DOI: 10.5586/asbp.3640

Publication history
Received: 2019-05-25
Accepted: 2019-11-27
Published: 2019-12-31

Handling editor
Jan Holeksa, Faculty of Biology, Adam Mickiewicz University in Poznań, Poland

Authors' contributions
PS: research design, writing the manuscript, preparing samples and laboratory work, data analysis; DS: research design, writing the manuscript, data analysis; OS: field work, preparing samples; AK: research design, writing the manuscript, field work, preparing samples; DW: research design, field work, preparing samples; AP: research design, writing the manuscript, preparing samples and laboratory work; OB: preparing samples and laboratory work

Funding
The research was financially supported by the Polish Ministry of Science and Higher Education.

Competing interests
No competing interests have been declared.

Citation
Sugier P, Sugier D, Sozinov O, Kołos A, Wołkowycki D, Plak A, et al. Characteristics of plant communities, population features, and edaphic conditions of *Arnica montana* L. populations in pine forests of mid-Eastern Europe. Acta Soc Bot Pol. 2019;88(4):3640. https://doi.org/10.5586/asbp.3640

ORIGINAL RESEARCH PAPER

Characteristics of plant communities, population features, and edaphic conditions of *Arnica montana* L. populations in pine forests of mid-Eastern Europe

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Abstract
Mountain arnica, *Arnica montana* L., an herbaceous plant species critically endangered in Europe, is the source of raw material, which is abundant in its secondary metabolites. During the field investigation conducted in Augustów and Knyszyn forests (Poland) and in Grodno Forest (Belarus), the plant species composition and population characteristics were measured. Additionally, to evaluate the edaphic conditions of arnica populations, soil samples were taken and analyzed. The sandy and very nutrient-poor soils are characterized by strong acidity and a very low concentration of macro- and microelements. The analyzed characteristics of the studied populations indicate a good status of populations located in Grodno Forest. However, the very small number of individuals and the very small proportion of flowering individuals in the populations in Augustów Forest and Knyszyn Forest indicate the need for active protective actions. *Calamagrostis arundinacea* can play the role of a competitor; therefore, during planning active protection, individuals of this species should be eliminated, and particular attention should be paid to the frequency and coverage of this plant species and the plant height of the herb layer. The dependence between population characteristics, especially the proportion of flowering stems and the concentration of available phosphorus, may indicate the effect of the concentration of this macroelement on flowering and, in consequence, provide a greater chance for the generative propagation of this plant species.

Keywords
Arnica montana; population characteristics; edaphic conditions; pine forest populations

Introduction

Arnica montana is an herbaceous plant species that is endemic to Europe and occurs in the heathlands and grasslands of lowland areas and in meadows of the Alps, Sudetes, and Eastern Carpathians, as well as in the latitude gradient from Norway to the Pyrenees and the Balkan Peninsula [1–5]. Natural populations constitute resources of important genetic diversity in Europe. This plant species is a source of raw material, which is abundant in it secondary metabolites; therefore, it has several different applications, e.g., it is widely used in the pharmaceutical and cosmetic industries [6–8]. Recently, arnica genotypes taken from collections and from natural sites have been the subject of various studies considering agricultural factors modifying the yield and chemical composition of raw material [8–13]. Arnica has been used in folk medicine for many years, and the demand has exerted pressure on the environment by increasing its collection for medicinal purposes, which has led to a rapid decline in this species. Presently, this species is covered by various forms of protection [14–18]. However, flower heads are still obtained from natural sites in Romania and Ukraine, where the species is not subjected to strict protection; this has a negative impact on the populations [19–21]. In addition, the biggest threats in recent decades, especially in Western Europe, are posed by nutrient enrichment through fertilization, land-use change, habitat fragmentation, grazing pressure [19,20], and the deposition of nitrogen from the atmosphere [5,22,23]. The last factor mentioned was the cause of eutrophication and changes in soils in the Karkonosze Mountains [24], and, in consequence, the reduction of arnica populations. Nitrogen has a significant impact on the expansion and dominance of grasses in grassland plant communities, which are important competitors of arnica [22,24,25].

Generally, arnica habitats are highly diverse. This species was registered in nutrient-poor and dry grasslands and heathlands in the Netherlands [4,22,26], as well as in the grasslands and shrublands of mountain environments [1–3]. It also grows in subalpine grasslands and dwarf-shrub vegetation, meadows on siliceous soils, wet meadows, montane pastures, marginal parts of spruce forests, and along roads in pine forests, as well as openings in coniferous forests and their edges [17,18,27,28]. In Western Europe, arnica arouses great interest as a research object. The habitat characteristics, reproduction, stage structure, and genetic structure of colline and montane arnica populations were studied in Belgium, Luxembourg, and Germany [5,29]. In turn, the phenotypic and genetic variation in *A. montana* was investigated in populations that have been fragmented by afforestation since 1930 in the Rhön, a mountainous region in central Germany [3]. The knowledge gained was essential in the active protection carried out recently in Belgium [30] and Germany [31]. In recent years, intensified research has also been conducted on arnica habitats in the Eastern Carpathians. Mardari et al. [32] described the characteristics of plant communities with *A. montana*. The distribution and participation of *A. montana* in phytocenoses and the effect of mowing and grazing pressure on the state and dynamics of the resources of this plant species were presented by Vantjuh [20,21]. However, there is very little knowledge about the number and population size of arnica occurring in the lowlands of Central and Eastern Europe, where the largest grouping of populations of this species occurs in a dense geographic range in mid-Eastern Europe [1,2], especially in Belarus. Similarly, the effect of edaphic conditions on arnica pine forests populations, especially in mid-Eastern Europe, is not known. The objective of our research is to supplement this gap in the knowledge. Therefore, the aims of the study were (***i***) to characterize plant communities with mountain *A. montana* in mid-Eastern Europe, (***ii***) to determine the main population characteristics and soil conditions of *A. montana* habitats, and (***iii***) to indicate edaphic factors determining fitness characteristics and favoring the persistence of populations of this endangered plant species.

Material and methods

Field study and population characteristics

In July 2017, we selected 20 populations of *A. montana* in coniferous pine forests; five in Augustów Forest (AF), five in Knyszyn Forest in Poland, and 10 in Grodno Forest (GF) in Belarus (Fig. 1). In each of the sites, one representative plot of 200 m^2 was set up in areas with *A. montana* and a phytosociological relevé was made. In each stand, the percent cover of the tree layer, shrub layer, herb layer, and bryophyte and lichen layer, as well as the height of herbaceous vegetation (HHL), were estimated. The HHL

Fig. 1 Distribution map of the studied *Arnica montana* populations in mid-Eastern Europe.

was estimated on the basis of 20 measurements within the vegetation patches where the phytosociological relevés were made. All vascular plant species, bryophyte species, and lichens were recorded and the covers were estimated. In each of the studied populations (within the vegetation patches where the phytosociological relevés were made), 10 soil samples were randomly taken using a sampler from the depth of 0–15 cm at each studied site and then pooled, packed in foil bags, and transported to the laboratory.

In each of the studied populations, the characteristics of *A. montana* were measured by estimating the total number of rosettes (TNR), total number of flowering stems (TNFS), total number of rosette aggregations (TNA), and population area (PA); and in the designated plots (each 25 m^2), the number of rosettes (NRP), number of rosette aggregations (NAP), and number of rosettes in the aggregation (NRAP). All of these research plots were established inside the vegetation patches where the phytosciological relevés were made. Moreover, the length of the longest leaf in the rosette (LL) and the width of the longest leaf in rosette (WL) were measured on 20 randomly selected rosettes from the area of the entire local population.

Analysis of soil samples

The organic carbon content (TOC) and total nitrogen concentration (Ntot) were determined using a LECO CNS Elemental Analyzer (LECO Truspec CN; LECO Corporation, St. Joseph, MI, USA). The determinations were tested in relation to a certified reference material (soil calibration sample ref. 502-062; LECO Corporation is A2LA accredited in accordance with the International Organization for Standardization ISO/IEC 17025: 2005 – certificate No. 3285.01). Soil granulometric composition was analyzed using a Malvern Mastersizer analyzer with a HydroG dispersal unit (Mastersizer MS-2000, Great Britain), and the soil determination methodology was based on the United States Department of Agriculture (USDA) [33]. The pH was measured potentiometrically in water and 1 M KCl. The content of $CaCO_3$ was assessed using the volume method proposed by Scheibler (Calcimeter DIN 19682/19684; Eijkelkamp Soil & Water, Giesbeek, the Nederlands) [34]. Soil acidity (Hh) was measured after extraction from 1 M KCl according to van Reeuwijk [35]. The total exchangeable bases (TEB) (Ca, Mg, K, and Na) was determined using an atomic absorption spectrophotometer (AAS; Agilent 240 FS F-AAS, Santa Clara, CA, USA). The cation exchange capacity (CEC) of soils was calculated as the sum of the hydrolytic acidity and TEB [33]. The plant available P and K forms in the soil were extracted with the Egner–Riehm method, and the calcium lactate was determined using Vis spectroscopy (Lambda 12 UV spectrophotometer; PerkinElmer, Waltham, MA, USA). To determine the pseudo total (hereafter referred to as the total) content of heavy metals, the soil samples were dissolved with aqua regia (ISO 11466). Potentially bioavailable forms of Cu (Cu-B), Cd (Cd-B), Cr (Cr-B), Ni (Ni-B), Pb (Pb-B), and Zn (Zn-B) were extracted using 0.01 M $CaCl_2$, as previously described [36]. Trace elements in soil extracts were determined using the F-AAS technique (Agilent 240 FS F-AAS).

Statistical analysis

The variation in the plant species composition of the studied plant communities and the variations in population characteristics with the proportion of *A. montana* plots were explored using principal component analysis (PCA), as the detrended correspondence analysis (DCA) results (the length of the first DCA axis not exceeding 3 *SD*) detected a modal structure in the vegetation data [37]. Ordination analyses were conducted using the MultiVariate Statistical Package (MVSP) [38]. Initially, all data were tested for normality and variance heterogeneity was checked. Because the majority of the data was not normally distributed (results of Shapiro–Wilk test) or the variance was not homogeneous (results of Levene's test), the nonparametric Mann–Whitney *U* test was used for the analysis of specific sample pairs for significant differences, and the non-parametric Spearman rank correlation coefficient was used to analyze the correlations between TNFS, LL, WL, and soil properties (except microelements and granulometric fractions). All results were expressed as means and minimum and maximum standard deviations, and differences were considered significant at $p < 0.05$. All statistical analyses were carried out using the STATISTICA 10.0 software.

Results

Characteristics of the forest plant communities with *A. montana*

The results of PCA prepared on the basis of the cover of particular plant species are presented in Fig. 2. The first axis explained 16.5% and the second axis accounted for 13.3% of the total variation. In the ordination space, two groups of sites can be distinguished. The first one on the right side comprises sites located in Augustów Forest (AF) and Knyszyn Forest (KF) in Poland, and on the left side, there are sites located in Grodno Forest (GF) in Belarus. In the Augustów and Knyszyn forest (AKF) sites, species such as *Chimaphila umbellata*, *Maianthemum bifolium*, *Thymus serpyllum*, *Cladonia rangiferina*, *Polytrichum commune*, and *Polytricum juniperinum* were not noted. In turn, *Fragaria vesca*, *Potentilla erecta*, *Rubus idaeus*, and *Veronica officinalis* were not found in the GF sites. In relation to the AKF plant communities, the GF plant communities with the proportion of *A. montana* are characterized by higher covers of *Pinus sylvestris*, *Juniperus communis*, and *Dicranum polysetum*, and a lower cover of *Calamagrostis arundinacea* (Tab. 1). The GF populations are characterized by a statistically higher cover of the tree layer and a higher cover of *Pinus sylvestris*, which is the dominant in this layer. In turn, the AKF populations are characterized by an approximately twofold higher HHL value. For the other characteristics of plant communities, no statistically significant differences were found.

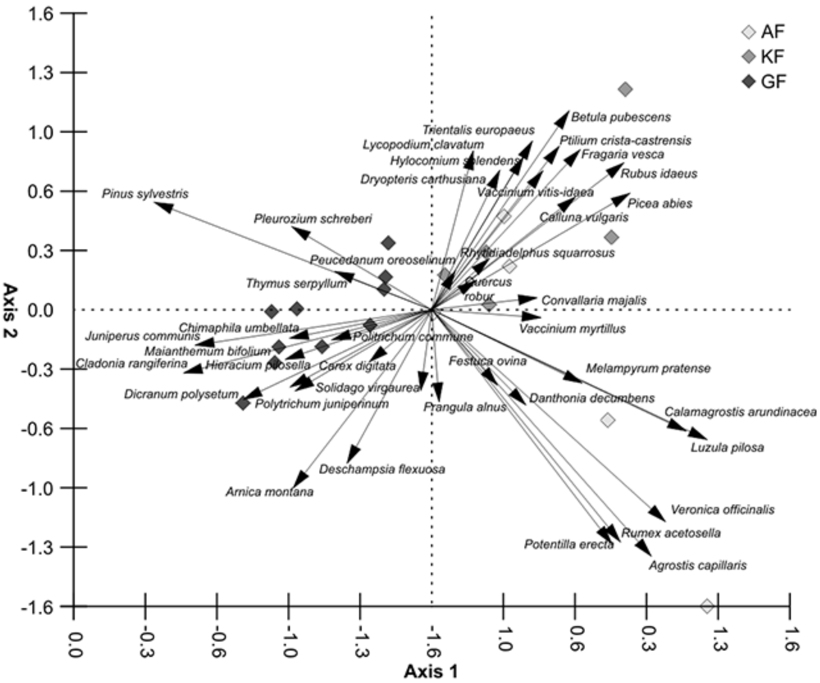

Fig. 2 Results of principal component analysis (PCA) based on the cover of the studied plant species in phytocoenoses with a share of *Arnica montana*; AF – Augustów Forest; KF – Knyszyn Forest; GF – Grodno Forest.

Tab. 1 Characteristics of the phytocoenoses with the proportion of *Arnica montana*.

	AKF				GF			
Characteristics	**Mean**	***SD***	**Min**	**Max**	**Mean**	***SD***	**Min**	**Max**
CA	41.0 [b]	27.5	15	95	71.0 [a]	18.9	13	65
CB	7.6	24.5	30	100	11.7	8.9	72	100
CC	46.0	26.8	5	80	37.7	20.6	20	90
CD	78.0	18.0	10	70	90.1	7.38	60	80
HHL	25.2 [a]	10.2	12	46	12.8 [b]	10.0	8	18
PINSYL	41.0 [b]	17.1	10	70	71.0 [a]	7.0	60	80
JUNCOM	1.0 [b]	1.5	1	5	12.0 [a]	8.6	1	30
CALARU	10.1 [a]	9.6	1	25	0.2 [b]	0.4	1	1
FESOVI	4.3	8.7	1	30	3.7	3.1	1	10
VACMYR	14.7	17.5	1	50	8.8	10.9	1	30
VACVIT	10.6	8.7	1	30	4.6	4.6	1	15
DICPOL	3.1 [b]	3.9	1	10	16.5 [a]	14.3	1	50
HYLSPL	27.5	27.9	5	80	20.0	20.4	5	70
PLESCH	48.5	22.4	20	80	55.5	18.9	30	90

CA – tree layer cover (%); CB – shrub layer cover (%); CC – herb layer cover (%); CD – bryophyte and lichen layer cover (%); HHL – height of herbaceous vegetation (cm); PINSYL – *Pinus sylvestris* cover (%); JUNCOM – *Juniperus communis* cover (%); FESOVI – *Festuca ovina* cover (%); CALARU – *Calamagrostis arundinacea* cover (%); VACMYR – *Vaccinium myrtillus* cover (%); VACVIT – *Vaccinium vitis-idaea* cover (%); DICPOL – *Dicranum polysetum* cover (%); HYLSPL – *Hylocomium splendens* cover (%); PLESCH – *Pleurozium schreberi* cover (%); AKF – Augustów and Knyszyn forests; GF – Grodno Forest. Different letters indicate significant differences according to the Mann–Whitney test ($p < 0.05$).

Population characteristics

As shown in the PCA ordination graph, the studied data revealed a clear distinction among the studied sites (Fig. 3). Two groups are separated in the ordination space of PCA: the AKF and GF populations. The first axis explained 44.0% and the second axis accounted for 24.6% of the total variation (Fig. 3). The first axis is positively correlated with TNFS, TNR, TNA, and PA. The second axis indicates a gradient in NRP and NAP. Arnica sites showing higher values of the population characteristics TNR, TNFS, TNA, and PA were placed on the right side of the ordination space (GF populations), while those having low values of these characteristics were located on the left side of Axis 1 (AKF populations). Moreover, there is a clear segregation between the groups of the arnica populations. The GF arnica populations cover a larger area and are more numerous. They are characterized by a TNR that is several times bigger, TNFS that is approximately 20 times higher, TNA that is five times higher, and PA that is 4 times higher than those in the AKF populations, whereas in turn, the WL was significantly lower (Tab. 2).

Soil properties

The analyzed soils in the arnica habitats did not contain $CaCO_3$ and were characterized by their strong acidity; the mean values of the pH(H_2O) and pH(KCl) of the AKF soils were significantly higher than those of the GF soils (Tab. 3). The analyzed soils were sandy and loamy sandy, and no clay fraction was recorded. However, the AKF soils were characterized by their higher content of silt fraction and lower content of sand fraction compared to those in the GF soils. The TOC, N, and C:N ratio in the studied surface levels did not differ in both locations. The studied soils exhibited low concentrations of bioavailable forms of P and K; however, the P concentration in the GF soils was significantly higher than that in the AKF soils. Moreover, very low abundances of Na^+,

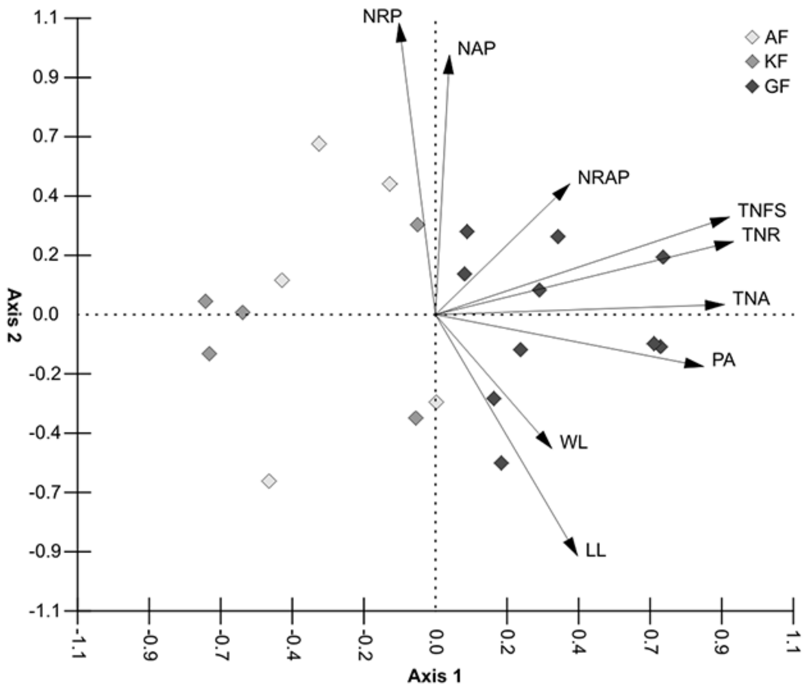

Fig. 3 Results of principal component analysis (PCA) based on the characteristics of the *Arnica montana* population. TNR – total number of rosettes in the population; TNFS – total number of flowering stems in the population; TNA – total number of rosette aggregations in the population; PA – population area; NRP – number of rosettes on the studied plot; NAP – number of rosette aggregations on the studied plot; NRAP – number of rosettes in the aggregation on the studied plot; LL – length of the longest leaf of rosette; WL – width of the longest leaf of rosette; AF – Augustów Forest; KF – Knyszyn Forest; GF – Grodno Forest.

Tab. 2 Characteristics of the *Arnica montana* populations.

	AKF				GF			
Characteristics	**Mean**	***SD***	**Min**	**Max**	**Mean**	***SD***	**Min**	**Max**
TNR	579 [b]	568	74	1,480	9,926 [a]	12,913	981	42,245
TNFS	20.1 [b]	21.9	1	53	411.2 [a]	368.5	57	994
TNA	46 [b]	50	6	154	273 [a]	269	45	935
PA	0.94 [b]	1.65	0.01	4.87	4.41 [a]	2.85	0.90	9.30
NRP	14.12	4.95	8	23	46.03	50.16	4	162
NAP	8.0	9.3	1	31	4.7	2.2	1	8
NRAP	98	101	20	352	73	43	15	140
LL	13.50	4.76	9.50	21.55	16.01	3.16	11.71	21.43
WL	3.34 [b]	0.60	2.63	4.66	3.83 [a]	0.51	2.99	4.82

TNR – total number of rosettes in the population; TNFS – total number of flowering stems in the population; TNA – total number of rosette aggregations in the population; PA – population area (ha); NRP – number of rosettes on the studied plot; NAP – number of rosette aggregations on the studied plot; NRAP – number of rosettes in the aggregation on the studied plot; LL – length of the longest leaf in rosette (cm); WL – width of the longest leaf in rosette (cm); AKF – Augustów and Knyszyn forests; GF – Grodno Forest. Different letters indicate significant differences according to the Mann–Whitney test ($p < 0.05$).

Tab. 3 Physicochemical properties of soils of *Arnica montana* habitats.

	AKF				GF			
Characteristics	Mean	*SD*	Min	Max	Mean	*SD*	Min	Max
pH(H_2O)	4.68 [a]	0.17	4.32	4.95	4.40 [b]	0.25	3.83	4.69
pH(KCl)	3.91 [a]	0.15	3.51	4.04	3.71 [b]	0.27	3.07	3.96
Ntot (%)	0.12	0.02	0.10	0.15	0.11	0.04	0.08	0.22
TOC (%)	2.52	0.52	1.69	3.08	2.42	1.02	1.05	4.81
Na^+ [cmol(+) kg^{-1}]	0.011	0.003	0.010	0.020	0.010	0.000	0.010	0.010
K^+ [cmol(+) kg^{-1}]	0.013	0.005	0.010	0.020	0.012	0.004	0.010	0.020
Mg^{2+} [cmol(+) kg^{-1}]	0.024	0.011	0.010	0.050	0.024	0.011	0.010	0.040
Ca^{2+} [cmol(+) kg^{-1}]	0.10	0.04	0.05	0.16	0.11	0.04	0.03	0.19
Hh[cmol(+) kg^{-1}]	10.26	1.83	7.35	12.67	10.30	2.77	7.42	16.57
CEC [cmol(+) kg^{-1}]	10.41	1.85	7.44	12.84	10.46	2.76	7.59	16.74
TEB [cmol(+) kg^{-1}]	0.14	0.05	0.09	0.24	0.16	0.04	0.07	0.23
BS (%)	1.39	0.38	0.87	2.01	1.63	0.67	0.61	2.98
P (mg kg^{-1})	7.12 [b]	1.67	5.08	10.33	9.47 [a]	2.72	4.71	14.68
K (mg kg^{-1})	19.71	7.58	11.80	36.80	15.65	5.21	9.54	23.10
FeT (mg kg^{-1})	3875	945	1700	5190	3381	1070	1381	4838
MnT (mg kg^{-1})	129.13	59.82	33.30	197.70	93.18	55.91	22.50	198.10
PbT (mg kg^{-1})	10.86 [a]	3.88	0.10	13.50	1.18 [b]	3.38	0.10	10.80
CuT (mg kg^{-1})	2.06	2.99	0.03	8.82	0.09	0.04	0.03	0.15
ZnT (mg kg^{-1})	45.98 [a]	14.93	28.20	77.50	24.23 [b]	7.89	11.90	36.50
CrT (mg kg^{-1})	8.39	1.64	5.28	11.30	6.96	2.47	2.76	10.80
FeB (mg kg^{-1})	7.50	4.73	1.50	14.40	10.70	7.52	2.04	23.10
MnB (mg kg^{-1})	8.89 [a]	6.17	1.70	19.10	1.81 [b]	1.28	0.03	4.51
ZnB (mg kg^{-1})	1.56 [a]	1.09	0.44	3.67	0.30 [b]	0.22	0.08	0.70
CuB (mg kg^{-1})	0.17	0.23	0.04	0.63	0.52	0.66	0.04	1.57
Fraction (mm):								
0.002–0.02	2.3 [a]	0.8	1.2	3.8	1.6 [b]	0.8	0.3	2.7
0.02–0.05	3.7	1.1	2.5	5.9	2.1	0.8	1.2	3.7
0.05–0.1	6.6 [b]	3.3	2.6	13.7	14.8 [a]	11.5	1.2	31.2
0.1–0.25	33.4 [b]	11.2	13.9	51.5	56.9 [a]	3.1	52.5	62.8
0.25–0.5	34.0 [a]	8.3	20.1	44.3	22.2 [b]	12.4	6.4	39.3
0.5–1.0	16.5 [a]	8.5	5.0	33.0	1.7 [b]	0.8	0.5	2.9
1.0–2.0	3.5	4.4	0.0	15.2	0.7	1.0	0.0	2.9

Ntot – total nitrogen; TOC – total organic carbon; Hh – hydrolytic acidity; TEB – total exchangeable bases; CEC – cation exchange capacity; BS – base saturation; T – total forms; B – bioavailable forms; AKF – Augustów and Knyszyn forests; GF – Grodno Forest. Different letters indicate significant differences according to the Mann–Whitney test ($p < 0.05$).

K^+, Mg^{2+}, and Ca^{2+}, and simultaneously very low values of parameters such as TEB, were detected in the two soil groups. The concentrations of NiT, CdT, PbB, NiB, and CdB were very low and below the detection limit (data not presented). The concentrations of other elements were similarly low. The AKF soils were characterized by a higher concentration of Mn in relation to the GF soils.

Population characteristics vs. soil properties

The Spearman correlation results showed only a positive relationship between the concentration of the available phosphorus and the TNFS, LL, and WL (Fig. 4).

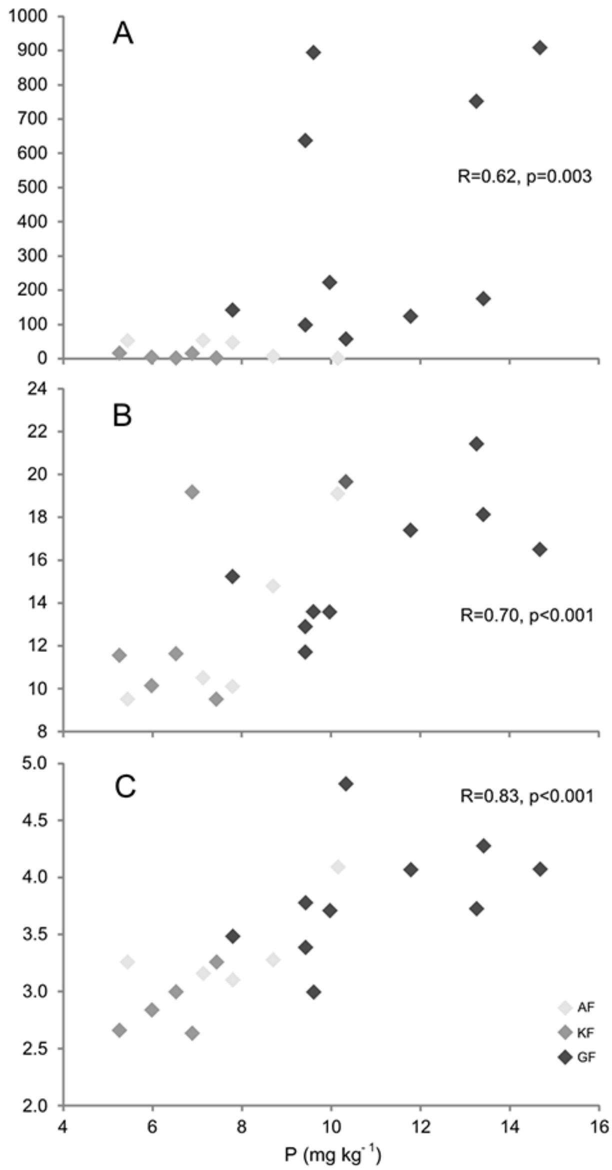

Fig. 4 Correlation between the concentration of available P (mg kg^{-1}) and total number of flowering stems in the population (**A**), length (cm) of the longest leaf in rosette (**B**), and width (cm) of the longest leaf in rosette (**C**). AF – Augustów Forest; KF – Knyszyn Forest; GF – Grodno Forest.

Discussion

The localization of the studied arnica stands near pathways and forest separating lines is typical for pine forest populations in this region of Europe [17,18,27,39]. Arnica rosette groups are characteristic for both nonforest [4,32] and forest populations [17,39], and is the result of generative and vegetative propagation [4]. AKF populations are similar in respect to structure; they are characterized by a similar number of rosettes per unit area and a similar number of rosette aggregations per unit area. However, the rosette density in the studied pine forest populations was several times lower than in the other European populations mentioned [5]. Similarly, the population size measured by estimating the number of flowering stems of the studied arnica pine forest population is several or even several tens of times smaller than the size of the populations in grassland vegetation from the *Violion caninae* alliance in Belgium, Luxembourg, France, and Germany (280–633 m a.s.l.), as well as in montane sites in the Vosges Mountains (France, 1,175–1,250 m a.s.l.) from the *Nardion strictae* alliance in Central Europe [5].

A big threat to arnica populations in Europe is grass, which negatively influences the vitality of this species and is regarded as a serious competitor [5,24,25,39,40]. Many grasses limit the development and establishment of seedlings and young arnica plants [41]. The main cause of the increased frequency of monocots in arnica populations in the Netherlands and Germany was the atmospheric nitrogen enrichment of grassland ecosystems accompanied by a loss of species richness, especially dicots [42]. Grasses are considered serious competitors in phytocoenoses with arnica, negatively influencing the vitality of this species [5,24,25,40]. *Calamagrostis arundinacea* is a natural component of pine forests in Lithuania, Poland, and Belarus [17,18,27], as well as in grassland communities in the Eastern Carpathians [20,21,32]. Moreover, in the presented results, there are clear differences between the cover of this plant and the cover of herbaceous vegetation in the AKF population, which is less abundant in relation to the GF populations. This confirms the thesis that the reed grass can be one of the factors determining the condition of the arnica population in Augustów and Knyszyn forests.

Another threat to arnica is posed by dwarf shrubs. In mountain areas, excessive grazing can facilitate the invasion of arnica sites by *Vaccinium myrtillus* [32]. Moreover, in pine forest populations, arnica individuals are shadowed by this dwarf shrub, which can play the role of a serious competitor [17,39]. The low frequency and cover of *V. myrtillus* suggest that arnica prefers sites without – or with very low cover of – this dwarf shrub [39]. A similar situation is observed in the present study. The *V. myrtillus* cover is low in the AKF and GF populations, i.e., it does not exceed 15%. In turn, the cover of *V. vitis-idaea* is even lower. Therefore, in the studied populations, dwarf shrubs that are natural components of pine forests are not likely to pose a threat.

The soils of the arnica habitats studied are typical Podzolic soils without the clay fraction, and with a small proportion of silt fraction. According to the USDA classification [33], the upper layer of soils was represented by the granulometric group of sands. The studied arnica pine forest habitats are located on nutrient-poor, strongly acidic, Podzolic soils, which are common in the studied regions of Poland and Belarus [43–45]. The acidity of the studied soils probably depends on processes related to climatic conditions, where rainfall exceeds evapotranspiration [46]. In turn, the organic matter content in soils is the basic indicator determining their physicochemical properties and biological processes. It determines the dynamics of biological activity, thereby influencing the quality of soils [47]. It is worth noting that the described arnica habitats were characterized by relatively high TOC contents [48,49]. The surface levels of the studied arable habitats in both Poland and Belarus were characterized by high nitrogen contents in relation to the agronomic categories of agriculturally used soils [50]. At the same time, based on the soil degradation criteria calculated with the C:N ratio [47], it can be concluded that the surface levels (0–20 cm) of the soils located in both Poland and Belarus were moderately degraded (average degradation C:N 18:1 – C:N 30:1). The properties of the studied soils are characteristic of dystrophic environments [48], and the content of heavy metals expressed as total and potentially bioavailable forms in the analyzed soils (Tab. 3) is evidently low and does not exceed the natural concentration level [51,52]. In the literature, researchers underline the role of the TEB in such soils as a very important factor for the existence of *A. montana* populations [22,39]. The results presented in this paper do not confirm this finding, because the soils of the small and large populations are characterized by very low values of this parameter, and the dependencies between the TEB and the characteristics of the population are invisible. However, there are clearly visible differences between the available phosphorus contents in the soils of the very small populations in AKF and the large populations located in GF (Tab. 3). Generally, in all cases, the content of available P in the soils is very low. In these unfavorable environmental conditions, P can be a limiting factor. This macroelement, like some other macro- and microelements, have an effect on flowering and seed or fruit production [53–55]. This effect is exerted by the use of B and humic substances in flowering and flower head production in arnica [10,12]. The available P concentration was very low. However, like the TEB [22,39], this factor can affect arnica flowering, generative plant propagation, and consequently population size and fitness even with very small changes in the concentration of this macroelement in very harsh edaphic conditions. The importance of phosphorus in the presented results was expressed by the positive correlation between the available P concentration in soils and population size, length of the longest leaf, and width of the longest leaf (Fig. 4). In the studies on arnica found in natural habitats, it was shown that the population size estimated by the number of flowering stems was positively correlated with the percentage of achenes containing a developed embryo, achene weight, and percentage of germinating seeds [3]. Therefore, a greater concentration of available phosphorus in soils under arnica populations can affect the flowering, generative plant propagation, and population viability of this plant species.

The GF populations are characterized by several times higher PA, TNR, and TNFS compared to those in AKF populations. It is possible that, in addition to the soil factors discussed earlier, climatic conditions are important in a well-preserved population such as that in Belarus. In the last few years of field observation, rosette death in the Polish populations was observed during the hot summer period (Kołos, field observations), similar to that seen in another arnica population in Polish lowlands (Załuski 2016, personal comments). The observations presented in this paper are in accordance with the recent literature reports. A negative impact of a dry spring and June on arnica achene germination was also reported during the in-situ resettlement of arnica in the northeast region of Upper Franconia (Bavaria) [31]. Moreover, in the studies conducted by Blachnik and Smaller [31], it was found that arnica rosettes grew slower in drier places. In the present study, the more numerous GF populations were characterized by higher tree cover (mainly *Pinus sylvestris*) and therefore probably a lower exposure of arnica individuals to the action of the sun during the hot summer. It is not excluded that despite the small distance between the AKF and GF populations, climatic conditions can differ and have an impact on arnica population fitness. In future studies, special attention should be paid to the occurrence of sudden weather phenomena (hot and dry summer periods) and their impact on the condition of arnica populations in Poland and Belarus (including geographic and climatic gradients).

Conclusions

The analyzed characteristics of the studied populations indicate the good status of populations located in GF (Belarus). However, the very small number of individuals and the very small proportion of flowering individuals in the populations located in AF and KF (Poland) indicates the need for active protection actions. In the forest arnica populations located in AF and KF, *Calamagrostis arundinacea* can play the role of competitor; therefore, during the planning of active protection, individuals of this species should be eliminated and particular attention should be paid to the frequency and cover of this plant species and height of the herb layer.

The dependence between population characteristics, especially the share of flowering stems and the concentration of available phosphorus, may indicate the effect of the concentration of this macroelement on flowering and, in consequence, provide a greater chance for the generative propagation of this plant species. In the unfavorable and extremely poor habitats of the studied pine forests, phosphorus can play an important and special function in the persistence of arnica individuals and exert an impact on the fitness characteristics of the populations. All actions of active protection should be preceded by an accurate assessment of soil conditions, and it is not excluded that mineral fertilization, as one of the elements of active protection, and P, as an element used to modify and improve the soil conditions, should be taken into account.

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