

Bioaccumulation of Macronutrients in the Herbaceous Plants of Mid-forest Spring Niches

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Abstract

The results of preliminary studies on nitrogen, phosphorus, potassium, magnesium and calcium bioaccumulation in the herbaceous plants of mid-forest spring niches are presented in this paper. Studies were conducted in the upper course of the Kamienna Creek, a left bank tributary of the Słupia River located in the area of the Forest Division Leśny Dwór (northern Poland). Groundwater, soil and plant samples were collected from three stands distributed within the headwater bog with Rheic Sapric Histosols covered by age-varied black alder. Based on N, P, K, Ca and Mg contents in the plants, groundwater and soils, bioaccumulation factors of these elements in plants were calculated in relation to groundwater (BF_w) and soil (BF_g). The investigated soils were characterized by strong decomposition of peat mass, acidic pH, high abundance in total nitrogen and low in phosphorus, potassium, magnesium and calcium. The chemical composition of the plants suggests an adequate nutrient supply, especially in the case of potassium, which accumulated in much higher amounts than physiological requirements. The investigated plant species were characterized by varied bioaccumulation factors of the studied elements. The highest bioaccumulation of N and Ca was noticed for *Urtica dioica*, P for *Urtica dioica* and *Solanum dulcamara*, Mg for *Caltha palustris* and *Urtica dioica* and K for *Geranium robertianum*. The shoots of *Solanum dulcamara* (N, P, K) and *Urtica dioica* (N, P, K, Ca, Mg) were characterized by a diversity of bioaccumulation factors. Our data confirm strong interactions between water, soil and plants as the main structural and functional components of headwater areas.

Key words Bioaccumulation, nutrients, herbaceous plants, headwater areas, Histosols, groundwater.

Introduction

Seasonal changes in the environment as a consequence of the seasons of the year and diverse demands of plants for macro- and microelements during their life cycle determine the dynamic character of the plant-environment relationship (Brinson 1990). The chemical composition of plants depends on many factors, particularly plant physiology, age and site conditions, which determine the bioavailability of individual elements. The bioavailability of nutrients in soil affects the growth and development of the plant cover (Pugnaire 2001) and depends on many factors, including groundwater level, which is of vital importance. Heavy moistening of soils, which is characteristic for headwater areas, affects the bioaccumulation of macroelements. Headwater areas constitute transitional zones between the underground and the surface part of water circulation in river catchments (Jekatierinczuk-Rudczyk 2007). They are open systems in the cycle of matter and energy, whose function is conditioned by the volume and chemical composition of

supplying waters and their constant flow on the surface and in the soil. Supplying waters bring, on the one hand, a specific load of ions to the ecosystem, but can wash out certain soil components on the other. Interactions between water, soils and plants in the ecosystems of headwater areas are very tight and multidirectional (Karlsson et al. 2005). Litterfall is a vital source of nutritional components and is one of the main mechanisms of their development (Astel et al. 2009; Jonczak et al. 2016). Nutrient elements gradually released during the process of decomposition are included again into biological turnover litterfall, and small quantities are eluted outside the reach of root systems (Jordan et al. 1989). Plants, as an integral part of riparian ecosystems, play an important role in filtering and retarding nutrients and other pollutants (Tanner 1996, Tabacchi et al. 2000, Broadmeadow and Nisbet 2004, Schoonover et al. 2005, Li et al. 2008, Dosskey et al. 2010, Vidon 2010, Yu et al. 2014a, Roberts et al. 2012). By intercepting and storing macro-components, plants play an important role in the protection of river waters against eutrophication (Hefting

et al. 2005, Hazlett et al. 2008, Lee et al. 2009, Yu et al. 2014b). The greater the species diversity and density of vegetation in the areas of headwaters, the more effective the protection against the penetration of pollutants into the lower courses of the river (Lyon and Segars 1998). Previous research studies done in headwater niches indicate considerable diversity of plant species (Decamps et al. 2004, Calçada et al. 2015, Pielech et al. 2015), which are characterized by a relatively high capacity of phytoaccumulation of macro- and microelements (Maine et al. 2006, Gottschall et al. 2007, Horska-Schwarz and Spalek 2008, Yu et al. 2014b). Nutrient dynamics of headwater riparian forests under natural conditions are complicated by the variety of riparian biotopes; thus, long-term studies are needed to obtain a representative view of nutrient accumulation dynamics. Studies on the accumulative properties of plants are vital due to the possible use of some species in the processes of phytoremediation (Xue et al. 2013, Galal and Shehata 2015). Despite this accumulated information, the influence of the chemical properties of soil and groundwater on accumulation processes in riparian plants remains poorly understood (Tabacchi et al. 2000, Zang et al. 2010).

The aims of this study were the following: (1) the identification of factors determining N, P, K, Mg and Ca contents, and (2) a comparison of bioaccumulation factors of macroelements in selected species of herbaceous plants occurring within mid-forest spring niches in the valley of the Kamienna Creek in northern Poland.

Materials and Methods

Stand characteristics

The research was done in the upper course of the Kamienna Creek, a left bank tributary of the Słupia River situated in the northern part of Poland within Leśny Dwór Forest Inspectorate area (54°19'N; 17°10'E). The area receives an average annual precipitation of about 770 mm and an average annual air temperature of 7.6°C. The area of the catchment of the Kamienna Creek is nearly wholly covered with forests with a spatially diverse species composition, with a predominance of beech, pine and spruce in its plateau part and black alder (*Alnus glutinosa* Gaertn.) in the valley bottom. This research was done in the headwater area covered with a 40- to 86-year-old stand. The tree stand grew over a domed moor made up of alder peat with layers of alder-sedge peat, cut by headwater streams. Within the area of the headwater riparian forest, a large number of species was discovered, which are generally considered to be the outstanding species of the assemblage: *Galium palustre* L., *Lycopus europaeus* L., *Lythrum salicaria* L., *Lysimachia vulgaris* L. and *Solanum dulcamara* L. Locally, there often appeared *Cardamine amara* L., *Chrysosplenium alternifolium* L., *Carex paniculata* L. and *Scirpus sylvaticus* L. (Sobisz et al. 2016).

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Soil sampling and analysis

Soils were described and sampled at the beginning of the experiment. Samples were collected from every 10 cm until the mineral bed. The methods of soil sampling and analysis were described in the paper by Jonczak et al. (2014). The contents of P, K, Mg and Ca were analyzed after sample digestion in 65% HNO₃ and 30% H₂O₂. The concentration of P in the solution was determined calorimetrically with ammonium molybdate and the remaining elements by microwave plasma atomic emission spectrometry (Agilent 4100 MP-AES).

Plant sampling and analysis

Plants were examined three times during the vegetation season (May, July and September) in 2012-2014. For chemical examination, selected plant species were chosen characterized by the highest frequency and density during the entire vegetation season (Sobisz et al. 2016). At each stand, samples of four plant species were taken. A sample comprised over-ground sprouts originating from several specimens of a given species taken within the area of one stand. In total, 108 samples were analyzed. After their delivery to the laboratory, the plant samples were washed in distilled water, dried at 65°C and homogenized in a laboratory grinder (IKA A11, Germany). Before analysis, the plant material was kept in tightly closed polyethylene bags.

In order to determine nitrogen and phosphorus, the samples were digested in a mixture of 98% H₂SO₄ and 30% H₂O₂ (1:1 by volume). Total content of nitrogen was determined by the Kjeldahl method (Büchi K-350, Switzerland), and that of phosphorus by the molybdate method (UV-VIS spectrophotometer, Hitachi U-5100, Japan). The concentrations of K, Mg and Ca in plants was determined by atomic absorption spectrometry (Analyst 300, Perkin Elmer, USA), after digestion in a mixture of 65% HNO₃ and 30% H₂O₂ (by volume 1:1). Merck standard solutions (1000 ppm) of these elements were used for the analysis. All the analyses were performed in three replicates. The analytical quality of the results was checked against the CRM 060 (aquatic plant) reference material, which was provided by the European Commission Institute for Reference Materials and Measurements. The recoveries were considered to be satisfactory if the results of the analyzed elements were within the confidence intervals of the certified values.

Water sampling and analysis

Within the patches, where the plants were collected, piezometers were installed, from which (at a depth of 30 cm) samples of groundwater were taken in May, July and September 2012-2014. In these samples, the pH was measured by a potentiometer (Elmetron CPI 551, Poland) and electrolytic conductivity by the conductometric method (Elmetron CC 315, Poland). The content of NH_4^+ , K^+ , Mg^{2+} , Ca^{2+} , NO_3^- and PO_4^{3-} ions was analyzed by ion chromatography (881 Compact IC pro, Metrohm, USA) after sample filtration using 0.2 mm net-mesh. Analysis quality was controlled using certified reference materials (Mutlielements Ion Chromatography, Certified 89 866-50ML-F, Fluka, France).

Data elaboration

The content of the macroelements in soils, groundwater and plants was tested by means of the Shapiro-Wilk test. The results referring to the soil, water and plant properties are specified in the tables, providing average, minimum and maximum values as well as standard deviations and variability coefficients. The obtained data were used to determine the Spearman correlation coefficients. To identify the factors determining the content of macroelements in sprouts of the examined plant species, factor analysis was applied (Principal Components Analysis, PCA). By PCA, two independent factors were determined, which explain the variability in the amounts of N, P, K, Mg and Ca in the sprouts of plants from a headwater riparian forest. Hierarchical cluster analysis (Ward's method) was used to classify plant species based on the macroelement contents in shoots.

On a basis of the results, the value of bioaccumulation factors of macroelements in relation to the content of the respective components in the soil (BF_s) (Harasimiuk 2006) and in the groundwater (BF_w) were calculated (Zayed et al. 1998). The variability in bioaccumulation factors during the period of the study is presented in the figures. The statistical significance of diversity among the examined plant species with regards to BF_s and BF_w was tested by the application of a non-parametric Kruskal-Wallis test. For evaluation of the results, a software package Statistica 7.1 was used.

Results

The physicochemical properties of soils

Sapric Histosols, which appear within the area of this headwater riparian forest, presented spatially varied thickness, which did not exceed 1 m. The soils were characterized by the content of organic matter at the level up to about 80% with maximum in topsoil and

pH from weakly acidic to acidic (Table 1). These soils are abundant in nitrogen, relatively abundant in calcium and poor in phosphorus, potassium and magnesium (Jonczak et al. 2015b). They represent a series of specific characteristics resulting from their function in the transitional zone between underground and surficial parts of water circulation in a river catchment. In these soils, there is a continuous, concentrated flow of groundwater towards the river, yielding characteristic vertical gradients in the concentration of various components (Jonczak et al. 2015b). These soils are regularly supplied with elements from litterfall (Jonczak et al. 2016), which nearly completely decay within the first year. The properties of the litterfall were described in detail by Jonczak et al. (2015a, 2016).

Table 1. Physicochemical properties of soil from headwater riparian forest

Depth [cm]	OM %	pH (KCl)	pH (H ₂ O)	N	P	K	Mg	Ca	
				g/kg					
Location 1									
0-10	72.5	6.1	5.5	32.99	1.21	0.87	1.10	22.28	
10-20	74.1	5.9	5.4	33.59	1.16	0.52	0.94	21.44	
20-30	75.3	5.8	5.3	32.41	0.70	0.36	0.93	23.97	
30-40	70.6	5.8	5.3	30.12	0.62	0.29	0.92	24.07	
Mean	73.1	5.9*	5.4*	32.27	0.92	0.51	0.97	22.94	
Location 2									
0-10	81.1	6.5	5.9	32.93	1.02	0.55	1.13	23.55	
10-20	80.4	6.4	5.8	31.95	0.78	0.35	1.11	24.19	
20-30	81.4	6.5	6.1	30.51	0.71	0.17	1.06	25.23	
30-40	79.5	6.5	5.9	29.75	0.60	0.16	1.04	26.53	
Mean	80.6	6.5*	5.9*	31.28	0.78	0.31	1.08	24.87	
Location 3									
0-10	80.5	5.9	5.2	29.74	1.56	0.51	1.01	20.17	
10-20	82.9	5.6	4.9	29.31	0.85	0.32	0.79	17.37	
20-30	79.5	5.6	5.1	33.19	0.77	0.29	0.84	18.83	
30-40	72.2	5.9	5.2	28.57	0.68	0.30	0.88	19.52	
Mean	78.8	5.8*	5.1*	30.20	0.96	0.36	0.88	18.97	

Note – *median

The physicochemical properties of groundwater

The groundwater within the examined headwater area was found to have a neutral or weakly alkaline pH, and the electrolytic conductivity (EC) remained on average from 224 to 255 $\mu\text{S}/\text{cm}^3$ showing variability within vegetation seasons (Table 2). The groundwater was characterized by a relatively low content of macroelements, which may be connected with a low level of peat mass decomposition (Jonczak and Parzych 2016), intensive uptake of nutritional components by plants and, in the bottom part of the soils, with continuous flushing of soils by water flowing towards the river (Jonczak et al. 2015b). Filtering rainwater also plays a role in the balance of nutritional ingredients (Jekatierynczuk-Rudczyk 2007, Parzych 2011). The variable concentration of biogenic components in groundwater leads to variable levels of electrolytic conductivity and pH. The concentrations of NH_4^+ , NO_3^- , K^+ , Mg^{2+} and Ca^{2+} ions showed variability in the

Table 2. Physico-chemical properties of groundwater in headwater riparian forest in 0.30 m depth in the years 2012-2014

Parameter	Location 1			Location 2			Location 3		
	Mean ± SD	Min-Max	CV, %	Mean ± SD	Min-Max	CV, %	Mean ± SD	Min-Max	CV, %
pH	7.24*±0.50	6.83-8.19	6	7.43*±0.27	6.94-7.84	4	7.54*±0.22	7.22-7.86	3
EC, µS/cm ³	231±75.90	158.00-343.00	33	255±52	211-380	21	224±28.95	187-268	13
NH ₄ ⁺ , mg/dm ³	1.33±1.30	0.00- 3.79	99	0.26±0.28	0.00-0.75	108	1.33±1.70	0.00-4.20	126
K ⁺ , mg/dm ³	2.37±1.70	0.92-6.28	73	1.32±0.41	0.74-2.04	31	1.64±0.60	0.93-2.50	35
Mg ²⁺ , mg/dm ³	2.29±1.80	0.00-5.55	81	2.93±2.15	0.51-5.79	74	2.34±1.90	0.22-5.77	85
Ca ²⁺ , mg/dm ³	39.1±13.1	26.36-67.28	33	55.26±9.70	40.60-71.89	18	53.91±11.22	39.82-71.43	21
NO ₃ ⁻ , mg/dm ³	2.07±2.10	0.06-6.67	99	1.12±0.96	0.17-3.35	86	1.67±1.43	0.38-4.07	86
PO ₄ ³⁻ , mg/dm ³	1.48±0.63	0.78-2.52	43	0.93±1.25	0.00-3.48	135	0.28±0.40	0.00-0.98	141

Notes: *median, EC – electrical conductivity, SD – standard deviation, CV – coefficient of variation

period of study, i.e. from 21% to 141% (Table 3, 4). The highest variability was found for PO₄³⁻, NH₄⁺ and NO₃⁻. In May, a decrease in the pH of groundwater was observed, resulting from intensive intake of alkaline cations (NH₄⁺, K⁺, Ca²⁺ and Mg²⁺) by plants. The reaction of the groundwater influenced the selectivity of plants as to the forms of nitrogen taken in. With slightly alkaline and neutral groundwater, many plant species show much higher demand for NO₃⁻ than NH₄⁺ (Britto and Kronzucke 2002). A reduced concentration of nitrates and phosphates in riparian forest groundwater has been demonstrated by Groffman et al. (1992) and Parzych (2011) in a pine forest.

demands (Table 3). This high accumulation of potassium by plants was also observed in studies by Krzywy (2007), Horska-Schwarz and Spałek (2008) and Parzych et al. (2015). A good supply of herbaceous plants with macronutrients is closely dependent on soil abundance, and especially on the bioavailability of these elements (Pugnaire 2001). High concentrations of macrocomponents in aboveground sprouts are fully substantiated due to the processes of photosynthesis, which take place in them (Sharma et al. 2006). The highest quantities of N, P, Mg and Ca were accumulated by sprouts of *Urtica dioica*, and K was the highest in *Geranium robertianum* and *Scirpus sylvaticus* (Table 3). The content of macrocomponents was arranged into decreasing series depending on species: K>N>Ca>Mg>P (*Carex echinata*, *Gymnocarpium dryopteris*, *Mentha aquatica*, *Solanum dulcamara*, *Valeriana officinalis*), K>Ca>N>Mg>P (*Ajuga reptans*, *Caltha palustris*, *Geranium robertianum*), Ca>K>N>Mg>P (*Urtica dioica*) and K>N>Ca>P>Mg

Table 3. The average content of macronutrients in the shoots of plants in headwater riparian forest (2012-2014)

Species	Mean (mg/kg) ± SD (CV, %)				
	N	P	K	Mg	Ca
Location 1					
<i>Ajuga reptans</i> L.	14786 ± 2758 (18.7)	2334 ± 570 (24.4)	22393 ± 3099 (13.8)	2722 ± 474 (17.4)	17874 ± 5472 (30.6)
<i>Gymnocarpium dryopteris</i> (L.) Newman	14432 ± 2551 (16.5)	2721 ± 837 (23.4)	22994 ± 380 (16.6)	3305 ± 477 (14.4)	8300 ± 4897 (59.0)
<i>Scirpus sylvaticus</i> L.	14024 ± 2022 (14.4)	2915 ± 799 (27.4)	28070 ± 5589 (19.9)	2495 ± 868 (34.8)	5818 ± 4390 (75.4)
<i>Urtica dioica</i> L.	18243 ± 3053 (16.7)	3803 ± 837 (22.0)	21805 ± 3146 (14.4)	5127 ± 581 (11.3)	29018 ± 8144 (28.1)
Location 2					
<i>Caltha palustris</i> L.	15293 ± 2895 (18.9)	2282 ± 561 (24.6)	27880 ± 7701 (27.6)	4965 ± 602 (12.1)	18863 ± 5157 (27.3)
<i>Geranium robertianum</i> L.	15755 ± 2250 (14.3)	2339 ± 353 (15.1)	29699 ± 7518 (25.3)	3243 ± 439 (13.5)	17770 ± 5412 (30.4)
<i>Solanum dulcamara</i> L.	17897 ± 4331 (24.2)	2582 ± 425 (16.4)	25242 ± 5077 (19.9)	3272 ± 730 (22.3)	10809 ± 2733 (25.3)
<i>Urtica dioica</i> L.	19047 ± 2805 (14.7)	3221 ± 936 (29.1)	24051 ± 5446 (22.6)	5116 ± 717 (14.0)	28555 ± 9513 (33.3)
Location 3					
<i>Carex echinata</i> Murray	12473 ± 2195 (17.6)	2157 ± 326 (15.1)	18289 ± 4315 (23.6)	3546 ± 850 (23.9)	6688 ± 3308 (49.5)
<i>Mentha aquatica</i> L.	14938 ± 2901 (19.4)	1908 ± 280 (14.7)	20694 ± 7708 (37.2)	3413 ± 655 (19.2)	13174 ± 1869 (14.2)
<i>Solanum dulcamara</i> L.	16984 ± 6931 (40.8)	2213 ± 570 (25.8)	26429 ± 3984 (15.1)	2869 ± 525 (18.3)	9734 ± 1989 (20.4)
<i>Valeriana officinalis</i> L.	12810 ± 437 (3.4)	1877 ± 345 (18.4)	22870 ± 951 (4.2)	2499 ± 1069(42.8)	9461 ± 2943 (31.1)
Minimum	12473	1877	18289	2495	5818
Maximum	19047	3803	29699	5127	29018
Natural content in plants	13000-31000 ¹	1000-4000 ¹	2000-18000 ¹	1000-3000 ²	1000-33000 ¹

Notes: ¹Ostrowska and Porębska 2002, ²Markert B. (1992)

Plants chemistry

Macronutrients play a vital role in many biological processes by conditioning the growth and development of plants. The examined plant species were characterized by sufficient concentrations of N, P, K, Mg and Ca, while K was accumulated in substantially higher quantities than required by the physiological

icus (Table 3). The content of macrocomponents was arranged into decreasing series depending on species: K>N>Ca>Mg>P (*Carex echinata*, *Gymnocarpium dryopteris*, *Mentha aquatica*, *Solanum dulcamara*, *Valeriana officinalis*), K>Ca>N>Mg>P (*Ajuga reptans*, *Caltha palustris*, *Geranium robertianum*), Ca>K>N>Mg>P (*Urtica dioica*) and K>N>Ca>P>Mg

(*Scirpus sylvaticus*). From among tested species, the shoots of *U. dioica* were characterized by the highest calcium content, which finds confirmation in the literature (Ştef et al. 2010, Gjorgieva et al. 2011). The contents of the examined macrocomponents in the sprouts of herbaceous plants of headwater niches presented substantial similarity to accumulative properties both of water (Parzych et al. 2015) and ground plants (Krzywy 2007, Parzych 2010, 2015).

Uptake of nutritional components by plants from soil and groundwater is, to a large extent, conditioned by factors dependent on plants, such as species, age, developmental stage as well as ion interactions of synergic and antagonistic character (Kabata-Pendias and Pendias 1999). The soil reaction is the main factor that determined the availability of macrocomponents. Nitrogen is most easily taken in by plants at pH 6.0-8.0, phosphorus at pH 6.5-8.0, potassium at pH 6.0-10.0 and calcium and magnesium at pH 6.5-8.5. The results (Tables 1 and 2) indicate that the pH of soil and groundwater in the examined environmental niches positively influenced the uptake of macrocomponents by plants (Table 3). This was also confirmed by many statistically significant correlation coefficients, along with the increase in the pH of groundwater, increase in concentration P in shoots (*C. echinata*), K in shoots (*S. sylvaticus*, *S. dulcamara*, *V. officinalis*), Mg in shoots (*A. reptans*, *C. palustris*, *C. echinata*, *G. dryopteris*, *V. officinalis*) and Ca in shoots (*C. echinata*) (Table 4).

Species diversity conditions variable demand for nutritional components (Broadmeadow and Nisbet 2004). Plants take in nitrogen as NH_4^+ and NO_3^- (Zang et al. 2008, Parzych 2011). The uptake dynamics of these ions depends on the species, reaction and soil moisture (Parzych 2010). Plants, which prefer acidic soils, are more likely to take up ions of NH_4^+ than NO_3^- . Changes in the soil reaction from acid to alkaline decrease the intensity of the intake of ions NH_4^+ to the benefit of ions of NO_3^- (Krzywy 2007). Statistically significant correlation coefficients between the concentration of NO_3^- in groundwater and the content of N in plant sprouts were found in the case of *C. palustris*, *G. dryopteris*, *U. dioica* and *V. officinalis* (Table 4). However, ions of NH_4^+ were easily taken in by the sprouts of *G. robertianum*, *M. aquatica*, *S. dulcamara* and *V. officinalis*. Of among examined species, only in the case of *V. officinalis* were statistically significant correlation coefficients found between the concentration of N in the shoots and the concentration of ions of NH_4^+ and NO_3^- in the groundwater. These data indicate that this species takes up nitrogen in the form of both cations and anions.

Ammonification and nitrification processes are phenomena with a positive impact on plants, since the

forms of nitrogen appearing as a consequence are easily assimilated. Ions of NH_4^+ , which have not been taken up, may, by the effect of exchange sorption, move to the sorption complex of the soil, and ions of NO_3^- , due to their high mobility, are easily washed out to deeper layers of the soil profile (Krzywy 2007, Parzych 2011). Ions of PO_4^{3-} (*G. robertianum*, *V. officinalis*), K^+ (*S. sylvaticus*, *V. officinalis*) and Ca^{2+} (*C. echinata*) in groundwater have a positive impact on nitrogen uptake by plants (Table 4).

Phosphorus uptake intensity from the soil solution is strongly affected by pH and moisture. Slightly acidic to neutral pH (pH_{KCl} 6-7) provides the optimum conditions for PO_4^{3-} uptake (Parzych 2011). High saturation of soils with water, which is typical for headwater areas, contributes to the maximum phosphorus uptake by plants (Krzywy 2007). Assimilation of phosphorus by plants depends, however, on the intensity of a series of physiologically conditioned biological and chemical processes. Statistically significant correlation coefficients were found between the concentration of PO_4^{3-} and P content in the aboveground sprouts of *G. robertianum*, *M. aquatica* and *V. officinalis* (Table 4). Concentrations of K^+ (*U. dioica*, *C. palustris*, *V. officinalis*), Mg^{2+} (*V. officinalis*) and Ca^{2+} (*A. reptans*), which

Table 4. The correlation coefficients results between concentrations of macronutrients (mg/kg) in the shoots of the plants and their concentration (mg/dm³) in groundwater (n=27, p<0.05, r_{crit}=0.324), significant correlation are in bold

	N	P	K	Mg	Ca	N	P	K	Mg	Ca
	<i>Ajuga reptans</i>					<i>Caltha palustris</i>				
pH	0.24	-0.15	0.21	0.84	-0.15	-0.18	-0.03	0.31	0.51	-0.46
NH ₄ ⁺	-0.02	-0.05	-0.38	-0.19	0.63	-0.26	-0.31	-0.18	0.33	0.46
NO ₃ ⁻	0.08	-0.31	-0.24	0.49	-0.11	-0.73	-0.47	-0.08	0.06	0.43
PO ₄ ³⁻	0.16	-0.13	-0.17	0.33	-0.62	-0.01	-0.09	0.15	0.15	0.22
K ⁺	-0.31	-0.18	-0.42	-0.15	0.42	0.15	0.34	0.43	0.24	0.61
Mg ²⁺	-0.18	-0.01	-0.12	-0.52	-0.13	-0.90	-0.63	-0.10	-0.24	0.61
Ca ²⁺	0.26	0.37	-0.15	0.07	0.07	-0.05	-0.03	-0.32	-0.03	0.23
	<i>Carex echinata</i>					<i>Geranium robertianum</i>				
pH	0.17	0.39	0.07	0.35	0.38	0.28	-0.51	0.02	0.32	-0.66
NH ₄ ⁺	0.08	-0.37	-0.71	-0.17	0.00	0.40	0.06	-0.18	-0.13	0.35
NO ₃ ⁻	-0.12	0.81	0.60	-0.06	0.56	0.19	-0.22	0.12	-0.08	-0.47
PO ₄ ³⁻	-0.69	-0.16	0.04	0.11	0.03	0.43	0.46	0.87	-0.41	0.50
K ⁺	-0.35	-0.24	-0.38	-0.16	0.11	-0.14	-0.05	-0.18	0.00	0.49
Mg ²⁺	-0.67	-0.34	-0.36	0.06	0.11	-0.35	0.24	0.09	-0.30	0.28
Ca ²⁺	0.57	-0.37	-0.64	-0.26	-0.36	0.30	-0.40	-0.37	-0.63	-0.23
	<i>Gymnocarpium dryopteris</i>					<i>Mentha aquatica</i>				
pH	-0.32	0.11	-0.71	0.43	0.03	-0.01	-0.43	-0.23	0.49	0.08
NH ₄ ⁺	-0.10	0.08	-0.38	-0.37	0.20	-0.59	-0.68	0.11	0.59	0.17
NO ₃ ⁻	-0.54	0.35	-0.73	0.57	0.18	0.13	0.37	-0.18	-0.06	0.01
PO ₄ ³⁻	-0.41	0.19	-0.14	0.46	0.04	-0.25	0.35	0.09	-0.41	0.16
K ⁺	-0.04	-0.01	-0.37	-0.06	-0.16	-0.81	-0.36	-0.69	0.40	0.15
Mg ²⁺	-0.11	-0.26	0.68	0.04	0.10	-0.77	-0.22	-0.56	-0.05	0.33
Ca ²⁺	-0.04	-0.19	0.43	-0.58	0.26	-0.32	-0.50	-0.40	0.40	0.13
	<i>Scirpus sylvaticus</i>					<i>Solanum dulcamara</i>				
pH	-0.09	0.31	0.94	0.19	0.06	-0.18	-0.36	0.40	0.32	0.03
NH ₄ ⁺	0.27	-0.02	-0.08	-0.16	-0.10	-0.46	-0.34	0.05	0.01	-0.01
NO ₃ ⁻	-0.08	0.08	0.54	-0.20	-0.40	0.12	-0.52	0.22	0.08	-0.03
PO ₄ ³⁻	-0.39	-0.19	0.36	0.36	-0.37	0.18	0.31	-0.09	-0.10	0.25
K ⁺	0.48	0.07	-0.25	-0.40	-0.12	0.12	-0.11	-0.01	-0.16	-0.02
Mg ²⁺	-0.21	-0.58	-0.63	0.01	-0.18	0.01	0.01	-0.19	0.33	0.44
Ca ²⁺	-0.44	-0.56	-0.21	0.53	0.07	-0.36	-0.08	0.19	-0.11	-0.01
	<i>Urtica dioica</i>					<i>Valeriana officinalis</i>				
pH	-0.18	-0.38	0.24	-0.31	-0.56	-0.99	-0.53	0.73	0.72	0.05
NH ₄ ⁺	-0.28	0.37	0.16	0.05	-0.01	0.99	0.49	-0.70	-0.63	-0.09
NO ₃ ⁻	-0.48	-0.29	-0.18	-0.02	-0.38	0.37	0.96	-0.86	0.60	0.94
PO ₄ ³⁻	-0.39	-0.07	-0.35	-0.07	0.33	0.68	0.97	-0.98	0.28	0.77
K ⁺	-0.27	0.36	0.09	0.17	0.03	0.42	0.98	-0.76	0.55	0.92
Mg ²⁺	0.13	-0.28	-0.01	0.18	0.44	0.21	0.91	-0.76	0.72	0.99
Ca ²⁺	0.30	-0.09	0.16	0.14	0.09	-0.82	-0.95	0.98	-0.06	-0.61

are available in groundwater, had a positive impact on the intake of phosphorus. These relationships indicate a close relationship between the chemistry of groundwater and vegetation (Karlsson et al. 2005). Phosphorus is one of the major structural elements of plants. Unlike nitrogen, it enhances plant growth and stimulates the generative organs of plants. However, in spite of geochemical stability, it undergoes migration in the environment and exerts negative pressure on the biogeochemical balance (Grzebisz 2003).

The physical and chemical parameters of groundwater and soils (Table 1, 2) within the area of the examined headwater were favourable to bioavailability and high accumulation of potassium in the sprouts of headwater plants (Table 3). The concentrations of NO_3^- (*C. echinata*, *S. sylvaticus*), PO_4^{3-} (*G. robertianum*, *S. sylvaticus*), K^+ (*C. palustris*), Mg^{2+} (*G. dryopteris*) and Ca^{2+} (*G. dryopteris*, *V. officinalis*), which are available in groundwater (Table 4), had a positive impact on the uptake of potassium, which confirms the close relationship between these environmental components.

However, substantial quantities of this element are washed out, especially from Histosols (Krzywy 2007). Potassium regulates the anion-cation balance, participates in the transport of metabolites and ions in the plant and limits the intake of other components, especially magnesium. It is a macrocomponent, which plants can take up in excess, often exceeding nutritional demand (Krzywy 2007, Horska-Schwarz and Spalek 2008, Parzych et al. 2015). During vegetation season, most K is accumulated in vegetative organs, i.e. in plant leaves and stems.

Calcium (Ca) and magnesium (Mg) penetrate into soil due to the weathering of minerals abundant in these components and during the decomposition of dead organic matter (Jonczak et al. 2015a). Precipitation may constitute an additional source along with the application of organic or mineral fertilizers (Szezwuk and Mazur 2004). The examined plants showed a series of statistically significant correlation coefficients between the contents of Mg (*A. reptans*, *S. dulcamara*, *V. officinalis*) and Ca (*C. echinata*, *V. officinalis*) in sprouts of these plants and the concentration of the respective nutritional ingredients in groundwater (Table 4). The intensity of calcium and magnesium assimilation by plants depends on the abundance of the substratum and on the content of other ions (Ostrowska and Porębska 2002). An increase in the magnesium content in plant shoots was significantly connected with the concentration of NH_4^+ (*C. palustris*, *M. aquatica*), NO_3^- (*A. reptans*, *V. officinalis*), PO_4^{3-} (*A. reptans*, *G. dryopteris*, *S. sylvaticus*), K^+ (*M. aquatica*, *V. officinalis*) and Ca^{2+} (*M. aquatica*). An increase in the calcium content in plant shoots was

significantly connected with the concentration of NH_4^+ (*A. reptans*, *C. palustris*, *G. robertianum*), NO_3^- (*C. echinata*, *C. palustris*), PO_4^{3-} (*U. dioica*, *G. robertianum*, *V. officinalis*) and K^+ (*A. reptans*, *C. palustris*, *G. robertianum*, *V. officinalis*). Magnesium can be taken in by both roots and leaves, but calcium is only taken up by roots of plants. As in the case of calcium, magnesium is a mobile element and can be easily washed out from the soil profile, especially from the layer of peat soils and peat and half-bog soils (Krzywy 2007). In addition, magnesium intake can reduce the pH of soil solution. Plants need a relatively small quantity of calcium for their optimal growth. A high content of assimilable potassium and ammonium nitrogen in soils can have an inhibitory effect on the intake of Mg^{2+} ions by certain species of plants.

The examples above demonstrate that the interrelationships between nutrients in the plant system and groundwater are quite complicated and interdependent (Ranade-Malvi 2011). Plants usually take up nutrients proportionally to their content in soil and groundwater (Parzych 2010, 2011, 2016). In the case of the plants of headwater niches, a high capacity to assimilate nutritional components was indicated. High contents of macrocomponents in the aboveground sprouts of plants at headwaters is probably conditioned by groundwater with a high level and a relatively low impact of anthropogenic factors. Similar high levels of biogenes in the shoots of headwater plants were observed by Horska-Schwarz and Spalek (2008).

In order to identify factors determining the participation of macrocomponents in the examined plants, a factor analysis was applied (PCA). For this calculation, monthly averages (for May, July and September) of the contents of macrocomponents (N, P, K, Mg and Ca) in the shoots of herbaceous plants (Table 5) during three consecutive years were used. By means of application of the method of main constituents, two independent factors were separated explaining 75% of the variability in the chemical composition of plants from

Table 5. Factor loading obtained from the principal components analysis (PCA) method on the basis of the macroelements content in the plants from headwater riparian forest

Element	Factor PC1	Factor PC2
N	-0.71	-0.47
P	-0.81	-0.15
K	-0.24	-0.82
Mg	-0.78	0.46
Ca	-0.83	0.35
Eigen values	2.49	1.25
Explained variance	0.50	0.25
[%]	75	

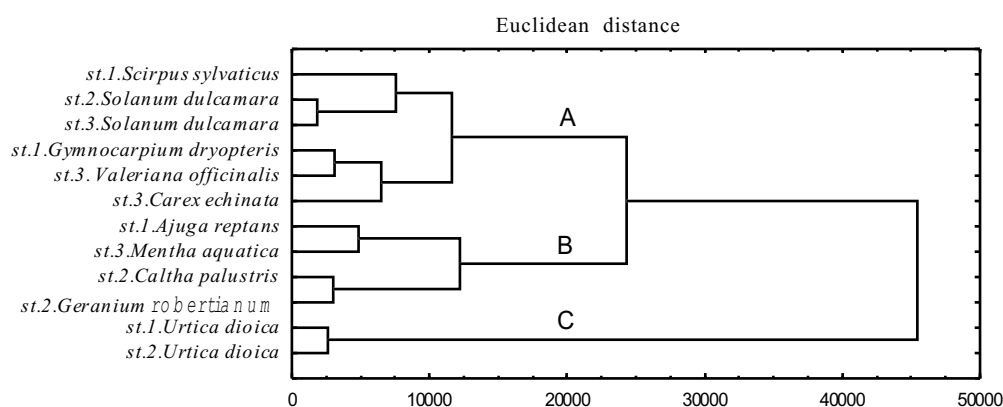
Note: in bold factor loading higher than 0.70 are highlighted

this headwater riparian forest. For interpretation of data, only factor loading values, which exceeded 0.7, were used. Factor 1 explained 50% of the variability and grouped N, P, Mg and Ca characterized by high, negative factor loads. Factor 2 explained 25% of the variability of the chemical composition of selected plant species and was created by K characterized by a high negative factor load. Plants generally have a high assimilation capacity for ions of K⁺. Potassium, due to its high mobility, is easily accumulated in the shoots of herbaceous plants in quantities exceeding the physiological demand of most species (Krzywy 2007).

Applying Ward's clustering method, three groups of plants were separated (Figure 1). Group A comprised the shoots of *S. sylvaticus*, *S. dulcamara*, *G. dryopteris*, *V. officinalis* and *C. echinata*, which were dominated by potassium, nitrogen and calcium (K>N>Ca). Group B included the shoots of *A. reptans*, *M. aquatica*, *C. palustris* and *G. robertianum*, with a dominating participation of potassium, calcium and nitrogen (K>Ca>N). Group C included the shoots of *U. dioica* (Ca>K>N). The diagram shows that *U. dioica* is distinguished from the other examined species by the

(42.7-81.4); the similar values were slightly lower for P (1.96-4.3), Mg (0.64-4.74) and Ca (0.25-1.26). Bioaccumulation factors (BF_s) had the lowest levels for N (0.41-0.61) depending on the species. In the case of *S. dulcamara* and *U. dioica* found at two different stands, no statistically significant differences were observed as to bioaccumulation factors (BF_s), which shows a strong influence of species characteristic features in the accumulation of nutrients, modified by environmental conditions. According to Ostrowska and Porębska (2002), the quantities of accumulated nutrients in plant shoots are characteristic for a given species. Comparable levels of bioaccumulation factors for K and Ca and slightly higher values for Mg were obtained in the case of aquatic plants by Łojko et al. (2015). Much higher values of bioaccumulation factors were obtained in relation to the content of the respective elements in groundwater (BF_w). The highest values were obtained in the case of K (9200-9280), N (8846-4140), P (4863-24589) and Mg (1068-2239). The lowest values were found for calcium (124-742). The levels of bioaccumulation factors showed diversity during the period of research, which was the effect of the variable demand

Figure 1. Variability of the investigated plant species in relation to the contents of N, P, K, Ca and Mg (Euclidean distance, Ward's clustering method)



highest concentration of N, P and Ca, independent of the location (Table 3). A high concentration of macrocomponents with a dominating participation of calcium in the shoots of *U. dioica* was also described by Ștef et al. (2010) and Gjorgieva et al. (2011), which is characteristic for this species. According to Szewczuk and Mazur (2004), *U. dioica* is characterized by an exceptionally high content of nutritional components in comparison to other plants.

Bioaccumulation factors

The values of the bioaccumulation factors exemplify the diverse accumulative properties of the examined species of herbaceous plants in reference to macrocomponents in soil and groundwater (Figure 2). The highest values of BF_s were obtained in the case of K

of plants for nutrients during the vegetative season (Figure 2). The high values of BF_s factors in relation to nitrogen, phosphorus, magnesium and calcium were characteristic for the shoots of *U. dioica*, and for *G. robertianum* in relation to potassium. In the case of BF_w, the highest average content of nitrogen was characteristic for the shoots of *C. palustris*, phosphorus for *C. echinata*, potassium for *G. robertianum*, and magnesium and calcium for *U. dioica*. The shoots of *S. dulcamara* (N, P, K) and *U. dioica* (N, P, K, Ca, Mg) were characterized by the highest diversity of bioaccumulation factors during the period of research. The non-parametric Kruskal-Wallis test showed statistically significant differences in the bioaccumulation factors of macrocomponents in the shoots of the examined plant species in this headwater riparian forest.

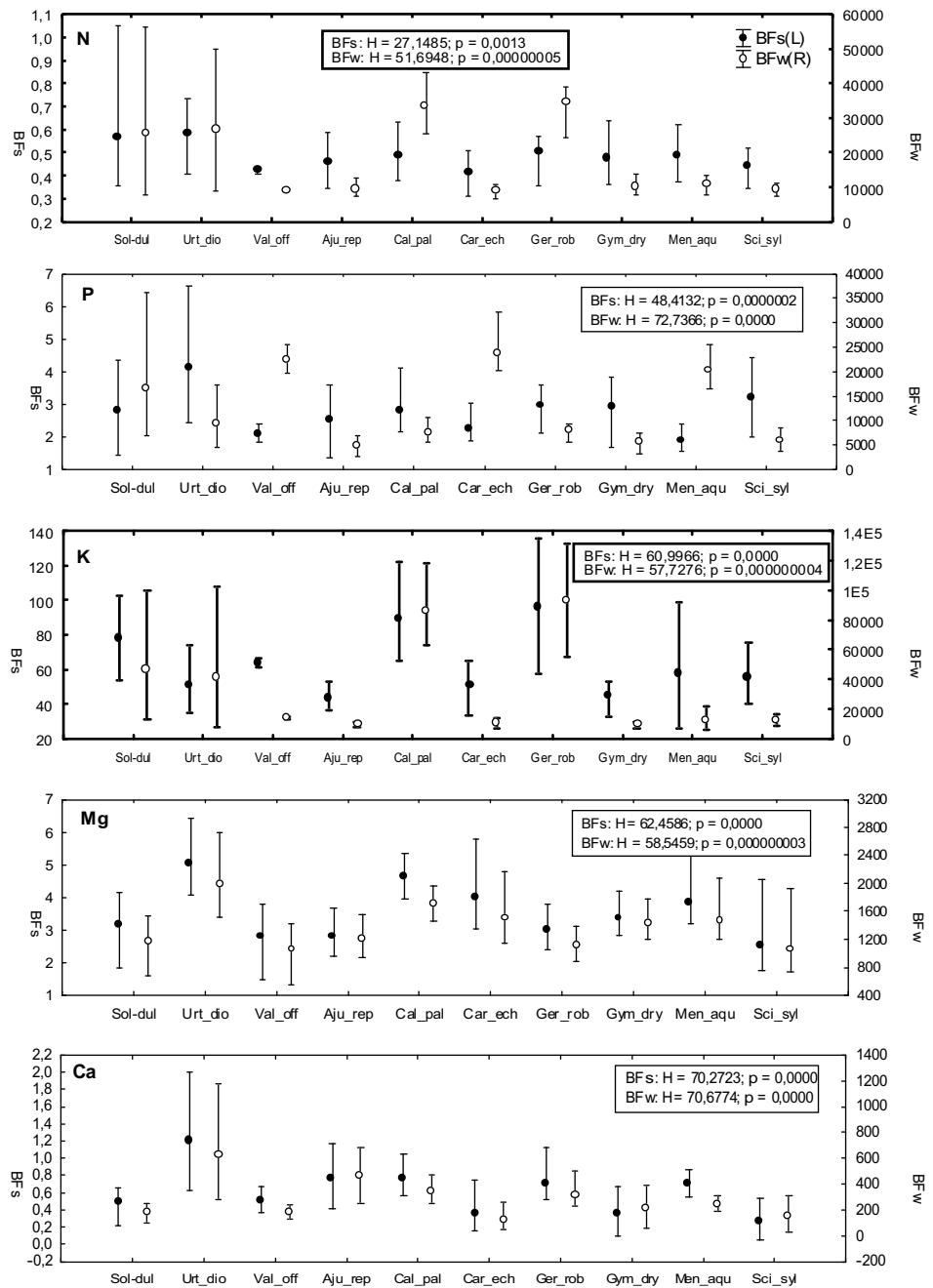


Figure 2. Mean, minimum and maximum values of bioaccumulation factors (BF_s and BF_w) of macronutrients in plants from headwater riparian forest with Kruskal-Wallis's test results

Conclusions

The examined soils were characterized by a weak acidic reaction, high nitrogen abundance and low phosphorus, potassium, magnesium and calcium abundance. The chemical composition of groundwater showed variability during the vegetation season and was shaped by diverse demand of plants for nutritional components. The aboveground sprouts of herbaceous plants were characterized by a good supply of N, P, K, Mg and Ca, while potassium was accumulated in larger quantities than required by physiological demand. This is evidence that

the species under consideration show a relatively high capacity for the assimilation of nutritional components. The high contents of macrocomponents in the plants of headwaters are conditioned by the high groundwater level. The series of statistically significant correlations between the content of macrocomponents in the shoots of the plants and the chemistry of groundwater shows close correlations among the examined components of the environment. The levels of bioaccumulation factors showed variability during the research period, which were the effect of variable demand of the plants for nutrients during the vegetative season.

The highest values of bioaccumulation factors (BF_s) were found for the sprouts of *U. dioica* (N, P, Mg, Ca), *S. dulcamara* (N, P) and *G. robertianum* (K). The lowest values of BF_s were found in the sprouts of *C. echinata* (N), *V. officinalis* (P), *A. reptans* (K) and *S. sylvaticus* (Mg, Ca). Similar values of BF_s obtained in the case of *S. dulcamara* and *U. dioica* growing at various stands reflect the strong impact of generic features in the accumulation of nutritional components, modified by environmental conditions.

Most nutritional components from groundwater (BF_w) were taken in by sprouts of *U. dioica* (NH_4^+ , NO_3^- , Mg^{2+} , Ca^{2+}), *S. dulcamara* (PO_4^{3-}) and *G. robertianum* (K^+). The highest average levels of BF_w in the case of nitrogen were found in the shoots of *C. palustris*, phosphorus in *C. echinata*, potassium in *G. robertianum*, and magnesium and calcium in *U. dioica*. The shoots of *S. dulcamara* (N, P, K) and *U. dioica* (N, P, K, Ca, Mg) were characterized by the highest variability of bioaccumulation factors during the period of study.

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