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Differentiation of *Pinus sylvestris* populations of the Tatra Mts and the Tuchola forest expressed in the needle anatomical traits

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Abstract: The differentiation among three populations of *Pinus sylvestris* of the Tatra National Park (TNP); Łysa Skała, Siwarowe Pańskie and Wielkie Koryciska has been investigated with regard to eight anatomical traits of needles. These populations were compared to a lowland population of Tuchola Forests. Five analyzed traits: the thickness of epidermis with cuticle, the mean of width of three epidermis cells, the width of needle cross-section, the height of a needle cross-section and the ratio of height to the width of a needle are characterized by a variation coefficient not exceeding 10%. Variation coefficients for the remaining traits: the number of resin canals, the distance between vascular bundles and a Marcet coefficient, range between 15.04 – 24.95%. The needles of the lowland population (Rzepiczna) turned out to be the widest as well as the thickest and with the biggest distance between vascular bundles. The Tatra populations of *P. sylvestris* have a considerably thicker cuticle together with epidermis compared with the lowland population of Tuchola Forests. None of the individual traits differentiate the populations which have already been regarded as distinct – Łysa Skała, found in the eastern part of the TNP and Wielkie Koryciska, from its western part. However, having carried out a discriminant analysis and Mahalanobis distances, the differentiation of the Tatra Scots pine in relation to its geographical location was reported. The individual character of *P. sylvestris* found in the eastern and western parts of the Tatra National Park might be a result of their different origin in the last glacial period.

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Introduction

The differentiation of *Pinus sylvestris* L. has been examined many times and in several aspects; biochemical, genetical, morphological and anatomical. In many works authors suggested that a profound influence on a present-day differentiation has the latest Ice Age. The populations of Scots pine were migrating and at the same time exchanging genes with one another. (Giertych 1979; Giertych and Oleksyn 1981; Staszkiewicz 1961a, b, 1963, 1993a, b; Latałowa et al. 2004). The plasticity of pine enabled it to settle in various locations (Obmiński 1970; Mamaev 1972; Przybylski 1993; Pravdin 1964; Hantley and Birks 1983; Giertych 1993; Urbaniak et al. 2003; Woźniak et al. 2005). Wright and Bull (1963) claim that the today's differentiation is a consequence of the isolation of populations of different origin in the glacial period, which eventually led to their different migrating directions in post-glacial period. On the other hand, Langlet (1959) taking into consideration adaptive abilities of pine to local conditions emphasizes a clinal variability of many traits mainly due to the impact of a climate. In the glacial period one of the most important refuges of the Scots pine was in the south of Europe, where a great interpopulational variability is observed (Staszkiewicz 1961b, 1963, 1968, 1993a, b; Prus- Głowacki et al. 2003; Cheddadi et al. 2004). It is believed, that the isolation of particular populations in the most elevated mountains over at least 150–200 generations, can be a reason for divergent migration routes from different glacial refuges (Wright and Bull 1963; Ruby and Wright 1976; Birks 1989; Berglund et al. 1996). In the case of small isolated populations, such as the ones in Tatra Mountains, the factor which influences the variability may even be a stochastic phenomenon – genetic drift, and the effects of local adaptations (Mitka 1997). *Pinus sylvestris*, a dominant ingredient of forest which occurs in the lowlands of Poland, in the south mountainous regions and foot-hills is significantly rare and replaced by the spruce (Białobok 1970). In the Tatra Mountains *Pinus sylvestris* forms small populations most often on rocks facing south (Łysek 1974; Zwijacz-Kozica 1998).

These relict populations possess many distinctive features concerning the morphology of cones, seeds and needles (Zajączkowski 1949; Staszkiewicz 1993b). They also have characteristic traits related to the increment and the duration of the vegetation period, height, vitality, stem, crown and bark types (Zajączkowski 1949; Skrzyszewski 2004). Staszkiewicz (1961a, b, 1993a, b) on the basis of the morphological structure of cones, distinguishes two different types of the Scots pine located in different regions of the Tatra National Park; *meridionalis* type found in Wielkie Koryciska in the western part; and a slightly changed *polonica*.. Also, morphological traits of needles turned out to be essential for the studies on regionalization of populations in the Tatra Mountains (Urbaniak et al. 2001; Androsiuk and Urbaniak 2005).

In the present study the following questions were addressed: (1) do populations from various regions of the Tatra Mountains have characteristic and contrastive anatomical features of their needles; (2) can these populations be grouped according to their geographical location on the basis of the anatomical traits of needles; (3) does the comparison of populations from the Tatra National Park with the lowland Scots pine native to Tuchola Forests enable us to find needles characteristics typical of mountain populations. (4) whether the reason for differentiation within populations is their adaptability which can be observed when different needle traits are being examined.

Material and methods

Plant material

Three of the analyzed populations were sampled in the Tatra National Park – Łysa Skała (49°15'N; 20°06'E) (1) – 24 investigated trees – in the eastern part, Siwarowe Pańskie (49°16'N; 19°56'E) (2) – 26 – in the central part, Wielkie Koryciska (49°15'N; 19°48'E) (3) – 25 – in the western part. The fourth population, Rzepiczna (4), is represented by 30 trees from the vicinity of the village Rzepiczna (53°43'N; 18°02'E) located in the Tuchola Protected Landscape Area (Fig. 1). A total 105 individuals were sampled in October 2003, each by collecting of 10 long-shoot, from the central part of the tree, from the crown edge and the southern part of crown. The investigated material from each tree consists of five two-year old needles coming from different dwarf shoots. The cross-sections of needles were performed in the middle of their length and preserved in polyvinyl alcohol. Altogether, sections of 525 needles were investigated. Eight anatomical characteristics of the needles were tested (Table 1). The usefulness of these traits was examined as suitable for the study on variability in many works (e.g. Bobowicz 1990; Bobowicz and Korczyk 1994; Urbaniak 1998).

Statistics

The results of the measurements of anatomical characteristics were subjected to statistical analysis with Statistica 7.1 software (StatSoft Inc.). Descriptive statistics (arithmetic means, standard deviations, minimum and maximum values of characters) and variation coefficients were computed to evaluate the range of variation of anatomical traits (Williams 1995; Łomnicki 2000; Watała 2002). The analysis included basic descriptive statistics for each population: means, minima, maxima, standard deviations, variation coefficients as well as the skewness and curtosis; Pearson's correlation coefficients; Tukey's test; discriminant analysis, determination coefficients of analyzed traits and discriminant variables, values of the F statistics and Mahalanobis distances (Krzyśko 1982, 1990; Urbaniak 1998; Ferguson and Takane 2007).

Results

Basic statistics. In the investigated populations, five traits: the thickness of epidermis with cuticle (b), the mean width of three epidermis cells (c), the width of needle cross-section (d), the height of needle cross-section (e), the ratio of height to the width of needle (f) have low variation coefficients not exceeding 10%. The lowest variation coefficient ranging between 4.62 and 6.09% was reported for the ratio of height to the width of needle (trait f). Three traits: the

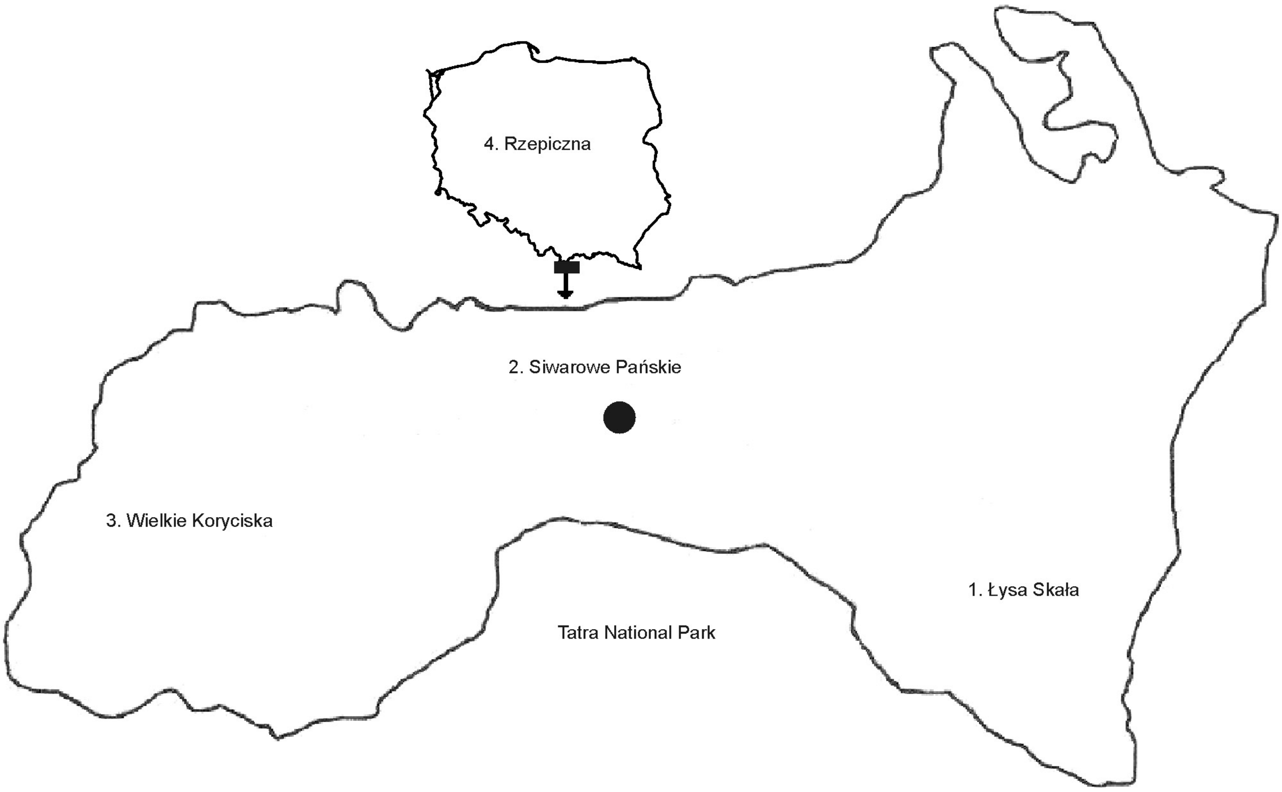

Fig. 1. Geographical location of studied populations from the Tatra National Park (1, 2, 3) and the Tuchola Forest (4)

number of resin canals (a), the distance between vascular bundles (g) and Marcet coefficient, trait d × g/e (h) are characterized by high variation, which is indicated by considerably high variation coefficient ranging 15.04 – 24.95% (Table 2).

Tukey's test. Tukey's test was conducted to determine statistically significant traits which make the populations different (Table 3). Population Rzepiczna (4) is statistically distinguished by the lowest number of resin canals (trait a) and the thinnest epidermis together with the cuticle (b). Moreover, it possesses the smallest width of three cells of the epidermis on the flat side of a needle (c). This trait makes it different from the other two populations of the Tatra Mountains, namely Łysa Skała (1) and Siwarowe Pańskie (2), but not from population Wielkie Koryciska (3). The population from the central part of the TNP Siwarowe Pańskie (2) differs from the population from the west part of the TNP Wielkie Koryciska (3) in regard to the mean width of three epidermis cells (trait c). Population Siwarowe Pańskie (2) has the narrowest needles (d) and significantly differs from population Rzepiczna (4), which has the widest needles (d). In addition, the ratio of needle thickness to its width, which value is the lowest in the case of population Rzepiczna (4) and the highest in case of population Siwarowe Pańskie (2), makes those populations different. Populations Łysa Skała (1) and Siwarowe Pańskie (2) have the smallest distance between vascular bundles (g) and are different from the population of the lowlands Rzepiczna (4) which shows the greatest distance between them. This trait distinguishes the Tatra Mountains popula-

Table 1. Needle characters analyzed

a	number of resin canals
b	thickness of epidermis with cuticle, μm
c	mean width of three epidermis cells, μm
d	width of needle cross-section, μm
e	height of needle cross-section, μm
f	ratio of height to width of needle (ratio of trait e to d)
g	distance between vascular bundles, μm
h	Marcet coefficient (trait d × g/e)

Table 2. Variability coefficient (V%) for eight (a –h) anatomical needle traits (a-h as in table 1)

Traits	Łysa Skała V%	Siwarowe Pańskie V%	Wielkie Koryciska V%	Rzepiczna V%
a	15.04	22.27	19.16	24.57
b	7.12	8.64	6.94	8.64
c	6.73	5.23	7.85	6.02
d	7.38	6.35	6.72	8.76
e	6.33	6.95	7.04	9.27
f	6.09	4.62	5.54	4.83
g	20.98	21.77	20.60	21.87
h	24.95	24.26	23.72	23.07

Table 3. The results of Tukey's test. Populations; Łysa Skała (1), Siwarowe Pańskie (2), Wielkie Koryciska (3), Rzepiczna (4). Traits; a–f (name of traits as in Table 1)

	1/2	1/3	1/4	2/1	2/3	2/4	3/1	3/2	3/4	4/1	4/2	4/3
a	0.21407	0.75878	0.00014*			0.00141*		0.75790	0.00016*			
b			0.00014*	0.99854	0.99994	0.00014*	0.99963		0.00014*			
c		0.39095	0.01780*	0.43206		0.00018*		0.01092*	0.47727			
d	0.06708	0.86413						0.30352		0.457897	0.000547*	0.10553
e	0.70923	0.99998	0.48388					0.72238			0.05154	0.43928
f			0.81283	0.18453	0.73712	0.01616*	0.73400		0.21360			
g	0.33874						0.28533	0.00344*		0.01607*	0.00016*	0.58286
h	0.22402						0.53949	0.00657*		0.01656*	0.00015*	0.32011

* – significant value at the level of 0.05

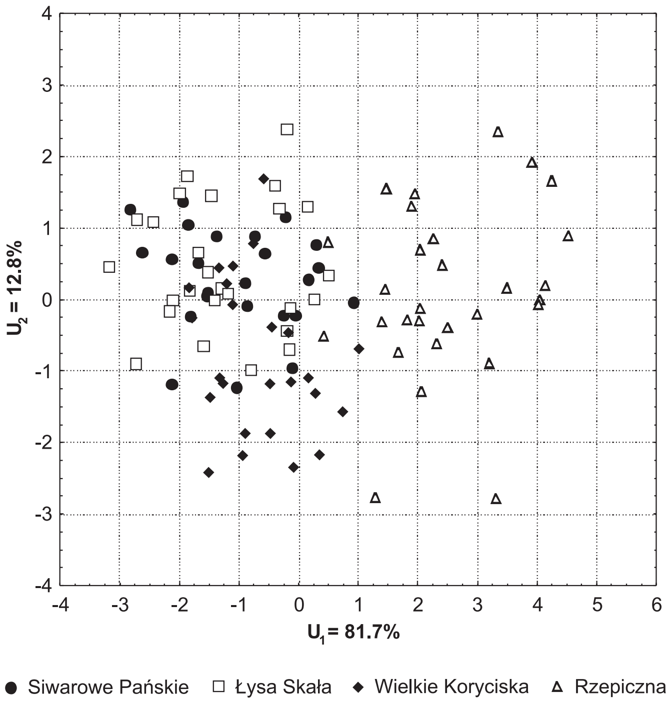

Fig. 2. Results of discriminant analysis of studied individuals on the plane of first two discriminant variables (U_1, U_2) containing altogether 94.5% information of the applied set of eight needle traits

tions Siwarowe Pańskie (2) and Wielkie Koryciska (3). As in the case of trait g (the distance between vascular bundles), identical dependences are observed for trait h (Marcet coefficient).

Discriminant analysis and determination coefficients between eight anatomical needle traits and the first three discriminant variables U1, U2 and U3. The first variable, carrying almost 81.7% of information, determined mostly by the thickness of epidermis together with cuticle (b), Marcet coefficient (h), the distance between vascular bundles (g) and the width of three epidermis cells (c) determines the distinct character of the lowland population Rzepiczna (4) (Fig. 2, Table 4). The distinct nature of western Wielkie Koryciska (3) and eastern Łysa Skała (1) Scots pine populations of the Tatra Mountains depends on the second discriminant variable, which contains 12.8% of information and is mostly determined by the distance between vascular bundles (g), the trait h (Marcet coefficient) and the width of epidermis cells (c) (Table 4). Traits: d (the width of needle cross-section), f (the ratio of height to the width of the needle, the ratio of trait e to d) and h (Marcet coefficient) are the main determinants of the third variable (U3) (containing only 5,44% information), which distinguishes the population of the east part of the TNP Łysa Skała (1) from the population of the central part of the TNP Siwarowe Pańskie (2) (Fig. 3 and Table 4).

Mahalanobis distances with F statistics. These statistics were used to demonstrate the evident differences between the populations from the lowlands Rzepiczna (4) and other three populations from the Tatra Mountains. The population from the western part of the Tatra National Park, i.e. Wielkie Koryciska (3), differed significantly from the population from the

Table 4. The determination coefficients between discriminant variables U1, U 2 and U3 and eight anatomical traits of needles (name of traits as in Table 1)

Traits	U1 81,71%	U2 12,84%	U3 5,44%
a	2,78	0,09	0,15
b	30,34	1,07	0,29
c	10,75	2,20	0,43
d	7,41	0,00	2,22
e	3,47	0,02	0,30
f	3,95	0,07	1,56
g	11,30	2,96	0,57
h	13,79	2,52	1,51

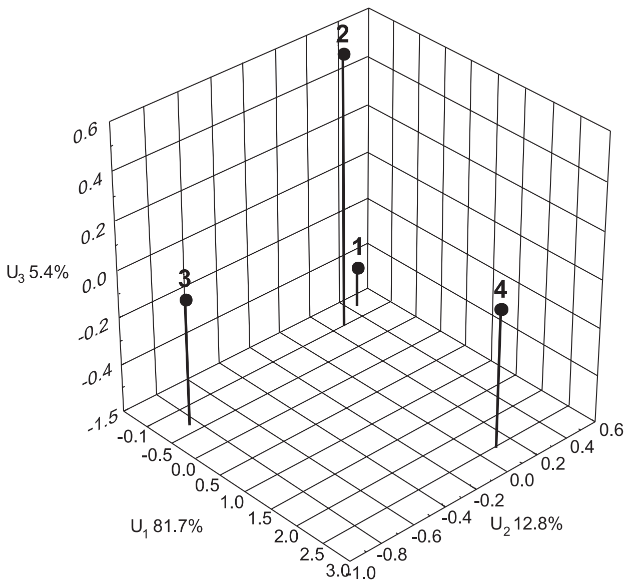

Fig. 3. Result of discriminant analysis of four populations from the Tatra Mountains and Tuchola Forest In the space of the first three discriminant variables containing 100% information of the applied set of 8 needles traits

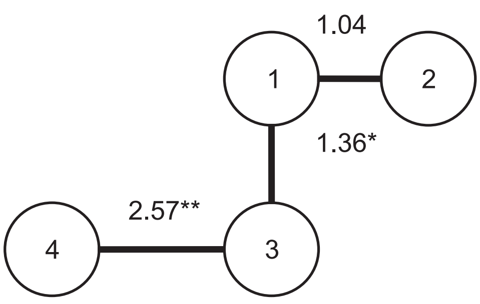

Fig. 4. Minimum spanning tree of four population of *P. sylvestris* constructed on the basis of the Mahalanobis distances (significant distances at the level 0.05*, 0.01**). Samples numbers as in Figure 1

Table 5. Table of Mahalanobis distances with values of F statistics (below), the critical values are mark by *(p<0.05), **(p<0.01). Samples numbers as in Fig. 1

2	1.04		
	1.509		
3	1.36*	1.53**	
	2.516	3.327	
4	2.74**	3.15**	2.57**
	11.216	15.540	10.056
	1	2	3

Tables 6.1–6.4. Correlation matrix between eight anatomical needle traits (name of traits as in Table 2)

6.1. Łysa Skała

b	0.03						
c	–0.15	0.33					
d	**0.58***	0.24	0.01				
e	**0.54***	0.11	0.00	**0.64***			
f	–0.17	–0.18	–0.01	**–0.59***	0.23		
g	0.34	0.02	0.06	**0.82***	0.39	**–0.62***	
h	0.30	0.05	0.06	**0.81***	0.25	**–0.74***	**0.98***
Traits	a	b	c	d	e	f	g

6.2. Siwarowe Pańskie

b	0.06						
c	0.06	0.23					
d	**0.59***	**0.44***	0.21				
e	**0.53***	**0.51***	0.05	**0.79***			
f	0.01	0.15	–0.20	–0.19	**0.45***		
g	**0.48***	0.05	0.15	**0.67***	0.36	–0.36	
h	**0.44***	0.00	0.18	**0.64***	0.24	**–0.50***	**0.99***
Traits	a	b	c	d	e	f	g

6.3. Wielkie Koryciska

b	–0.11						
c	**–0.45***	0.20					
d	**0.71***	0.11	–0.29				
e	**0.59***	**0.50***	0.33	**0.66***			
f	–0.12	**0.47***	–0.08	0.40	**0.41***		
g	**0.50***	0.01	–0.23	**0.77***	0.39	**–0.46***	
h	**0.46***	–0.12	–0.19	**0.76***	0.24	**–0.63***	**0.98***
Traits	a	b	c	d	e	f	g

6.4. Rzepiczna

b	0.17						
c	–0.04	0.22					
d	**0.61***	0.21	0.20				
e	0.34	0.18	0.11	**0.66***			
f	–0.32	0.24	0.05	**–0.54***	–0.16		
g	**0.65***	0.12	–0.02	**0.79***	**0.52***	**–0.38***	
h	**0.67***	0.06	–0.06	**0.82***	**0.49***	**–0.53***	**0.98***
Traits	a	b	c	d	e	f	g

eastern part of the TNP Łysa Skała (1) and the population from its central part Siwarowe Pańskie (2) (Table 5, Fig. 4).

Correlations of traits. The importance of correlation coefficients, the strength of the relationship between them, as well as the direction of those relationships were presented on the basis of Pearson's correlation coefficient (Tables 6.1 – 6.4). Trait d (the width of needle cross-section) shows high or very high positive correlation with traits: the number of resin canals (a), the height of needle cross-section (e), the distance between vascular bundles (g) and h (Marcet coefficient, trait h × g/e). Trait h displays high or very high negative correlation with f (the ratio of height to the width of the needle, the ratio of trait e to d) and correlates positively, almost completely, with the trait g (Tables 6.1–6.3).

Correlations found only in the populations native to the Tatra Mountains. Three investigated populations show high positive correlations of traits a (the number of resin canals) and e (the height of needle cross-section). Apart from positive ones, there are other correlations of traits typical of the two populations or only one of them (Tables 6.1–6.3).

Correlations found only in the population of the lowlands. Only in this population trait e (the height of needle cross-section) correlates with trait g (the distance between vascular bundles) and h (Marcet coefficient, trait h × g/e); those correlations are of high and positive value (Table 6.4).

Discussion

The last period of the Ice Age and particularly its youngest part had a profound influence on the changes of flora in the Tatra Mountains. According to Obidowicz (1996) the Scots pine could not grow there at that time as it first appeared in the Tatra Mountains 11.800–11.000 years ago in the last interstadial Alleröd. However, Staszkiewicz (1993a, b) argues that populations of the Scots pine found there in few isolated distribution areas are relicts dating back to the last interglacial. In the Tatra Mountains two different morphological types of Scots pine populations are observed: a *meridionalis* type found in Wielkie Koryciska in the western part; and a slightly changed *polonica* type found in the eastern part in Łysa Skała. Both of them possess a set of characteristic features of cones and their multivariate analysis allows to distinguish between them (Staszkiewicz 1961a, 1993a, 1993b). These morphological features of cones, typical of *Pinus sylvestris* in the Tatra Mts., are genetically determined, which was proved by Zajączkowski (1949). In comparative cultures they retain their characteristic features related to the speed of growth. They also maintain short needles, characteristic of the mountain population**.** It is not a result of local adaptations because similar populations are found in Europe; *meridionalis* type of populations is found in many places from the Pyrenean in the west, through the Central Massif in France, the Alps, Hungary, to Slovakia; however, *polonica* is found only in Poland and Finland (Staszkiewicz 1961a, 1970, 1993a, b).

The studies on interpopulational variability of *Pinus sylvestris* are often focused on resin canals. Pravdin (1964) and Vidyakin (1981) claim that their number, visible in the cross-section of a needle, constitutes a valuable diagnostic feature, however Lin et al. (2001) suggest that crown position and tree age has important influence to resin-canal density. The attempt to separate population groups by means of resin canals as a diagnostic marker may prove problematic. One reason for this is that the variability amplitudes of this feature may overlap in different areas of the range (Urbaniak 1998). In the eastern Carpathian Mountains, relict populations of *Pinus sylvestris* differ from each other in the number of resin canals which ranges from 9.3 to 14.7 (Molotkov et al. 1979) and from 14.3 to 14.8 (Yacyk 1977). In the three investigated populations in the Tatra Mountains, the variation in the number of resin canals is small and ranges from 11.3 to 12.5. Such differences are not statistically significant (Table 3). The Scots pine population from Tuchola Forests has fewer resin canals (8.9) than populations from the Tatra Mountains. However, this value is smaller than in other populations from the area of Poland, where it usually ranges from 10.0 to 12.5 (Sokołowski 1931; Bobowicz and Korczyk 1994; Urbaniak, 1998**)**. In this broad context the populations from the Tatra Mountains do not differ considerably in regard to the number of resin canals from other populations of the Polish lowlands; there is even a similarity between them. The highest number of resin canals – 12.53 – belongs to the morphological type *polonica* of the eastern part of the Tatra Mountains from Łysa Skała and a relatively geographically close lowland population from Janów Lubelski (Bobowicz and Korczyk 1994). The number of resin canals is correlated with the width and thickness of needles and the distance between vascular bundles (Tables 6.1–6.4). The comparison of the thickness of cuticle with epidermis of different populations indicates that the more northerly the population is located, the thicker the both features (Sokolov 1928; Mamaev 1972). But the populations of the Scots pine of the Tatra Mountains have a considerably thicker cuticle together with epidermis than the population of the lowlands from Tuchola Forests (Tables 2 and 3). This can stem from a modifying influence of the environment which is similar in many parameters in the north and at the height of 1100 meters above sea level where the studied Tatra populations are found. One of the reasons for such a characteristic of the trait can be also a very strong insulation of south slopes where the populations grow. A thicker epidermis with cuticle would prevent excessive evaporation. Whether this is a genetically reinforced feature resulting from natural selection or a consequence of environmental modifications could be determined by thorough research conducted on comparative cultures, but plant material sampled according to standards (e.g. at the same time, at the same crown location, tree age etc.) makes such meaningful comparisons possible.

With regard to the remaining anatomical features of needles, the populations of the Scots pine found in the Tatra Mountains are also differentiated (Tables 4, 5). The three individual features: c (the width of three epidermis cells), g (the distance between vascular bundles) and h (Marcet coefficient, trait d × g/e) distinguish the populations of the Tatra Mts. – 3 (Wielkie Koryciska) from the western part of the TNP and 1 (Łysa Skała) from eastern parts of the TNP (Table 4). Discriminant analysis which maximized distances between the populations proved the diversification of the Tatra Scots pine which had already been considered distinct – 1 (Łysa Skała) and 3 (Wielkie Koryciska) (Staszkiewicz 1993a, b) with regard to its geographical location (Fig. 3).

It seems, that the way to describe this differentiation can only be done by a complex analysis of research on both cones (Staszkiewicz 1993a, b) and needles (Androsiuk and Urbaniak 2006). The authors of this research focus on dissimilarity of lowland and eastern and western Tatra populations. Such a dissimilarity, in terms of morphological traits of needles, is determined by the number of stomata, in most cases regardless of the environment (Urbaniak et al. 2003).

The individual character of populations native to the eastern and western part of the Tatra National Park arises from their history which is related, among other factors, to migration routes from glacial refuges. That would correlate with conception (Wright and Bull 1963; Cheddadi et al. 2004), who claim that the today's differentiation is a consequence of the isolation of populations of different origin in the glacial period. What supports the thesis is a simple conclusion: since the morphological type of populations, *meridionalis*, is widely distributed in southern parts of Europe, it must have been shaped there in the last glacial period. It is not found north of the Carpathian Mountains. Hence, the Scots pine of the Tatra Mountains in Wielkie Koryciska will be the north outpost of *meridionalis* population which developed as a result of the migration to the north. Although the *polonica* type does not have any equivalents in southern Europe, it does have some in the north of the continent, which would prove, in line with Staszkiewicz (1970, 1993a, b), that it was formed in the area of Poland and Slovakia (Lewockie Mts.) and that the Scots pine from Łysa Skała is a relatively old population from historical point of view – even interglacial or early Holocene relict. The comparison of populations from the Tatra National Park with the lowland Scots pine native to Tuchola Forests enables us to find characteristics typical of mountain populations. The needles of population 4 (Rzepiczna) are the widest (traits d), the thickest (traits e) and have the greatest distance between vascular bundles (traits g) (Tables 2, 3).

During the last glacial period *P. sylvestris* was constrained under severe climatic conditions to survive in scattered and restricted refugial areas. The combination of morphological and anatomical data can support thesis that the long-term isolation in the glacial refugia and the migration process during the Holocene had played a major role in shaping the modern diversity of *P. sylvestris* in Europe. The unique character of Tatra pine determine its value. Their today's condition seems to be the best guarantee of their future existence.

References

Androsiuk P., Urbaniak L. 2005. Zróżnicowanie tatrzańskich populacji sosny zwyczajnej (*Pinus sylvestris* L.) na podstawie cech morfologicznych igieł. In: Taksonomia, chorologia i ekologia roślin w dobie zagrożenia różnorodności biologicznej. Materiały Konferencji Naukowej Dedykowanej Prof. W. Żukowskiemu. Poznań. 3-4: 227–231.

Białobok S. 1970. Sosna w lasach Polski od późnego glacjału po czasy współczesne. In: Białobok S. (ed.). Sosna zwyczajna *Pinus sylvestris* L. PWN, Warszawa–Poznań, pp. 20–54.

Birks H.J.B. 1989. Holocene isochrone maps and patterns of tree-spreading in the British Isles. Journal of Biogeography 16: 503–540.

Berglund B.E., Birks H.J.B., Ralska-Jasiewiczowa M. 1996. In; Wright H.E. (eds.) Paleoecological events during the last 15000 years. Willey, Chichester, pp.764.

Bobowicz M. A. 1990. Mieszańce *Pinus mugo* Turra × *Pinus sylvestris* L. z rezerwatu „Bór na Czerwonem" w Kotlinie Nowotarskiej. Wydawictwo Naukowe UAM, Poznań.

Bobowicz M.A., Korczyk A.F. 1994. Interpopulational variability of *Pinus sylvestris* L. in eight Polish localities expressed in morphological and anatomical traits of needles. Acta Societatis Botanicorum Poloniae 63: 67–76.

Cheddadi R., Vendramin G.G., Litt T., Francois L., Kageyama M., Lorentz S., Laurent J.M., De Beaulieu L., Sadori L., Jost A., Lunt D. 2004. Imprints of glacial refugia in the modern genetic diversity of *Pinus sylvestris*. Global Ecology and Biogeography 15: 271–282.

Ferguson G.A., Takane Y. 2007. Analiza statystyczna w psychologii i pedagogice. Wydawnictwo Naukowe PWN, Warszawa.

Giertych M. 1979. Summary of results on Scots pine (*Pinus sylvestris* L.) height growth in IUFRO provenance experiments. Silvae Genetica 28: 136–152.

Giertych M. 1993. Zmienność proweniencyjna. In: Białobok S., Boratyński A., Bugała (eds.), Biologia sosny zwyczajnej: Wyd. Sorus, Poznań-Kórnik, pp. 325–339.

Giertych M., Oleksyn J. 1981. Summary of results on Scotch pine (*Pinus sylvestris* L.) volume production in Ogijevskij pre-revolutionary Russian provenances. Silvae Genetica 41: 133–143.

Hantley B., Birks H.J.B. 1983. An atlas of past and present pollen maps for Europe: 0-13 000 years of age. Cambridge University Press.

Krzyśko M. 1982. Analiza dyskryminacyjna. Wydawnictwo Naukowe UAM, Poznań, Ser. Mat. No. 6, Poznań; 1–98.

Krzyśko M. 1990. Analiza dyskryminacyjna. Wydawnictwo Naukowo-Techniczne, Warszawa.

Latałowa M., Tobolski K., Nalepka D. 2004. *Pinus* L. subgenus *Pinus* (subgen. *Diploxylon* (Koehne) Pilger) – Pine. In: Late glacial and holocene history of vegetation in Poland based on isopollen maps. W. Szafer Institute of Botany PAN, Kraków, pp. 165–178.

Lin J., Sampson D.A., Ceulemans R. 2001. The effect of crown position and tree age on resin-canal density in Scots pine (*Pinus sylvestris* L.) needles. Canadian Journal of Botany 79: 1257–1261.

Łomnicki A. 2000. Wprowadzenie do statystyki dla przyrodników. Wydawnictwo Naukowe PWN, Warszawa.

Łysek S. 1974. Sosna *Pinus sylvestris* L. Studia Ośrodka Dokumentacji Fizjograficznej PAN 3: 87–110.

Mamaev S.A. 1972. Formy vnutrividovoj izmencivosti drevesnych rastenij. Nauka, Moskva.

Mitka J. 1997. Małe, izolowane populacje na skraju zasięgu geograficznego – niektóre procesy ekologiczne i genetyczne. Wiadomości Botaniczne 41: 13–34.

Molotkov P.I., Yacyk R.M., Molcenko L.L. 1979. Izmencivost nekotorych morfo-anatomiceskich priznakov sosny obyknovennoj relikovogo proischozdenia v Karpatach. Lesovodstvo i agrolesomelioracia 54: 14–22.

Obidowicz A. 1996. A late Glacial-Holocene history of the formation of vegetation belts in the Tatra Mts. Acta Paleobotanica 36: 159–206.

Obmiński Z. 1970. Zarys ekologii. In: Białobok S., Boratyński A., Bugała (eds.), Biologia sosny zwyczajnej: Wydawnictwo Sorus, Poznań–Kórnik, pp. 45–66.

Pravdin L.F. 1964. Sosna obyknovennaja izmencivost vnutrividovaja sistematicka i selekcija (Scots pine, Variability, Systematics and Selection). Nauka, Moskwa.

Prus-Głowacki W., Stephan B.R., Bujas E., Alia R., Marciniak A. 2003. Genetic differentiation of autochthonous populations of /Pinus sylvestris/ (Pinaceae) from the Iberia peninsula. Plant Systematics and Evolution 239: 55–66.

Przybylski T. 1993. Autekologia i synekologia. In: Białobok S., Boratyński A., Bugała (eds.), Biologia sosny zwyczajnej: Wydawnictwo Sorus, Poznań-Kórnik, pp. 255–276.

Ruby J.L., Wright J.W. 1976. A revised classification of geographic varieties in Scots pine. Silvae Genetica 25: 169–175.

Skrzyszewski J. 2004. Charakterystyka morfologiczno-przyrostowa sosny zwyczajnej (*Pinus sylvestris* L.) w polskiej części Karpat i Sudetów. Zeszyty Naukowe AR Kraków no. 411.

Sokolov I.A. 1928. K voprosu o geograficeskich rasach *Pinus silvestris* L. Izvestija glavnogo botaniceskogo sada: 559–586.

Sokołowski S. 1931. Prace biometryczne nad rasami sosny zwyczajnej (*Pinus sylvestris* L.) na ziemiach Polski. Prace Rolniczo-Leśne PAU 5: 1–106.

Staszkiewicz J. 1961a. Biometric studies on the cones of *Pinus sylvestris* growing in Hungary. Acta Botanicorum Academy of Science Hungary 7: 451–455.

Staszkiewicz J. 1961b. Zmienność współczesnych i kopalnych szyszek sosny zwyczajnej (*Pinus silvestris* L.). Fragmenta Floristica Geobotanica 7: 97–160.

Staszkiewicz J. 1963. Recherches biometriques sur la variabilite des cones du Pin Sylvestre (*Pinus sylvestris* L.) du Massif Central en France. Fragmenta Floristica Geobotanica 9: 157–187.

Staszkiewicz J. 1968. Badania nad sosną zwyczajną z Europy południowo-wschodniej i Kaukazu oraz jej stosunkiem do sosny z innych obszarów Europy, oparte na zmienności morfologicznej szyszek. Fragmenta Floristica Geobotanica 14: 259–315.

Staszkiewicz J. 1970. Systematyka i zmienność In: Białobok S. (ed.), Sosna zwyczajna *Pinus sylvestris* L. Nasze drzewa leśne PWN, Warszawa–Poznań; 1: 55–57.

Staszkiewicz J. 1993a. Od trzeciorzędu do holocenu. In: Białobok S., Boratyński A., Bugała (eds.), Biologia sosny zwyczajnej: Wydawnictwo Sorus, Poznań-Kórnik, pp. 11–17.

Staszkiewicz J. 1993b. Zmienność morfologiczna szpilek, szyszek i nasion. In: Białobok S., Boratyński A., Bugała (eds.), Biologia sosny zwyczajnej: Wydawnictwo Sorus, Poznań–Kórnik, pp. 33–42.

Urbaniak L. 1998. Geographical differentiation of Scots pine (*Pinus sylvestris* L.) from the area of

Eurasia on the basis of anatomical and morphological characters of needle. A. Mickiewicz University Press, Seria Biologia., no. 58.

Urbaniak L., Ślósarz M., Karliński L. 2001. Description of the relict Scots pine (*Pinus sylvestris* L.) populations in the Tatra and Pieniny mountains by needle characters. Proceedings of the Third Balkan Scientific Conference "Study, conservation and utilization of forest resources". Sofia. Vol. II: 191–198.

Urbaniak L., Karliński L., Popielarz R. 2003. Variation of morphological needle characters of Scots pine (*Pinus sylvestris* L.) populations in different habitats. Acta Societatis Botanicorum Poloniae 72: 37–44.

Watała C. 2002. Biostatystyka- wykorzystanie metod statystycznych w pracy badawczej w naukach biometrycznych. Bielsko-Biała, pp. 423.

Williams B. 1995. Biostatistics concepts and applications for biologists. Chapman and Hall, New York.

Woźniak T., Androsiuk P., Nowak D., Urbaniak L. 2005. The expression of morphological needle characters of Scots pine (*Pinus sylvestris* L.) populations growing in various habitats in Puszcza Notecka. In: Prus-Głowacki W. and Pawlaczyk E. (eds.). Variability and Evolution – New Perspectives. Wydawnictwo UAM, Poznań, pp.449–462.

Wright J.W., Bull W.I. 1963. Geographic variation in Scots pine . Silvae Genetica 12: 1–40.

Vidyakin A.N. 1981. Izmencivost anatomo-morfologicheskogo stroenija sosny v geograficheskich kulturach Kirovskoj oblasti. Lesoved. 5: 18–25.

Yatsyk R.M. 1977. O populacionnoj izmencivosti sosny obyknovennoj reliktovogo proisozdenia v Karpatach. Lesovodstvo i agrolesomelioracia 48: 21–26.

Zajączkowski M. 1949. Studia nad sosną zwyczajną w Tatrach i Pieninach. Studies on the common pine in the Tatra and Pieniny mountains (Western Carpatians). Prace Rolniczo-Leśne PAU 45.

Zwijacz-Kozica T. 1998. Występowanie sosny zwyczajnej *Pinus sylvestris* L. w Tatrzańskim Parku Narodowym. Parki Narodowe i Rezerwaty Przyrody 17.2: 55–68.