, where E [y|x,z] = $\exp(\alpha + \beta \cdot x + \gamma \cdot z) = \hat{y}$. The data were tested to verify the dependence of $(y \sim)$ N–NO₃⁻ and N–NH₄⁺ concentrations using continuous variables such as the soil properties from the groups of physical–chemical, chemical and biochemical, and categorical variables (i.e., 'ForType', 'Position' and 'Gap size'), tested both with and without interactions. Graphics were created using the 'ggplot2' package, version 2.1.0 (Wickham and Chang 2016). The final model was selected using Akaike's information criterion (AIC), variance inflation factors (VIF) and p value when alpha = 0.05.

Results

In terms of the parameters considered, the individual stands and their canopy gaps are soil-specific (Tables 2, 3). Overall mineral nitrogen content was highest in ForType-Spruce compared to beech and mixed forest types and had higher average contents of both nitrate N (significantly) and ammonium N (more balanced with ForType-Beech), as well as a higher proportion of mineral nitrogen N_{min} in total nitrogen Nt.

Principal Component Analysis (PCA) (Fig. 1) shows the relationships between the individual variables and ForType position in factorial plane. ForType-Beech is determined particularly by soil enzymatic activity and related physical-chemical properties. ForType-Mixed is determined by two variables: C/N ratio and T-CEC with strong positive correlation and significance for the second PCA component. ForType-Spruce is bound to the mineral nitrogen fraction, C_{ox} and C_{mic} content (Fig. 2).

Optimized model result for N-NO₃⁻ content (Table 4, Fig. 3) is a determined $N-NO_3^-$ dependency on base cation content (S-CEC) and ureolytic activity (UreasePot, UreaseNat) as continuous variables, and on ForType and Position as categorical variables. N-NO₃⁻ content is strongly negatively correlated with S-CEC and specific within trends for each stand type (see *p* values in Table 4). $N-NO_3^-$ production, (in the process of nitrification, it includes the acidification process and subsequent release of H⁺ in the soil solution), causes decreasing base cation content fixed on the soil sorption complex. This relationship is more striking in neighbouring forests; it might be caused by, among others, the absolutely higher N-NO₃ concentration values in the gap centre. The most striking negative N-NO₃⁻ content dependency on UreasePot is in ForType-Spruce.

The optimized model for N–NH₄⁺ content (Table 5, Fig. 4) resulted in finding the negative N–NH₄⁺ dependence on hydrolytic acidity (Ha-CEC) and ureolytic activity (UreasePot, UreaseNat) as a continuous variable, and on ForType and Position as categorical variables. N–NH₄⁺ concentration slightly decreased in ForType-Beech and Mixed with increasing Ha-CEC; in Spruce it is strongly positively dependent, both in the case of Ha-CEC and UreasePot (see *p* values in Table 5). In ForType-

 Table 2
 Characteristics of soil

 properties grouped in
 categorical variables

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Variable	Stat	Unit	Gap size	e(n = 8)				
			Beech		Mixed		Spruce	
			Big	Small	Big	Small	Big	Small
wH ₂ O	Mean	%w	31.56	29.35	22.44	16.51	27.62	27.13
	SD		9.73	7.00	8.13	3.52	10.33	9.76
pH/H ₂ O	Mean	-	4.45	4.38	4.03	3.96	3.45	3.61
	SD		0.28	0.25	0.51	0.53	0.06	0.11
pH/KCl	Mean	-	4.08	3.93	3.68	3.56	3.07	3.13
	SD		0.24	0.27	0.49	0.46	0.03	0.12
Ha-CEC	Mean	mmolChemeq kg ⁻¹	104.85	127.47	198.68	244.34	199.44	223.96
	SD		17.51	31.57	68.88	29.91	15.88	27.38
S-CEC	Mean	mmolChemeq kg ⁻¹	188.96	150.24	139.78	183.32	70.86	96.24
	SD		19.37	9.17	23.13	40.87	10.43	19.55
T-CEC	Mean	mmolChemeq kg ⁻¹	293.82	277.71	338.46	427.66	270.29	320.20
	SD		9.54	32.44	63.03	21.42	18.31	44.36
BS-CEC	Mean	%	64.28	54.69	42.92	42.63	26.19	29.85
	SD		6.08	6.03	10.94	8.18	3.26	2.60
Cox	Mean	%	7.53	7.21	7.12	8.28	8.19	8.73
	SD		1.61	1.32	1.06	1.60	0.99	1.60
Nt	Mean	%	0.40	0.40	0.25	0.32	0.41	0.47
	SD		0.08	0.06	0.04	0.11	0.07	0.09
C/N	Mean	-	18.79	18.01	28.93	27.08	20.39	18.99
	SD		1.20	1.31	2.82	3.99	1.30	2.55
$N-NH_4^+$	Mean	$\mu g g_{drymass}^{-1}$	13.30	12.14	11.77	8.96	16.77	21.24
	SD		4.70	2.36	4.80	1.10	13.99	4.63
$N-NO_3^-$	Mean	$\mu g g_{drymass}^{-1}$	1.68	3.01	3.13	2.60	6.69	4.72
	SD		1.04	1.55	0.67	1.53	1.54	1.31
Catalase	Mean	ml O ₂ $g_{drymass}^{-1}$ 15 min ⁻¹	61.27	46.19	25.74	32.26	39.39	33.07
	SD		19.63	12.54	6.54	12.91	17.91	10.34
Protease	Mean	μg L-tyrosine $g_{drymass}^{-1} h^{-1}$	85.20	56.83	31.07	25.45	13.81	26.81
	SD		36.43	16.31	12.76	9.05	8.14	6.12
UreasePot	Mean	μ g N–NH ₄ ⁺ g ⁻¹ _{drymass} h ⁻¹	115.48	118.00	69.08	49.18	24.32	45.53
	SD		24.14	37.22	32.61	25.65	7.95	18.20
UreaseNat	Mean	μ g N–NH ₄ ⁺ g ⁻¹ _{drymass} h ⁻¹	87.31	68.04	54.18	42.05	20.10	31.61
	SD	-	17.05	22.56	26.13	14.72	6.09	13.18
Cmic	Mean	$\mu g C g_{drymass}^{-1}$	690.07	468.00	464.99	488.05	624.88	611.18
	SD	-	166.59	61.92	86.39	90.79	118.86	94.66

Gap size wH₂O is moisture; pH/H₂O and pH/KCl are soil reaction active and exchangeable, respectively; Ha-CEC is hydrolytic acidity; S-CEC is base cation content; T-CEC is cation exchange capacity; BS-CEC is base cation content; Cox is oxidative carbon; Nt is total nitrogen; C/N is Cox: Nt ratio; N–NH₄⁺ and N–NO₃⁻ are ammonium and nitrate nitrogen, respectively; catalase, protease, UreasePot and UreaseNat are enzymatic activities; Cmic is microbial carbon)

Spruce, there is a statistically significant dependence of soil acidity, respectively concentration of proton H^+ , and $N-NH_4^+$ content, respectively ammonium nitrogen ions as acidity promoters, thus acid cations also bound in the soil sorption complex.

Discussion

The canopy gaps led to an accelerated decomposition of organic matter, as well as to mineralization leading to an increase of available nutrients which can be utilized by plants or microbial biomass (Muscolo et al. 2014).

 Table 3 Characteristics of soil properties grouped in categorical variables ForType and position

Variable	Stat	Unit	ForType	n = 16		Position	(n = 8)				
			Beech	Mixed	Spruce	Beech		Mixed		Spruce	
						Centre	Parental	Centre	Parental	Centre	Parental
wH ₂ O	Mean	% w	30.45	19.48	27.37	38.21	22.69	24.04	14.92	35.02	19.73
	SD		8.54	6.93	10.05	4.38	2.53	7.20	1.64	8.77	9.24
pH/H ₂ O	Mean	-	4.41	3.99	3.53	4.43	4.40	4.11	3.87	3.50	3.56
	SD		0.27	0.52	0.12	0.24	0.29	0.42	0.58	0.04	0.43
pH/KCl	Mean	-	4.01	3.62	3.10	4.07	3.95	3.63	3.62	3.06	3.14
	SD		0.27	0.48	0.09	0.20	0.31	0.49	0.46	0.05	0.48
Ha-CEC	Mean	mmolChemeq kg ⁻¹	116.16	221.51	211.70	98.56	133.76	201.71	241.32	204.06	219.34
	SD		27.92	57.80	25.52	12.73	27.89	71.77	27.33	12.64	67.94
S-CEC	Mean	mmolChemeq kg ⁻¹	169.60	161.55	83.55	178.63	160.57	145.60	177.50	77.27	89.83
	SD		24.58	39.71	20.16	26.12	19.07	13.72	49.56	6.63	22.52
T-CEC	Mean	mmolChemeq kg ⁻¹	285.76	383.06	295.25	277.20	294.33	347.31	418.81	281.33	309.17
	SD		25.23	64.84	42.12	17.56	28.60	72.26	25.13	8.07	56.76
BS-CEC	Mean	%	59.49	42.77	28.02	64.16	54.81	43.69	41.86	27.52	28.53
	SD		7.72	9.66	3.47	5.95	6.34	9.49	9.75	2.70	12.13
Cox	Mean	%	7.37	7.70	8.46	7.63	7.11	7.29	8.11	8.95	7.97
	SD		1.48	1.48	1.36	1.55	1.36	0.73	1.87	1.48	1.13
Nt	Mean	%	0.40	0.28	0.44	0.41	0.39	0.24	0.33	0.44	0.44
	SD		0.08	0.09	0.08	0.08	0.07	0.02	0.11	0.08	0.10
C/N	Mean	-	18.40	28.00	19.69	18.62	18.18	30.28	25.72	20.73	18.64
	SD		1.31	3.57	2.14	1.41	1.18	2.28	3.15	1.58	6.74
N-NH4+	Mean	$\mu g g_{drymass}^{-1}$	12.72	10.36	19.00	15.53	9.91	11.11	9.62	21.05	16.96
	SD	·	3.77	3.76	10.66	3.36	1.13	4.86	1.86	11.94	2.70
$N-NO_3^-$	Mean	$\mu g g_{drymass}^{-1}$	2.34	2.87	5.71	3.36	1.33	3.71	2.02	5.50	5.91
	SD	·	1.47	1.21	1.73	1.42	0.54	0.69	1.01	1.77	1.21
Catalase	Mean	ml O ₂ $g_{drymass}^{-1}$ 15 min ⁻¹	53.73	29.00	36.23	64.95	42.51	30.82	27.18	41.02	31.44
	SD		18.12	10.74	14.96	18.87	6.98	12.31	8.52	16.91	17.30
Protease	Mean	μg L-tyrosine $g_{drymass}^{-1} h^{-1}$	71.02	28.26	20.31	87.17	54.86	29.45	27.06	19.71	20.91
	SD		31.59	11.41	9.71	32.07	21.10	13.30	8.98	11.68	21.93
UreasePot	Mean	μ g N–NH ₄ ⁺ g ⁻¹ _{drymass} h ⁻¹	116.74	59.13	34.93	116.55	116.93	55.95	62.32	30.16	39.69
	SD		31.40	30.98	17.60	23.08	37.93	34.36	26.81	16.76	48.24
UreaseNat	Mean	$\mu g \text{ N-NH}_4^+ g_{drymass}^{-1} h^{-1}$	77.68	48.11	25.86	92.57	62.78	51.02	45.20	23.44	28.28
	SD	2	22.19	22.05	11.77	14.77	17.98	25.67	17.23	10.17	24.80
Cmic	Mean	$\mu g C g_{drymass}^{-1}$	579.04	476.52	618.03	628.09	529.98	477.21	475.83	649.96	586.10
	SD	·	167.70	89.36	107.66	210.31	84.86	87.42	91.26	105.63	80.11

wH₂O is moisture; pH/H₂O and pH/KCl are soil reaction active and exchangeable, respectively; Ha-CEC is hydrolytic acidity; S-CEC is base cation content; T-CEC is cation exchange capacity; BS-CEC is base cation content; Cox is oxidative carbon; Nt is total nitrogen; C/N is Cox: Nt ratio; N–NH₄⁺ and N–NO₃⁻ are ammonium and nitrate nitrogen, respectively; catalase, protease, UreasePot and UreaseNat are enzymatic activities; Cmic is microbial carbon)

The assessment of causes in changes or differences in $N-NO_3^-$ and $N-NH_4^+$ content is subject to a comprehensive approach considering more aspects from the soil properties categories but also within the forest stand context and litter characteristics of the relevant ForType.

Urease, as a constitutive enzyme (Gobat et al. 2010), is a result of prolonged soil chemistry when, under long-term nitrate presence, ureolytic activity involved in N uptake in ammonium form is reduced. If nitrogen is present in the soil, albeit in the form of nitrate, soil biota are not "driven" to energy-consuming biochemical processes of urea



Fig. 1 Ordination plot of soil parameters in projection together with ForType

Fig. 2 Results of correlation analysis of soil parameters: with correlation coefficient below diagonal (higher values of correlation coefficient correspond with larger numbers), and histograms of data distribution on the diagonal (pH/KCl is soil reaction exchangeable; Ha-CEC is hydrolytic acidity; S-CEC is base cation content; BS-CEC is base saturation; N-NH4⁺ and N-NO₃⁻ are ammonium and nitrate nitrogen, respectively; catalase, protease, UreasePot are enzymatic activity)

decomposition (Xu et al. 2015). This is more striking in parental treatment than in the gap centres. Increasing the availability of NH_4^+ can suppress nitrite reductase synthesis or inhibit NO_3^- transport into cells (Hart et al. 1994); therefore, NO_3^- concentration increases in the soil.

In several studies, enzymatic activities showed immediate changes in soil properties as a result of felling, as well as from natural regeneration processes (Alvear et al. 2005; Muscolo et al. 2015; Settineri et al. 2018). The reaction of microbial activity results from disturbances, including changes in abiotic conditions, namely temperature and moisture and consequently, changes in form and amount of available nitrogen and in the C/N ratio (Armas et al. 2009). The influence of abiotic conditions on microbial activities were described in detail by Hortal et al. (2015) who reported an increase of enzyme activities (dehydrogenase, β -glucosidase, urease, phosphatase), and also changes in microbial community composition.

Our results have shown increased enzymatic activities in the beech and mixed forest soils (with the exception of UreasePot). Similar results were described by Muscolo



Table 4Selection of final model exiteeither with or without interaction (ca	plaining the determination of ttegorical)	f nitrate nitrogen (N–NC	3 ⁻) depending on independ	lent variables S-CEC, U	reasePot (continuous) and I	² orType and Position
Model ($y = N-NO_3^-$)/coefficients	S-CEC + UreaseNat + Fc	$rType \times Position$	S-CEC + UreasePot + Fe	orType + Position	S-CEC + UreasePot + F	orType × Position
	<i>p</i> value	vif	<i>p</i> value	vif	<i>p</i> value	vif
Intercept	< 0.001	I	< 0.001	I	< 0.001	I
S-CEC	< 0.001	3.78	< 0.001	3.00	< 0.001	3.43
UreaseNat	0.1624	3.30	I	I	I	I
UreasePot	I	I	0.0094	2.77	< 0.001	2.78
ForType-Mixed	0.1111	I	0.3769	I	0.0077	I
ForType-Spruce	0.0101	I	0.8036	I	< 0.001	I
Position—P	< 0.001	I	< 0.001	I	< 0.001	I
ForTypeMixed:PositionP	0.0037	I	I	I	0.0031	I
ForTypeSpruce:PositionP	< 0.001	I	I	I	< 0.001	I
ForType	I	12.43	I	5.05	I	11.70
Position	Ι	3.30	Ι	1.03	I	3.12
ForType:Position	Ι	10.24	Ι	I	I	9.19
AIC/R ²	145.4322/	0.78	160.76/	0.67	133.35/	0.83





et al. (2007a) in European beech and silver fir stands in middle gap sizes (410 m²), and by Muscolo et al. (2007b) for black pine stands with small gap sizes (380 m²) as well as large gap sizes (1520 m²). Settineri et al. (2018) also observed increased protease and catalase activities in comparison with soils in clearcuts with parental stand. An exception was acid phosphatase and cellobiohydrolase activities (Mayer et al. 2017) in large gaps (1000 m²) of a mixed forest type where the activities decreased.

Gap size has an immediate effect on the rate of change in microclimatic and edaphic conditions (Zhang and Zak 1995; Ferreira De Lima 2005; Gálhidy et al. 2006; He et al. 2015). The selection system is an optimal forest management system in terms of species diversity and microclimate stability (Brunet et al. 2010). However, in terms of harvest practice, it is not always possible to apply this method; the limit of 15–30 trees (Parsons et al. 1994) is considered a cutting-area size. In our case, this means that no significant nitrogen losses occur: the gap sizes fluctuate on a range of small and medium size classes in the case of small gaps, and big size class in case of the big gaps. Only $N-NO_3^-$ in relation to gap size shows irregular fluctuations and dynamics with no obvious trend.

Changes in nitrate concentrations in soils do not necessarily have to be related to the selected management system, to gap size, or to subsequent changes in microclimatic conditions (Prescott et al. 2003). Changes may be due to the nature of the litter that results from the stand type, or to the harvest itself.

The differences in treatment position correspond to the biological aspects of the tree layer and its effect on the soil water regime. However, some authors report different results, e.g., gaps at the micro-site scale show a determined disturbance and a biological water pump deprivation (Aragão 2012), and these constitute the initiation factor or subsequent changes in biological and enzymatic activities, humus ratios, soil chemistry and other parameters, including water regime and soil aeration (Guntinas et al.

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Model (y = $N-NH_4^+)/coefficients$	Ha-CEC + Urease	Nat + ForType × Position	Ha-CEC + UreaseP	ot + ForType + Position	Ha-CEC + UreasePot	$(+ For Type \times Position)$
	p value	vif	<i>p</i> value	vif	p value	vif
Intercept	< 0.001	1	< 0.001	I	< 0.001	I
Ha-CEC	0.0194	3.23	0.0111	3.17	0.0106	3.20
UreaseNat	0.0262	2.93	I	I	I	I
UreasePot	I	I	0.0059	2.76	0.0053	2.77
ForType-mixed	0.0144	I	0.0734	I	0.0116	I
ForType-spruce	0.2405	I	0.0211	I	0.2727	I
Position—P	0.0767	I	0.0108	I	0.0023	I
ForTypeMixed:PositionP	0.2205	I	I	I	0.0577	I
ForTypeSpruce:PositionP	0.4432	1	I	I	0.0931	I
ForType	I	13.40	I	3.86	I	10.52
Position	I	3.78	I	1.19	I	3.48
ForType:Position	I	10.55	I	I	I	9.40
AIC/R ²	237.14/	0.57	235.38	0.55	234.26/	0.60
The correlation matrix (Fig. 2) indi- which is correlated only to N–NH ₄ ⁻ with pH/KCl, S-CEC and BS-CEC properties	cates a frequently stati ⁺ . Ammonium N gene and a positive correla	stically significant correlation rally appears to not be predeten tion of borderline significance	correlation coefficient mined by monitored pa with Ha-CEC. Enzyme	R^2 at significance level $\alpha =$ rameters, in contrast to nitra sactivities are frequently in	0.05; R ² crit = 0.2377), an tte N which shows a signi n strong mutual correlatio	nong others in case of Ha, ficant negative correlation n with physical-chemical

Table 5 Selection of final model explaining determination of ammonium nitrogen (N–NH₄⁺) on independent variables S-CEC, UreasePot (continous) and ForType and Position, either with or without interaction (categorical)





2012; Olajuyigbe et al. 2012). Thus, changes in the significance of nitrogen transformations can be expected in gaps, especially in terms of significantly increased moisture (cf., Tables 2, 3), when both nitrates are transferred to the ammonium form in the process of dissimilatory nitrate reduction to ammonium or when, during the denitrification process, they are converted to a gaseous form (van Groenigen et al. 2015).

Conclusion

The concentration of N mineral forms was most affected by three factors from the group of soil parameters, (Ha-CEC and UreasePot for N–NH₄⁺; S-CEC and UreasePot for N– NO₃⁻), and two factors from the group of categorical variables (ForType and Position for both N–NH₄⁺ and N– NO₃⁻). In our study, gap size was not a significant factor. The N–NO₃⁻ relationship with the base cation content was more significant in terms of stand type when using the viewpoint of Position-Centre versus Parental. Thus, N–NO₃⁻ was determined more by base cation content and ureolytic activity with respect to stand type, and dependence decreased in the gap Centre. Equally, the N–NO₃⁻ linkage to ureolytic activity was reduced in the gap. As for stand type, N–NO₃⁻ concentration was most clearly determined by urease in the spruce stand.

 $N-NH_4^+$ showed different trends in individual stand types; in beech and mixed stands, $N-NH_4^+$ dependence on hydrolytic activity and potential urease activity was very weak, and in the spruce stand, it showed a significantly strong dependence. A significant negative dependence of nitrate nitrogen concentration on ureolytic activity may indicate a reduced need to stimulate the energy-demanding biochemical process of urea decomposition. Acknowledgements This work was supported by the Faculty of Forestry and Wood Technology, Mendel University, Brno (IGA Mendelu in Brno "GAPS" 84/2013; IGA Mendelu in Brno LDF_PSV_2017006), and the Ministry of Agriculture of the Czech Republic (QJ1320050) and (MZe RO418).

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