BRC
www.brc.amu.edu.pl

Biodiv. Res. Conserv. 3-4: 220-226, 2006

Do needle characteristics of *Pinus uncinata* depend on climatic factors?

Krystyna Boratyńska & Ewelina Muchewicz

Polish Academy of Science, Institute of Dendrology, Parkowa 5, 62-035 Kórnik, Poland, e-mail: borkrys@man.poznan.pl

Abstract: Samples of 10 previous-year needles each were collected from 30 individuals of *Pinus uncinata* in the Vall de Ransol (Pyrenees) in 1999, 2000 and 2003. Each needle was analysed separately for 15 characters. The dependence of average values of particular needle characters in the particular year on average monthly temperatures, average annual temperature, average temperature in June-September, the number of days with average temperatures > 10^{O}C per year, annual precipitation and precipitation in June-September were tested. Results of the preliminary analyses suggest that numbers of stomata, stomatal rows and resin canals in needles can be determined by temperature and precipitation regimes of the year of initiation of the needle (i.e. bud formation), while needle dimensions can be influenced by the temperatures and precipitation in the year of their development and intensive growth.

Key words: plant variation, climatic conditions, Pyrenees, *Pinus uncinata*

1. Introduction

Morphological and anatomical characters of pine needles are recognized as differentiating between species, among other diagnostic features for the taxa from the complex of *Pinus mugo* L., which includes *P. uncinata* Ramond ex DC. (Marcet 1967; Szweykowski 1969; Staszkiewicz & Tyszkiewicz 1972; Szweykowski & Bobowicz 1977; Christensen 1987; Minghetti 1997; Boratyńska & Bobowicz 2001; Boratyńska *et al*. 2003; Boratyńska 2004). Previous-year needles are tested in such investigations, sometimes sampled in different years.

Individual years are characterized with more or less different climatic factors, reflected in average annual and monthly temperatures, average annual precipitation, average number of days with temperatures > 10^{O}C per year, etc. Those factors influence to some degree the morphological and anatomical characteristics of the plants. The commonly known dendrochronological analyses utilize this influence, and analyses on *P. uncinata* wood have shown the correlations of precipitation and temperatures with annual ring widths (Génova 1986; Bosch & Gutierez 1999). Also the influence of climate on needle characters of *P. sylvestris* was tested several times. Needle length in Scots pine depends on the various climatic factors of the year of needle growth (Sokołowski 1931; Pravdin 1964; Żelawski & Gowin 1966; Żelawski & Niwiński 1966; Mamaev 1972; Vidâkin 1981; Staszkiewicz 1993; Fedorkov 2002). Similar studies for *P. uncinata* have not been carried out yet.

The main aim of this work was to examine the 3 population samples collected in the same population in 3 years, to verify which of the needle characters differ statistically significantly among them. Another aim was to verify the dependence of values of particular needle characters on climatic factors during the year of needle initiation and year of needle growth.

2. Material and methods

2.1. Plant material and measuring procedures

Previous-year needles were sampled from 30 individuals in the Vall de Ransol (Andorra) in the Pyrenees at an altitude of about 2000 m, in 1999, 2000 and 2003. The needles were collected in the same population, every year from adult, cone-bearing individuals without symptoms of injury. The trees were not marked, so the population samples were probably represented by different individuals every year. Ten normally developed dwarf shoots were sampled on every individual, from the sunny part of the crown, about 6-7 m above the ground level. One needle of every dwarf shoot was utilized, i.e. 10 needles per individual, according to procedures described earlier (Boratyńska & Bobowicz 2000; Boratyńska *et al*. 2005). The 15 needle characters examined are listed in Table 1.

2.2. Meteorological data

We assessed the dependence of average values of individual characters in the particular year on the average monthly temperatures, average annual temperature, average temperature in June-September, the number of days with average temperatures > 10°C per year, annual precipitation, and precipitation in June-September. The climatic data came from the Ransol meteorological station (altitude 1640 m) of FEDA (Forces Electriques d'Andorra). The influence of the meteorological data on characters of the needles was tested for the year of needle initiation (i.e. bud formation) and year of needle growth.

2.3. Statistical methods

The average values of every character were calculated separately for individuals and the population for every year. The Student's *t*-test for independent samples was utilized to verify the statistical differences between the years of collection. The relations among the population samples representing particular years of collection were verified by using the analysis of discrimination on the measured characters (Łomnicki 2000; Watała 2002).

The power of connection between the average values of characters of needles collected in particular years and corresponding meteorological data detected with the use of correlation coefficients. Results should be treated as provisional because only 3-year data were accessible.

The package Statistica PL for Windows was used in the calculating procedures.

3. Results

The average values of most of characters differed at a statistically significant level. Only epidermal cell height and stomatal rows ratio (characters 10 and 13, respectively) did not show any significant differences among population samples collected in different years. The distance between vascular bundles and Marcet's coefficient (characters 9 and 12, respectively) differed significantly between all 3 years, while the other traits between 2 of them (Fig. 1). The highest number of traits differed between population samples collected in 1999 and 2003.

The multivariate relations between population samples representing different years were tested by the analysis of discrimination. In spite of statistically significant differences between population samples in values of particular needle characters, the individuals from all of them form a single conglomeration on the plane of the first 2 discrimination variables, which account for 100% of total variation (Fig. 2). The population sample collected in 1999 differed slightly from those collected in 2000 and 2003 in respect of the first variable, accounting for more than 70% of total variation. The needles sampled in 1999 had more stomatal rows on both sides of the needle (characters 2 and 3) and higher values of needle width (character 7) and distance between vascular bundles (character 9). Also the number of resin canals and needle length was of some discriminating value (character 6 and 1).

Results of the preliminary analyses indicated the possible connections of some needle traits with climatic factors. Numbers of stomatal rows on both sides of the needle, numbers of stomata on both sides of the needle,

Table 1. Needle characters analysed

No.	Characters	Precision
1	Needle length	1 mm
2	Number of stomatal rows on convex (abaxial) side of needle	
3	Number of stomatal rows on flat (adaxial) side of needle	
4	Number of stomata on 2-mm long section of needle, on convex (abaxial) side	
5	Number of stomata on 2-mm long section of needle, on flat (adaxial) side	
6	Number of resin canals	
7	Needle width	0.1 mm
8	Needle thickness	0.1 mm
9	Distance between vascular bundles	1 µm
10	Epidermal cell height	1 µm
11	Epidermal cell width	1 µm
12	Marcet's coefficient (=traits 9×7/8)	
13	Stomatal rows ratio (=traits 2/3)	
14	Needle thickness/width ratio (=traits 8/7)	
15	Epidermal cell width/height ratio (=traits 11/10)	

and number of resin canals (characters 2, 3, 4, 5 and 6, respectively) depended on temperature and precipitation in the year of needle initiation. The above-mentioned characters of the needle correlated positively most closely with average temperatures in April and May in the year of needle initiation (Figs. 3a, 3b, 3g and 3m). High temperatures during the summer months seemed to reduce the number of stomatal rows (Figs. 3e and 3f),

Character 1

2000		
2003	xx	xx
	1999	2000

Character 2

2000	xx	
2003	xx	
	1999	2000

Character 3

2000	xx	
2003	xx	
	1999	2000

Character 4

2000	x	
2003		x
	1999	2000

Character 5

2000	xx	
2003		
	1999	2000

Character 6

2000	xx	
2003	x	
	1999	2000

Character 7

2000	xx	
2003	xx	
	1999	2000

Character 8

2000	xx	
2003	xx	
	1999	2000

Character 9

2000	xx	
2003	xx	x
	1999	2000

Character 11

2000		
2003	xx	xx
	1999	2000

Character 12

2000	xx	
2003	xx	x
	1999	2000

Character 14

2000	xx	
2003	xx	
	1999	2000

Character 15

2000		
2003	xx	x
	1999	2000

Fig. 1. Differences among the average values of needle characters in *Pinus uncinata* sampled in 3 years in the Vall de Ransol, Andorra, tested with Student's *t*-test; xx-$P = 0.01$; x-$P = 0.05$ (character numbers as in Table 1)

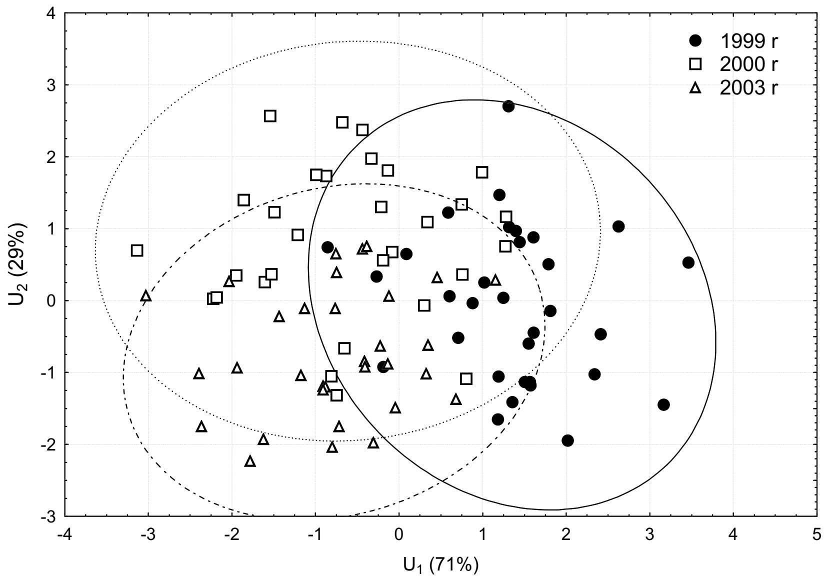

Fig. 2. Results of discrimination analysis for the individuals of *Pinus uncinata* sampled in 3 years in Vall de Ransol, Andorra

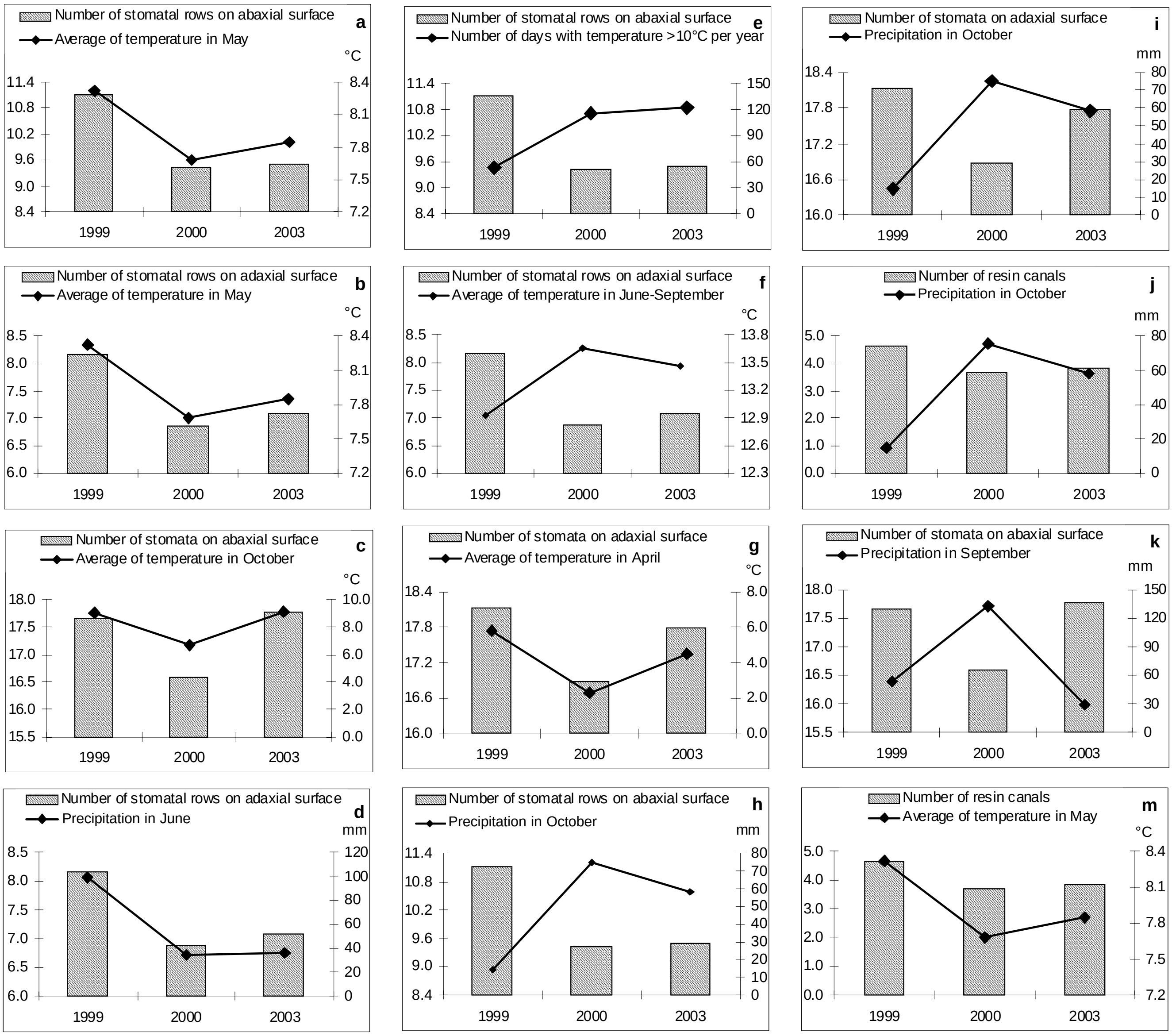

Fig. 3. Numbers of stomata, stomatal rows and resin canals in needles versus climatic factors in the year of needle initiation in *Pinus uncinata*

but these characters correlated positively with the average temperatures in October (Fig. 3c). Generally, numbers of stomatal rows and resin canals are connected positively with the average temperature in the year of needle initiation.

The precipitation in June in the year of needle initiation stimulated the numbers of stomatal rows (Fig. 3d), but precipitation in autumn months reduced them strongly (Figs. 3h, 3i and 3k). Precipitation in autumn had a negative effect also on the number of resin canals (Fig. 3j). The number of stomata on both sides of the needle correlated positively with annual precipitation but negatively with precipitation in June-September in the year of needle initiation.

The quantitative traits, as for example needle length, width and thickness, were influenced by climatic conditions in the year of intensive growth of the needle. Needle length, width and thickness (characters 1, 7 and 8, respectively), correlated negatively with average temperatures in the first and last month of the growing period, but positively with temperatures in summer months (Figs. 4a, 4b, 4e and 4f). Epidermal cell height and width (characters 10 and 11) correlated positively with the average temperature in June (Fig. 4c), but negatively with average temperatures in other months (Fig. 4g) and with the number of days with temperatures > 10°C per year (Fig. 4m).

The dimensional characters of the needle generally were negatively connected with precipitation in spring, but positively with the precipitation at the end of the summer in the year of intensive growth of the needle. The high precipitation in that year and high precipitation in June-September reduced strongly the length of the needle, but also its thickness (Figs. 4i and 4j). The connection of the width and height of epidermal cells with precipitation was not clear. A positive correlation

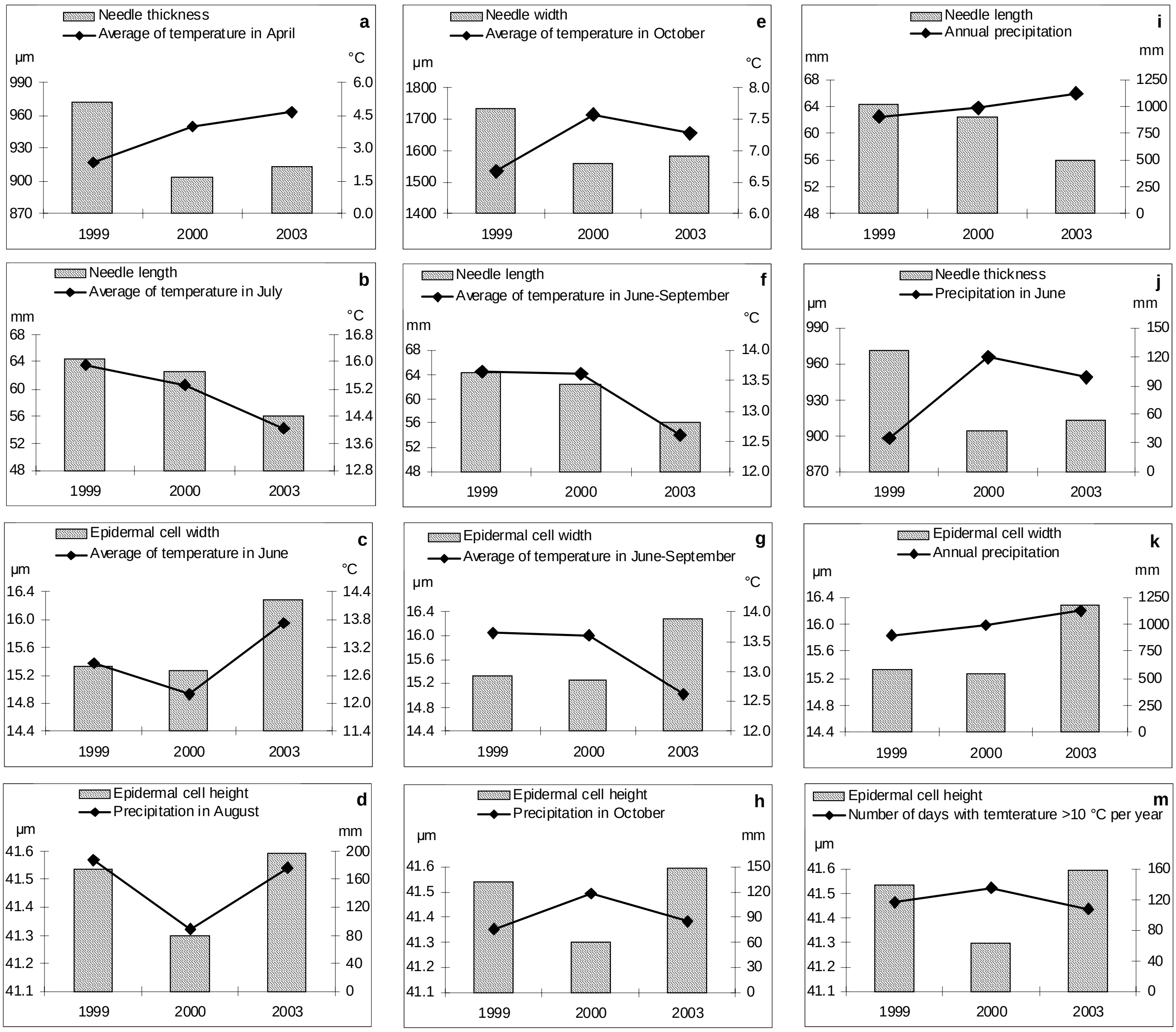

Fig. 4. Dimensional characters of needles of *Pinus uncinata* versus climatic factors in the year of needle development and intensive growth

was detected for epidermal cell height with precipitation in August, but negative with precipitation in October (Figs. 4d, 4h). Epidermal cell width was slightly positively connected with annual precipitation (Fig. 4k).

The distance between vascular bundles did not show clear connections neither with temperature nor with precipitation.

4. Discussion

The length of the needles sampled in 3 years in the same population in our study varied, but only the year 2003 showed statistically significant differences. The length of 64, 63 and 56 mm in the years 1999, 2000 and 2003, respectively, only slightly differed from the average data known from a previous study. The average value of this character was estimated by Boratyńska & Bobowicz (2000) at 66 mm, with variation coefficient at 11%, which corresponds closely with the data for the years 1999 and 2000 in the present study. The shortest needles in the population of *Pinus uncinata* in Vall de Ransol were formed in 2003, probably as a result of a very high level of precipitation in 2002. Similar connections were found also between the width and thickness of the needle with a high level of precipitation in the year of initiation of the needle. However, the influence of lack of precipitation (drought) during the year of growth of the needle was a reason of its smaller size in the case of *Pinus canariensis* (Grill *et al*. 2004). The length of *Pinus mugo* needle in the Tatras depends on temperature and air humidity in the year before their formation (Copińska 1975 by Staszkiewicz 1993).

Epidermal cell height (i.e. thickness of epidermis) in *Pinus sylvestris* needles changes with geographic latitude and with altitude in the mountains (Mamaev 1972), so it shall be influenced by changes of temperatures and precipitation regime. The direct connection of epidermal cell height and also the distance between vascular bundles with climatic factors has not been studied. It was shown in *P. sylvestris* that the distance between vascular bundles has higher values in the more southern populations, but it was rather disputed as an effect of provenance differentiation rather than of climatic influence (Mamaev 1972; Vidâkin 1981). The distance between vascular bundles of *P. uncinata* in our data also was not influenced by climatic factors in the 3 different years.

The number of stomata on needles of *Pinus sylvestris* depends on geographic latitude (Marcet 1967; Fedorkov 2002) and also on precipitation (Mamaev 1972). The number of stomata on *P. uncinata* needles in our study also showed a connection with the precipitation in the year of needle initiation.

The influence of the climatic factors on the number of resin canals in the needle of *Pinus sylvestris* has been studied by Vidâkin (1981), who did not find any direct connection. The variation in the number of resin canals was rather explained as a result of differences in the age of individuals (Petrenko 1967). The number of resin canals grew with the age of the tree and also depended on the position in the crown and length of the needle (Lin 2001). The influence of these factors was eliminated in our study by sampling from trees of similar age and at a similar height of their crowns. The significantly higher number of resin canals sampled in 1999 can be connected with low temperatures in the summer of 1998, when the needles grew intensively. The number of days with average temperature > 10°C was a half lower in 1998 than in 1999 and 2002, the years of growth of the needles sampled in 2000 and 2003. The years of growth of needles of all 3 population samples did not differ in annual precipitation, but in 1997, i.e. in the year of initiation of the needles sampled in 1999, there was very high snowfall, significantly higher than in the corresponding years of the other 2 population samples, i.e. in 1998 and 2001.

5. Conclusions

The results of this preliminary study suggest that some needle traits, such as numbers of stomata and stomatal rows, and also number of resin canals (characters 2-6) can be determined by temperature and precipitation regimes of the year of initiation of the needle. The dimensional traits of the needle (characters 1, 7-10, Table 1) can be influenced by the temperatures and precipitation in the year of its development and intensive growth.

Acknowledgements. The work was partly financed by the Institute of Dendrology, Kórnik, Poland. Material for the study was collected thanks to bilateral cooperation between the Polish Academy of Sciences and Consejo Superior de Investigaciones Cientificas, Spain.

References

Boratyńska K. 2004. Relacje taksonomiczne między sosnami z kompleksu *Pinus mugo* (*Pinaceae*) na podstawie cech igieł. Fragm. Flor. Geobot. Polonica 11(2): 235-255.

Boratyńska K. & Bobowicz M. A. 2000. Variability of *Pinus uncinata* Ramond ex DC as expressed in needle traits. Dendrobiology 45: 7-16

Boratyńska K. & Bobowicz M. A. 2001. *Pinus uncinata* Ramond taxonomy based on needle characters. Plant. Syst. Evol. 227: 183-194.

Boratyńska K., Boratyński A. & Lewandowski A. 2003. Morphology of *Pinus uliginosa* (Pinaceae) needles from populations exposed to and isolated from the direct influence of *Pinus sylvestris*. Bot. J. Linn. Soc. 142: 83-91.

Boratyńska K., Marcysiak K. & Boratyński A. 2005. *Pinus mugo* in the Abruzzi Mountains: high morphological variation of isolated population. Bot. J. Linn. Soc. 147: 309-316.

Bosch O. & Gutiérrez E. 1999. La sucesión los bosques de *Pinus uncinata* del Pirineo. De los anillos de crecimiento a la historia del bosque. Ecologia 13: 133-171.

Christensen K. I. 1987. Taxonomic revision of the *Pinus mugo* complex and *P.* ×*rhaetica* (*P. mugo* × *P. sylvestris*). Nord. J. Bot. 7: 383-408.

Fedorkov A. L. 2002. Variation in the anatomical characters of needles and their resistance to technogenic and climatic stress in Scots Pine. Russian J. Ecol. 33(1): 65-67.

Génova R. 1986. Dendroclimatology of mountain pine (*Pinus uncinata* Ram.) in the Central Plain of Spain. Tree-ring Bulletin 46: 3-12.

Grill D., Tausz M., Pöllinger U., Jimčnez M. S. & Morales D. 2004. Effects of drought on needle anatomy of *Pinus canariensis*. Flora 199: 85-89.

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. Can. J. Bot. 79: 1257-1261.

Łomnicki A. 2000. Wprowadzenie do statystyki dla przyrodników. 263 pp. PWN, Warszawa.

Mamaev S. A. 1972. Formy vnutrividovoi izmenchivosti drevesnych rastenii. 282 pp. Nauka, Moskva.

Marcet E. 1967. Über den Nachweis spontanen Hybriden von *Pinus mugo* Turra and *Pinus sylvestris* L. aufgrund von Nadelmerkmalen. Ber. Schweiz. Bot. Gesell. Bull. Soc. Bot. Suisse 77: 314-360.

Minghetti P. 1997. Contributo alla conoscenza di *Pinus mugo* agg. in Trentino (Italio): un approccio biometrico. Webbia 52(1): 67-85.

Petrenko E. C. 1967. Izmenchivost chisla smolânykh kanalov v khvoi sosny obyknovennoi. Lesoved. 6: 76-83.

Pravdin L. F. 1964. Sosna obyknovennaya. Izmenchivost vnutrividovaya sistematika i selekciya. 190 pp. Nauka, Moskva.

Sokołowski S. 1931. Prace biometryczne nad rasami sosny zwyczajnej (*Pinus sylvestris*) na ziemiach Polski. Prace rolniczo-leśne PUJ 5: 1-106.

Staszkiewicz J. 1993. Zmienność morfologiczna szpilek, szyszek i nasion. In: S. Białobok, A. Boratyński & W. Bugała (eds.). Biologia sosny zwyczajnej, pp. 33-43. PAN Instytut Dendrologii, Kórnik.

Staszkiewicz J. & Tyszkiewicz M. 1972. Zmienność naturalnych mieszańców *Pinus sylvestris* L. × *Pinus mugo* Turra (= *P.* ×*rotundata* Link) w południowo-zachodniej Polsce oraz na wybranych stanowiskach Czech i Moraw. Fragm. Flor. Geobot. 18(2): 173-191.

Szweykowski J. 1969. The variability of *Pinus mugo* Turra in Poland. Bull. Soc. Amis Sci. Lett. Poznań, ser. D 10: 39-54.

Szweykowski J. & Bobowicz M. A. 1977. Variability of *Pinus mugo* Turra in Poland IV. Needles and cones in some Polish populations. Bull. Soc. Amis Sci. Lett. Poznań, ser. D 17: 3-14.

Vidâkin A. N. 1981. Izmenchivost anatomo-morfologocheskogo stroenya khvoi v geograficheskikh kulturakh Kirovskoi oblasti. Lesoved. 5: 18-25.

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

Żelawski W. & Gowin T. 1966. Variability of some needle characteristics in Scots pine (*Pinus sylvestris* L.) ecotypes grown on the comparative plantation. Ekol. Pol. 14A(17): 275-283.

Żelawski W. & Niwiński Z. 1966. Variability of some needle characteristics in Scots pine (*Pinus sylvestris* L.) ecotypes grown in native conditions. Ekol. Pol. 14A(19): 301-308.