TREE-LIKE PINES ON THE MSHANA PEAT BOG IN THE GORGANY MOUNTAINS: A TRACE OF *PINUS ULIGINOSA* MIGRATION IN THE EAST CARPATHIANS?

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ABSTRACT

The taxonomic position of the population of tree-like, mostly polycormic individuals of pines from the Mshana peat bog in the Gorgany Mountains (East Carpathians, Ukraine) has been studied on the basis of the morphological characteristics of cones and needles, and anatomical characteristics of the needles. These features have been compared with the surrounding *Pinus mugo* population as well as *P. uliginosa*, *P. mugo*, *P. sylvestris* and *P. uncinata* from natural populations of the taxa. Tree-like individuals were found to have the most similar needles to *P. uliginosa*, but most similar cone characteristics to *P. mugo*. It was concluded, that the tree-like population has a relic character and can present the trace of the early migration of *P. uliginosa* from the West and its hybridisation with *P. mugo*.

KEY WORDS: *Pinus uliginosa*, *P. mugo*, *P. sylvestris*, *P. uncinata*, needle, cone, morphology, variability, taxonomy.

INTRODUCTION

Pinus uliginosa Neumann is a polycormic tree up to about 15-18 m high, with dark bark on the trunk and branches and large, about 3 cm long, asymmetric cones with hooked apophyses. It occurs in several places across the Czech Republic, south-eastern Germany, south-western Poland and northern Austria. Despite investigations on its taxonomic position it remains controversial. It has been treated as a hybrid between *P. mugo* Turra and *P. sylvestris* L. (e.g. Staszkiewicz and Tyszkiewicz 1972; Staszkiewicz 2001) or between *P. mugo* and *P. uncinata* Ramond (Christensen 1987), a synonym of *P. rotundata* Link (Businský 1999) or lately as a subspecies of *P. uncinata* (Businský and Kirschner 2006; Businský 2008). Businský (2008) suggests that the peat bog pine is the subspecies *P. uncinata* subsp. *uliginosa* (Neumann) Businský and this rank of the taxon has been adopted in this study.

The problems with the morphological delimitation of *P. uncinata*, *P. uliginosa*, *P. mugo*, *P. rotundata* and *P. ×rhaetica* Brügger arise mainly from the hybridisation among them in the areas where these taxa occur together. Intensive hybridisation between *P. mugo* and *P. uncinata* takes place in the western Alps, and between *P. mugo* and *P. uliginosa* in the low mountains north of the Alps (Christensen 1987; Christensen and Dar 1997, 2003; Businský 1999, 2008; Boratyńska et al. 2003; Kormuták et al. 2005; Wachowiak and Prus-Głowacki 2008). The latter taxon, *P. uliginosa*, belongs to rare plants connected with central Europe. Most localities of *P. uliginosa* are endangered because of the disappearance of peat bogs, where this species grows (Gołąb 1999; Businský 1999; Danielewicz and Zieliński 2000; Mach et al. 2009; Bastl et al. 2009). This also concerns localities protected in nature reserves or national parks (Businský 1999; Gołąb 1999; Danielewicz and Zieliński 2000; Staszkiewicz 2001). The easternmost isolated localities of *P. uliginosa* have been recorded in the Eastern Carpathians. The occurrence of the species had been described in the Pogórze Przemyskie (Schramm 1925), but two localities in that region have since disappeared

(Schramm 1973; Staszkiewicz 2001). Specimens resembling *P. uliginosa* have also been described in the peat bog near Dolina at the northern base of the Eastern Beskids (Windakiewicz 1873) and in the Kikhola peat bog in the Gorgany Mountains (Trampler 1937), but neither has recently been confirmed (Tsaryk et al. 2006). Finding several tree-like, polycormic individuals of pines resembling *P. uliginosa* in the Mshana peat bog in the Gorgany Mountains (Jasińska et al. 2009), about 350 km south-east of existing localities of the species in the Nowy Targ Basin (Kotlina Nowotarska) shall be treated as very interesting in case of confirmation of their taxonomical position.

The aims of this study were: 1) to verify the influence of the tree-like versus shrubby growth form to the morphological characteristics of cones and morphological and anatomical characteristics of needles; and 2) to test the taxonomic status of the tree-like specimens of mountain pine from the Mshana peat bog using the morphological characteristics of the cones and morphological and anatomical characteristics of the needles (Szweykowski 1969; Staszkiewicz and Tyszkiewicz 1972; Boratyńska and Bobowicz 2000; Boratyńska and Boratyński 2007; Marcysiak and Boratyński 2007; Boratyńska and Lewandowska 2009).

MATERIALS AND METHODS

Materials

The partly drained Mshana peat bog ("Boloto Mshana") in the Gorgany Mountains is located in the valley of the Mshana stream near Osmoloda, at an altitude of 830 m and surrounded by extensive Norway spruce forest with admixture of *Betula pendula* Roth (Table 1). Only 16 tree-like

TABLE 1. Tree-like pines from the Mshana peat bog and comparative material of *P. mugo*, *P. uncinata and P. uliginosa*.

Taxon	Locality	Acronym	Geographic coordinates	Altitude (m)	Subject	Source of data
Pinus	**Ukraine, Carpathians, Gorgany Mts, Osmoloda, Mshana peat bog**	**PN**	**48°40'33"N 23°55'19"E**	**830**	**needles, cones**	**This paper**
Pinus mugo	Ukraine, Carpathians, Gorgany Mts, Osmoloda, Mshana peat bog	M_1	48°40'33"N 23°55'19"E	830	needles, cones	This paper
	Ukraine, Carpathians, Chornokhora Mts, N slopes of Breskulec	M_2	48°06'25"N 24°35'00"E	1650	cones	Marcysiak and Boratyński 2007
	Ukraine, Carpathians, Chornokhora Mts, N slopes of Khoverla	M_3	48°08'00"N 24°37'30"E	1550	cones	Marcysiak and Boratyński 2007
	Poland, Carpathians, Tatry Mts, Dolina Pięciu Stawów	M_4	49°13'09"N 20°03'05"E	1700	needles	Boratyńska et al. 2004
	Poland, Carpathians, Tatry Mts, Grześ-Wołowiec ridge	M_5	49°13'07"N 19°45'50"E	1620	cones	Marcysiak and Boratyński 2007
	Poland, Sudetes, Karkonosze Mts, Czarny Kocioł Jagniątkowski	M_6	50°47'05"N 15°35'30"E	1350	needles	Sobierajska and Boratyńska 2010
Pinus uliginosa	Poland, Sudetes, Karkonosze Mts, Kocioł Małego Stawu	M_7	50°44'41"N 15°47'34"E	1380	cones	Sobierajska and Boratyńska 2010
	Poland, Sudetes, Stołowe Mts, Batorów peat bog	UL_1	50°27'36"N 16°15'25"E	750	needles	Boratyńska et al. 2003
	Poland, Bory Dolnośląskie, Nature Reserve Węgliniec	UL_2	51°17'50"N 16°15'25"E	200	needles, cones	Marcysiak et al. 2003; Boratyńska et al. 2003
	Poland, Bory Dolnośląskie, Węglowiec	UL_3	51°18'61"N 15°11'34"E	200	needles	Sobierajska and Boratyńska 2008; Boratyńska and Lewandowska 2009
Pinus uncinata	Andora, Pyrenees, Vall de Ransol	UN_1	42°37'30"N 01°35'37"E	2025	needles, cones	Boratyńska and Boratyński 2007; Boratyńska et al. 2004
	Spain, Pyrenees, Vall de Nuria	UN_2	42°22'40"N 02°10'49"E	2150	cones	Marcysiak and Boratyński 2007
Pinus sylvestris	Poland, Sudetes, Stołowe Mts, Szczeliniec Wielki Mt.	S_1	50°28'55"N 16°16'46"E	900	needles	Boratyńska and Lewandowska 2009
	Poland, Bory Tucholskie Forest, Krówka	S_2	53°21'42"N 17°52'16"E	90	cones	Marcysiak and Boratyński 2007
	Poland, Bory Dolnośląskie Forest, Węgliniec	S_3	51°18'61"N 15°11'34"E	200	needles	Boratyńska et al. 2003

TABLE 2. Result of Student's *t*-test for characters of needles and cones from two samples from the Mshana peat bog: PN (type *P. uliginosa*) and M_1 (type *P. mugo*).

Character	Arithmetical means		t	df	p
	M_1	PN			
1. Needle length (mm) *	35.65	37.41	-0.83	42	0.412
2. Number of stomatal rows on the convex (abaxial) side of needle	8.34	7.59	2.88	42	0.006
3. Number of stomatal rows on the flat (adaxial) side of needle	5.34	5.08	1.45	42	0.153
4. Number of stomata on a 2 mm long section of needle, on the convex (abaxial) side *	19.00	18.17	2.44	42	0.019
5. Number of stomata on a 2 mm long section of needle, on the flat (adaxial) side *	18.84	17.74	3,37	42	0.002
6. Number of resin canals *	3.49	2.78	3.93	42	0.000
7. Needle width (µm)	1191.75	1221.60	-1.07	42	0.289
8. Needle thickness (µm)	742.53	746.19	-0.23	42	0.819
9. Distance between vascular bundles (µm)	80.78	92.11	-1.28	42	0.207
10. Height (thickness) of epidermial cells (µm)	34.01	34.52	-0.50	42	0.620
11. Width of epidermal with hypodermal cells (µm)	15.69	16.01	-0.91	42	0.366
12. Width of epidermal cells (µm)	23.25	23.98	-0.81	42	0.421
13. Width of hypodermal cells (µm)	10.76	10.60	0.46	42	0.648
14. Number of sclerenchymatic cells layer over the vascular bundles	1.42	0.84	4.28	42	0.000
15. Marcet's coefficient (=traits 9×7/8) *	130.51	151.59	-1.40	42	0.170
16. Stomatal rows ratio (=traits 2/3) *	1.60	1.53	1.60	42	0.118
17. Stomata ratio (=traits 4/5)	1.01	1.03	-1.57	42	0.124
18. Needle thickness/width ratio (=traits 8/7)*	0.62	0.61	1.69	42	0.099
19. Epidermis with hypodermis cells width/thickness ratio (=traits 11/10) *	0.47	0.47	-0.12	42	0.901
20. Epidermis cell width/thickness ratio (=traits 12/10)	0.69	0.68	0.37	42	0.712
21. Character of cells between vascular bundles (%)*					
A – fibre-like cells	4.09	0.98	1.38	42	0.176
B – intermediate, semi-fibrous cells	9.50	17.82	-1.66	42	0.105
C – intermediate	32.24	54.45	-4.80	42	0.000
D – cells with thin walls and large lumens	54.11	26.96	3.73	42	0.001
22. Character of cells around the resin canals (%)*					
A – fibre-like cells	33.19	70.79	-5.44	42	0.000
B – intermediate cells	48.51	28.13	4.73	42	0.000
C – cells with thin walls and large lumens	17.96	1.08	3.25	42	0.002
23. Length of cone (mm)	31.13	30.53	-0.42	42	0.678
24. Maximal diameter of cone (mm)	17.82	19.35	2.63	42	0.012
25. Cone scale number	82.81	86.94	1,58	42	0.120
26. Length of cone scale apophysis (mm)	5.69	5.91	0.88	42	0.386
27. Width of cone scale apophysis (mm)	6.85	6.34	-2.14	42	0.038
28. Thickness of cone apophysis (mm)	1.85	2.26	6.07	42	0.000
29. Distance between umbo and scale top (mm) *	3.01	3.07	0.41	42	0.685
30. Diameter of cone top (mm) *	4.68	4.73	0.34	42	0.739
31. Diameter of cone midpoint between the top and maximal diameter (mm) *	14.20	14.70	1.10	42	0.278
32. Measurement of convex cone side from stalk to top (mm)	45.74	45.64	-0.06	42	0.955
33. Measurement of concave cone side from stalk to top (mm)	40.78	40.57	-0.13	42	0.896
34. Ratio of cone length/maximal diameter (23/24) *	1.74	1.57	-3.76	42	0.001
35. Ratio of cone length/number of scales (23/25) *	0.38	0.36	-1.36	42	0.182
36. Ratio of cone scale apophysis length/width (26/27) *	0.84	0.93	3.77	42	0.001
37. Ratio of cone scale apophysis length/thickness (26/28) *	3.14	2.67	-3.40	42	0.001
38. Cone asymmetry (ratio of convex/concave cone measurements, 32/33) *	1.13	1.14	0.52	42	0.607

*characters used in discriminant and cluster analyses

specimens resembling peat bog pines were found there, surrounded by numerous shrubby individuals, typical of *P. mugo* (Jasińska et al. 2009). Material was collected separately from tree-like and shrubby individuals, the first as tree-like pines (PN) and the second as *P. mugo* (M_1). Ten two-year-old dwarf shoots (brachyblasts) from the central part of a long shoot with undamaged needles and ten mature cones were gathered from each individual according to the methods described earlier (Boratyńska and Bobowicz 2000; Boratyńska et al. 2003). Some tree-like individuals were in a bad condition or had only a few cones. Finally, 14 individuals were represented in needle samples and 15 in cone samples and 140 needles and 86 cones have been gathered. Similarly, following the sampling method described by Boratyńska et al. (2003) altogether, 300 needles and 248 cones were collected from 30 individuals of a *P. mugo* population. Sampled individuals were separated by distances of least 40 m to avoid collecting from the same genet.

The tree-like pines and *Pinus mugo* from the Mshana peat bog were biometrically compared with another populations of *P. mugo* and with closely related *P. uncinata*, *P. uliginosa* and *P. sylvestris* using data from previously published works (source of data in Table 1). The comparative material included: three samples of *P. mugo* needles and five of cones from the Carpathians in Poland and Ukraine and the Sudetes in Poland, three samples of *P. uliginosa* needles and one of cones from the Sudetes and lowland in Poland; one sample of *P. uncinata* needles and two of cones from the Pyrenees in Spain and Andorra; and two samples of *P. sylvestris* needles and one of cones from the Sudetes and lowland in Poland (Table 1).

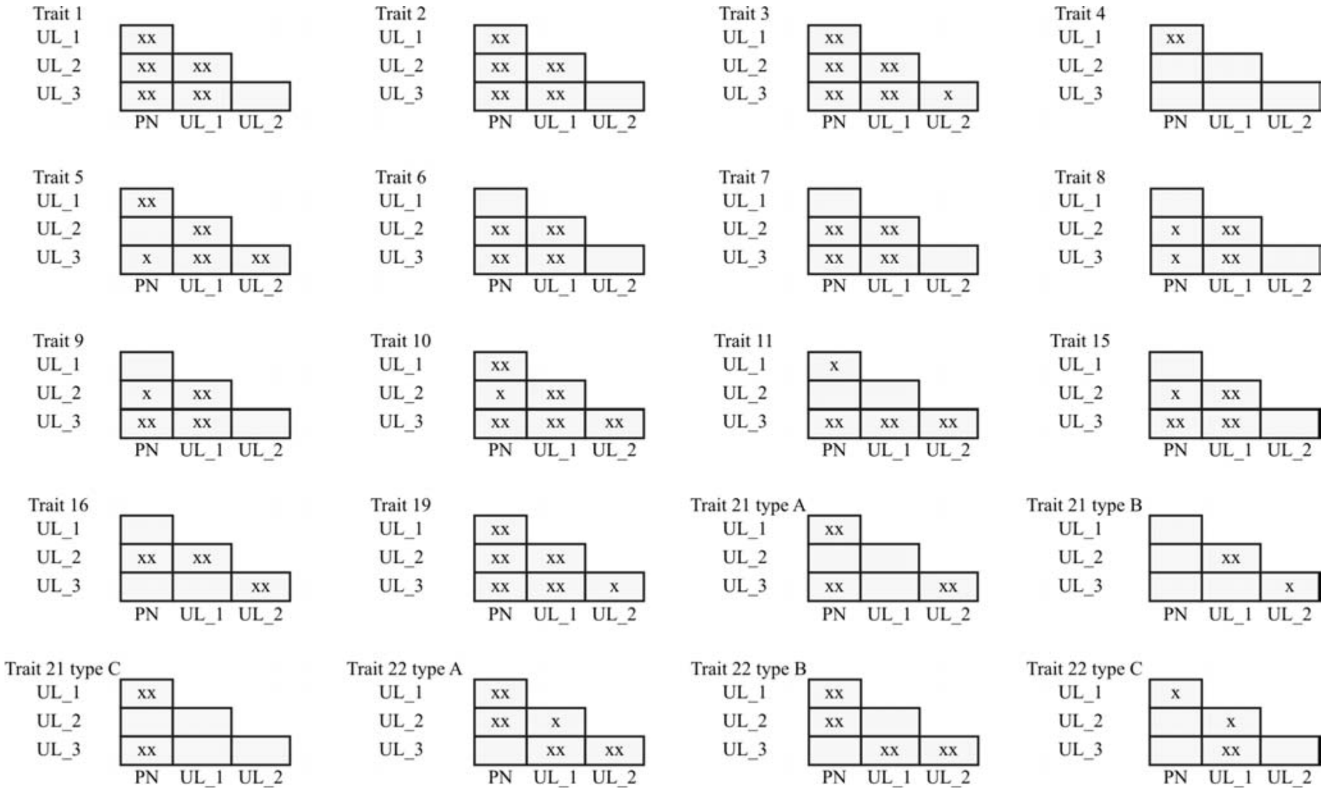

Fig. 1. Student's *t*-test differences between verified tree-like pines from the Mshana peat bog (PN) and three Sudetan populations of *Pinus uliginosa* (UL_1, UL_2 and UL_3, acronyms as in table 1); x – differences statistically significant at p=0.05, xx – differences statistically significant at p=0.01.

Biometrical measurements

The 22 traits of needles and 16 of cones were measured and/or evaluated (Table 2) following procedures and using methods described in previous works (Szweykowski 1969; Staszkiewicz and Tyszkiewicz 1969, 1972; Boratyńska and Bobowicz 2000; Marcysiak et al. 2003; Marcysiak 2004; Marcysiak and Boratyński 2007; Boratyńska and Boratyński 2007; Boratyńska et al. 2009).

Statistical treatment

The arithmetic means, standard deviations and coefficients of variation were calculated and analysed. The values of particular characters between the two samples from the Mshana Peat-bog in the Gorgany Mountains were compared using the Student's *t*-test for independent samples to testify the differences between them. Discriminant analysis was performed and the position of individuals from the particular subpopulations and samples from the Sudetes and Carpathians was examined along the first two discriminant variables to show the intra- and interpopulational variation (Sokal and Rohlf 2003; Stanisz 2007). Cluster analysis based on the shortest Euclidean distances, following the unweighted pair group average (UPGMA), was also used to verify the findings (Sokal and Rohlf 2003). The distribution of every trait was checked using the Shapiro-Wilks test and those characterised with biased distribution were transformed to fulfil the demands of multivariate analyses. The values of every trait were standardised prior to analyses. The percentage data were arcsine transformed prior to statistical analyses (Watała 2002). STATISTICA PL 7.0 for Windows (StatSoft) was used in the calculations.

RESULTS

Tree-like pines versus Pinus mugo from the Mshana peat bog

The traits of the tree-like pines (PN) and *P. mugo* (M_1) on the Mshana peat bog in the Gorgany Mountains differ at statistically significant level in respect to several traits (Table 2). The highest differences were found in the number of the strata of sclerenchyma cells above the phloem on the needle cross-section (character 14). Statistically significant

TABLE 3. Result of Student's *t*-test for cone characters of tree-like pines from the Mshana peat bog (PN) and *Pinus uliginosa* from Węgliniec Nature Reserve (UL_2).

Character	Arithmetical means		t	df	p
	UL_2	PN			
23	33.26	30.53	1.90	59	0.063
24	16.85	19.35	-4.19	59	0.000
25	75.35	86.94	-4.29	59	0.000
26	6.84	5.91	3.31	59	0.002
27	7.03	6.34	3.36	59	0.001
28	3.71	2.26	6.48	59	0.000
29	4.37	3.07	5.89	59	0.000
30	3.35	4.73	-9.64	59	0.000
31	12.31	14.7	-4.61	59	0.000
32	48.47	45.64	1.53	59	0.131
33	34.75	40.57	-3.55	59	0.001
34	1.99	1.57	8.30	59	0.000
35	0.45	0.36	5.06	59	0.000
36	0.98	0.93	1.38	59	0.174
37	1.98	2.67	-4.52	59	0.000
38	1.42	1.14	6.26	59	0.000

TABLE 4. Discriminant power testing for needle characters of tree-like pines from the Mshana peat bog, *P. mugo*, *P. uliginosa*, *P. uncinata* and *P. sylvestris*.

No	Character	Partial Wilks' lambda	*F* statisti	*P* value
1	Needle length	0.6580	16.22	0.0000
4	Number of stomata on a 2 mm long section of needle, on the convex (abaxial) side	0.9025	3.37	0.0006
5	Number of stomata on a 2 mm long section of needle, on the flat (adaxial) side	0.8941	3.70	0.0002
6	Number of resin canals	0.8178	6.96	0.0000
15	Marcet's coefficient (=traits 9×7/8)	0.8965	3.60	0.0003
16	Stomatal rows ratio (=traits 2/3)	0.6043	20.44	0.0000
18	Needle thickness/width ratio (=traits 8/7)	0.7437	10.76	0.0000
19	Epidermis with hypodermis cells width/thickness ratio (=traits 11/10)	0.4087	45.18	0.0000
21A	Percentage of fibre-like cells between vascular bundles	0.7426	10.82	0.0000
21B	Percentage of intermediate, semi-fibrous cells between vascular bundles	0.6889	14.10	0.0000
21C	Percentage of intermediate cells between vascular bundles	0.7943	8.09	0.0000
21D	Percentage of cells with thin walls and large lumens between vascular bundles	0.7730	9.17	0.0000
22A	Percentage of fibre-like cells around resin canals	0.7460	10.63	0.0000
22B	Percentage of intermediate cells around resin canals	0.7000	13.40	0.0000
22C	Percentage of cells with thin walls and large lumens around resin canals	0.6562	16.36	0.0000

differences were also found in the number of resin canals, the numbers of stomata and stomata rows and percentages of particular types of sclerenchymatic cells between vascular bundles and around resin canals (characters 6, 5, 2, 21C, 21D and 22A, 22B and 22C, respectively). Among the analysed characters of cones the thickness of the apophysis of cone scale (character 28) revealed the biggest differences. The cones from tree-like individuals with thicker apophyses were more rounded and showed a larger diameter (characters 36, 37, 34 and 24, respectively) than those of *P. mugo* (Table 2).

Tree-like pines from the Mshana peat bog versus P. uliginosa from Poland

Most of the tested characters of needles and cones from the tree-like pines from the Mshana peat bog differed from all three populations of *Pinus uliginosa* from Poland (Fig. 1). Only needle shape and percentage of thin walled cells between the vascular bundles (characters 18 and 21D) do not differentiate them. Insignificant differences were also found in the number of stomata on the convex needle side (character 4), but only between the tree-like pine from the Mshana peat bog and *P. uliginosa* populations from the Batorów peat bog (populations PN and UL_1) (Fig. 1).

The cones from the tree-like pines from the Mshana peat bog in the Gorgany Mountains (PN) did not differ from the cones of the *P. uliginosa* population from the Węgliniec Nature Reserve (the only cones available) in respect to cone length, measurement of cone convex side from stalk to top and shape of cone scale apophysis (characters 23, 32 and 36, respectively). All other characters differed significantly from the characters of compared populations. The tree-like pines from the Gorgany Mountains had cones with a higher number of cone scales (character 25), whereas the scale dimensions (characters 26, 27 and 28) were significantly smaller than in Bory Dolnośląskie. Additionally, the cones of tree-like population from the Gorgany were thinner (characters 24, 30 and 31), more rounded (character 34) and less asymmetric (character 38) than the population of *P. uliginosa* (Table 3).

Tree-like pines from the Mshana peat bog versus P. uncinata, P. mugo, P. uliginosa and P. sylvestris

Discriminant analysis on the 10 needle characters (Table 2) of tree-like pines from Mshana peat bog in the Gorgany Mountains and the three populations of *P. mugo* (including the population of shrubby individuals from the Mshana peat bog), the three populations of *P. uliginosa*, one of *P. uncinata* and two of *P. sylvestris* showed that the shape of epidermis and hypodermis cells, stomatal row ratio, percentage of sclerenchyma cells with thin walls around resin canals and needle length (characters 19, 16, 22C and 1, respectively) differed at the highest level (Table 4).

The shape of epidermis and hypodermis cells, percentage of fibre-like cells between vascular bundles and number of resin canals (characters 19, 21A and 6, respectively) determined at the highest level of the first discrimination variable (U_1), which was responsible for almost 61% of the total variation. This differentiated the needles of *P. sylvestris* from the needles of *P. mugo*, *P. uncinata* and *P. uliginosa* (Fig. 2A). The second discriminant variable, responsible for only 15% of the variation, was mostly determined by the percentage of thin walled cells around the resin canals, the shape of the needle cross-section and shape of epidermis cells (characters 22C, 18 and 19, respectively). The U_2 discriminant variable differentiated among populations of *P. uliginosa*, *P. uncinata* and *P. mugo*. Tree-like pines population from Mshana peat bog (PN) seemed closest to the group formed by all *P. uliginosa* populations (UL_1 – UL_3), but, surprisingly, the population of *P. mugo* from Mshana (M_1) also entered this group (Fig. 2A). The latter population is characterised additionally by a broad scale of variation overlapping part of the ranges of the variation of *P. mugo*, *P. uncinata* and *P. uliginosa* (Fig. 2A).

The UPGMA dendrogram constructed on the shortest Euclidean distances between the centroids of the compared populations confirmed the close connection between tree-like pines and shrubs of *P. mugo* from the Mshana peat bog and their affinities to *P. uliginosa*, but not to *P. mugo* (Fig. 2B). It also confirmed the largest distance of *P. uncinata* from all other taxa of the *P. mugo* complex as well as the separateness of the *P. sylvestris* populations.

Discriminant analysis of eight cone characters (Table 2) of tree-like pines from Mshana peat bog in the Gorgany Mountains and five populations of *P. mugo* (including the population of shrubby individuals from the Mshana peat bog), one population of *P. uliginosa*, two of *P. uncinata* and one of *P. sylvestris* showed that the distance between the top of cone scale and umbo, cone top diameter and shape of apophysis (characters 29, 30, 36 and 37, respectively)

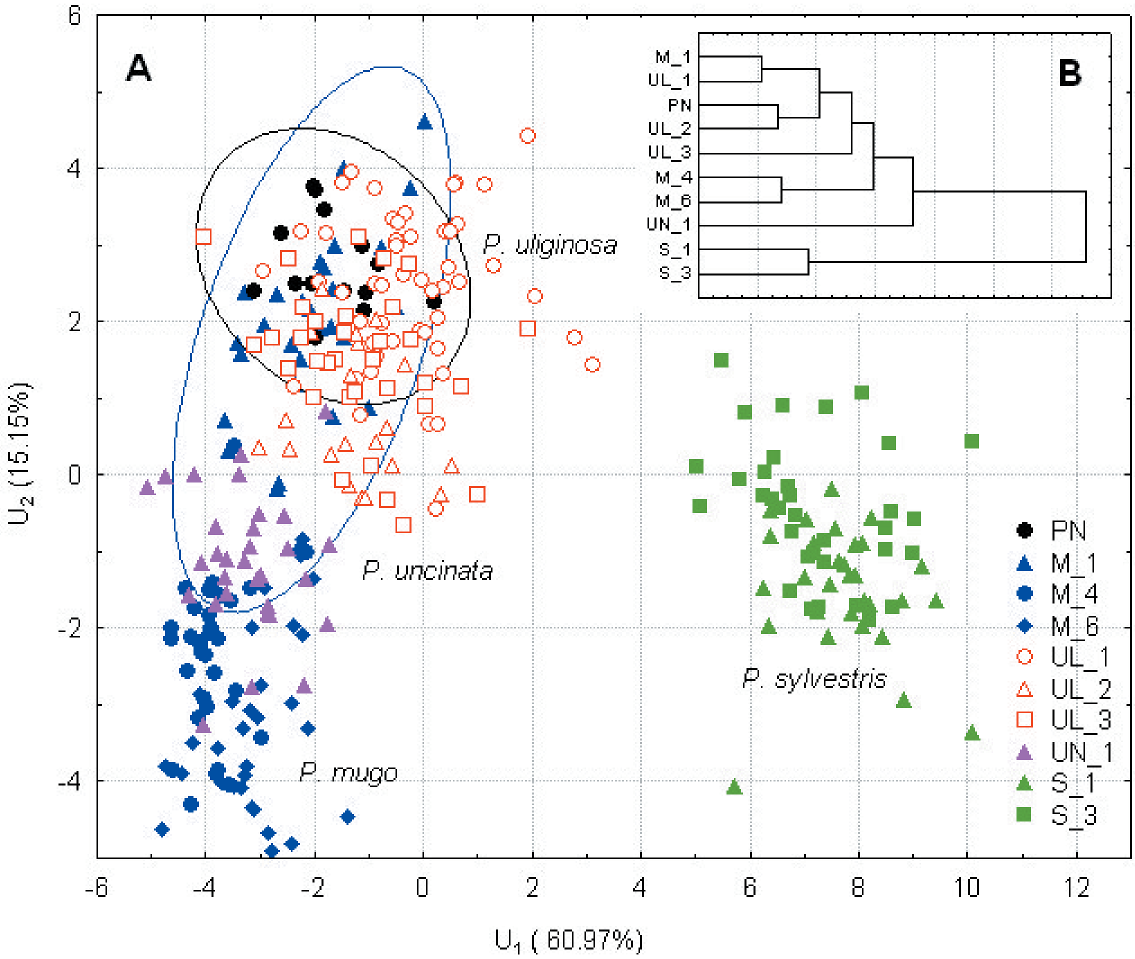

Fig. 2. Position of tree-like pines from the Mshana peat bog (PN) among the compared *P. sylvestris* (S_1 and S_2), *P. mugo* (M_1, M_4 and M_6), *P. uliginosa* (UL_1, UL_2 and UL_3) and *P. uncinata* (UN_1) populations on the scatter-plot showing results of discriminant analysis (A) and on the dendrogram constructed on the shortest Euclidean distances according to Ward's method (B), based on the needle characters.

differentiated among them at the highest level (Table 5). All other characters discriminated among the analysed taxa and populations at a somewhat lower but still highly statistically significant level.

The two populations of *Pinus uncinata* were clearly distinct from the others, according to the space determined by the first two discriminant variables U_1 and U_2 (Fig. 3A), which were responsible for about 74% of the total variation. Both populations of *P. uncinata* were separated by the first variable, which covers more than 56% of the variation. This was determined mostly by distance from the cone scale top to umbo, cone diameter at the midpoint between the top and maximal diameter and cone asymmetry (characters 29, 31 and 38, respectively). The second discriminant variable U_2 was responsible for more than 17% of variation and was determined mostly by cone diameter at the top and at the midpoint between the top and maximal diameter than by cone and cone scale apophysis or shape and ratio of cone length/number of cone scales (characters 30, 31, 34, 36 and 35, respectively). The second discriminant variable differentiated *P. uliginosa* (UL_3), *P. sylvestris* (S_2) and *P. mugo* (M_1, M_2, M_3, M_5 and M_7). The population of tree-like pines from the Mshana peat bog seemed most close to the populations of *P. mugo* from the same place than to other *P. mugo* populations, and not to *P. uliginosa* (Fig. 3A).

This result is confirmed by UPGMA dendrogram constructed on the Euclidean distances from the centroids of

TABLE 5. Discriminant power testing for cone characters of tree-like pines from the Mshana peat bog, *P. mugo*, *P. uliginosa*, *P. uncinata* and *P. sylvestris*.

No	Character	Partial Wilks' lambda	*F* statisti	*P* value
29	Distance between umbo and scale top	0.4399	53.06	0.0000
30	Diameter of cone top	0.5542	33.51	0.0000
31	Diameter of cone midpoint between the top and maximal diameter	0.6063	27.06	0.0000
34	Ratio of cone length/maximal diameter (23/24)	0.6868	19.01	0.0000
35	Ratio of cone length/number of scales (23/25)	0.6256	24.94	0.0000
36	Ratio of cone scale apophysis length/width (26/27)	0.5866	29.34	0.0000
37	Ratio of cone scale apophysis length/thickness (26/28)	0.6041	27.31	0.0000
38	Cone asymmetry (ratio of convex/concave cone measurements, 32/33)	0.6393	23.51	0.0000

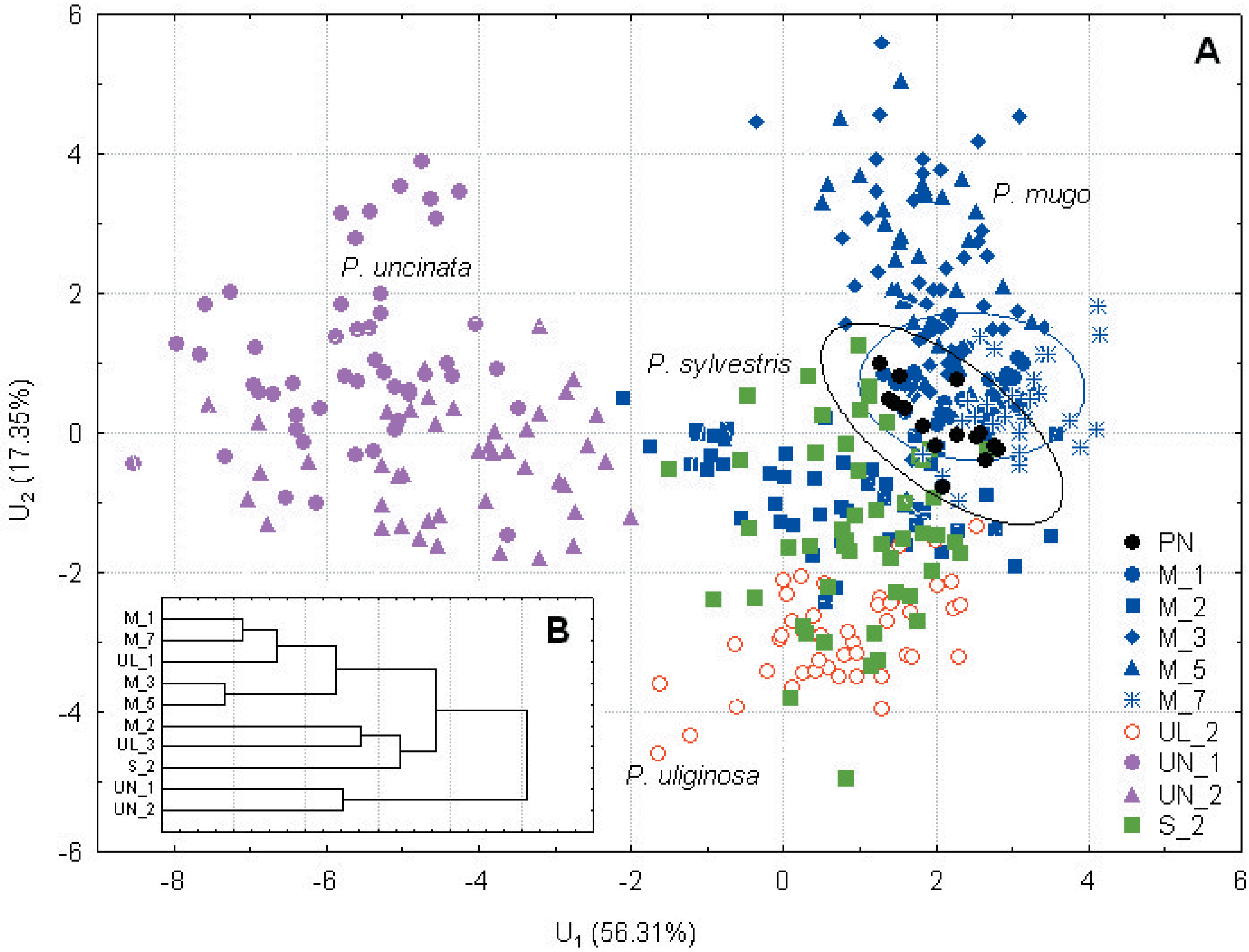

Fig. 3. Position of tree-like pines from the Mshana peat bog (PN) among the compared *P. sylvestris* (S_2), *P. mugo* (M_1, M_2, M_3, M_5 and M_7), *P. uliginosa* (UL_1 and UL_3) and *P. uncinata* (UN_UN_1 and UN_2) on the scatter-plot showing results of discriminant analysis (A) and on the dendrogram constructed on the shortest Euclidean distances according to Ward's method (B), based on the cone characters.

populations. The populations of *P. uncinata* were separated from all other taxa, but were also not closely related to each other (Fig. 3B). Tree-like pines from the Mshana peat bog were placed among *P. mugo* populations, close to the population of *P. mugo* from the Mshana peat bog. *P. uliginosa* seemed similar to the population of *P. mugo* from the East Carpathians, and *P. sylvestris* from the lowland of Poland is also similar to *P. mugo*/*P. uliginosa*.

DISCUSSION

The needles of tree-like pines and *P. mugo* from the Mshana peat bog in the Gorgany Mountains differed significantly on several characteristics, including number of stomata rows on the convex (abaxial) side of the needle, number of stomata rows on the flat (adaxial) side of the needle, number of resin canals, number of sclerenchyma cell layers over the vascular bundles, percentage of intermediate and thin walled sclerenchyma cells between the vascular bundles and the characteristics of the sclerenchyma cells around the resin canals (characters 2, 5, 6, 14, 21C, 21D, 22A, 22B and 22C, respectively). All other analysed characters showed no differences between the compared populations (Table 2). In the Sudetes, needle length and width, number of stomata rows on the flat (adaxial) side of the needle, needle thickness, distance between vascular bundles, thickness of epidermal cells, stomatal rows ratio, needle thickness/width ratio, epidermis cell width/thickness ratio and percentages of all types of sclerenchyma cells around the resin canals and between vascular bundles (character 1, 7, 3, 8, 9, 10, 16, 18, 20 and 21A, 21B, respectively) also revealed statistically significant differences between *P. mugo* and *P. uliginosa* (Boratyńska et al. 2003; Boratyńska and Boratyński 2007; Boratyńska and Lewandowska 2009). This indicates a higher level of similarity between tree-like pines and shrub of *P. mugo* from the Mshana peat bog, than of *P. uliginosa* and *P. mugo* in the Sudetes. This was also visible in the discrimination analysis (Fig. 2A) and UPGMA dendrogram (Fig. 2B). Even smaller differences in cone characteristics were found between tree-like pines and shrubs of *P. mugo* from the Mshana peat bog (Fig. 3A and 3B). Additionally, the range of variations in needle and cone characters of tree-like pines population is much more restricted compared with the variation of the other taxa (Fig. 2A and 3A). The 95% confidence intervals of dispersion of tree-like individuals from the Mshana peat bog based on the characters of needles (Fig. 2A) and cones (Fig. 3A) are about a half as broad as for the other populations compared.

The restricted range of variation of tree-like pine population might indicate an ancient founder effect. A reduction of differences between this population and nearby *P. mugo* might be because of gene flow from the latter species. However, this hypotheses will need to be verified using molecular methods, if markers that allow us to distinguish between *P. mugo* and *P. uliginosa* could be found.

Several characters of needles of tree-like individuals from the Gorgany Mountains have values typical for *P. uliginosa*, intermediate values of the character among all three compared populations of the latter species or are close to one of them (compare Table 2 and data from Boratyńska et al. 2003; Boratyńska and Boratyński 2007; Boratyńska and Lewandowska 2009). The length, width and thickness of needles and distance between vascular bundles (characters 1, 7, 8 and 9, respectively) have values intermediate among the three populations of peat bog pine, which can be treated as typical for *P. uliginosa*. The percentages of particular types of sclerenchyma cells between vascular bundles were also similar to those observed in *P. uliginosa*, whereas around the resin canals in the population of tree-like individuals a higher frequency of fibre-like cells were observed (compare Table 2 and data from Boratyńska and Boratyński 2007; Boratyńska and Lewandowska 2009).

Cones of tree-like individuals from the Ukraine are more symmetrical than cones of *P. uliginosa* from the Węgliniec nature reserve and have a higher number of scales with thicker, longer and wider apophyses (Marcysiak et al. 2003). The cones collected from the Mshana tree-like individuals were more similar to cones of *P. mugo* than to cones of *P. uliginosa*.

The analyses performed on cone characters did not allow to include the investigated tree-like individuals of pines in the Gorgany Mountains into taxon of *P. uliginosa*. Their origin from the ancient hybridisation between *P. mugo* and *P. sylvestris* should be also excluded. The characters of needles place them among *P. uliginosa* and close to *P. mugo*, but far from *P. sylvestris*. The closest connections of the examined tree-like individuals with *P. uliginosa* regarding needle characters suggest their identity with the latter taxon, but the characters of cones are more similar to cones of *P. mugo*. All these intermediate characteristics can be interpreted as a trace of ancient migration of *P. uliginosa* and then hybridisation with *P. mugo*. The latter process reduced expression of some morphological characters, conserving another, similarly as in the case of *P. uncinata* in the Giant Mountains (Boratyńska et al. 2009). It is note worthy, that mountain pine with hooked apophyses has been recorded from the Gorgany Mountains by Trampler (1937), who named them "*Pinus uncinata rotundata gibba* Wilk." (Trampler 1937: 33-34). These data have not been confirmed (Tsaryk et al. 2006), but the genes of *P. uliginosa* can be conserved in their progeny. The tree-like growth form and characters of the needles of some number of mountain pine on the Mshana peat bog indicate an influence of the genes of latter taxon, however, it can not be determined as typical *P. uliginosa*. The locality on the Mshana peat bog can be treated as relict from the time of the end of last glaciation and early Holocene (Obidowicz 1996).

The possibility of migration of mountain species, to which *P. uliginosa* can be included, from the West to East Carpathians has been confirmed by verifying the DNA variation of *Abies alba* (Gömöry et al. 2004; Liepelt et al. 2009).

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