The shrub and Black Locust communities of chosen parts of the Hron downs, the Slovak Republic

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Abstract

BENČAŤOVÁ, B., KOPRDA, J., BENČAŤ, T. 2013. The shrub and Black Locust communities of chosen parts of the Hron downs, the Slovak Republic. *Folia oecol.*, 40: 157–162.

The scrubland and Black Locust phytocoenoses belong to the substitute communities which constitute an important component in present cultural landscape. They arose and evolved according to certain rules. Their presence and arrangement is mainly dependent on the type of agricultural land. In the study area, around the Arborétum Mlyňany, these systems have become a permanent component of the vegetation. In our article there are given phytocenological and ecological characteristics of scrubland and Black Locust forest stands in the municipalities of Vieska nad Žitavou, Tesárske Mlyňany and Slepčany. Within the scrubland we determined association *Ligustro-Prunetum* R.Tx. 1952 with the ecological variations of *Prunus spinosa* and *Vitis vinifera* and within the Black Locust we determined association *Chelidonio-Robinietum* Jurko 1963, with ecological variant with *Hedera helix* and with facias with *Rubus caesius* and *Vinca minor* and association *Bromo sterilis-Robinietum* Jurko 1963 prov. For the allocation of communities were used numerical methods (JUICE, TWINSPAN), ecological analysis was conducted in the program JUICE. On the basis of the performed ecological analysis we can conclude that the communities are very similar in their ecological claims.

Keywords

the Arborétum Mlyňany, Black Locust, cultural landscape, phytocoenoses

Introduction

The scrubland and Black Locust phytocoenoses belong to the substitute communities which constitute an important component in present cultural landscape. They arose and evolved according to certain rules. Their presence and arrangement is mainly depended on the type of agricultural land. In the study area, around the Arborétum Mlyňany, these systems have become a permanent component of the vegetation. In the past not much attention was paid to the study of both types of communities and even nowadays the syntaxonomy of these communities is not definitely worked out.

Besides JURKO (1964) the study of shrubs in Slovakia was performed mainly by KONTRIŠ (1966) who

described the shrub field communities of north-western part of Liptovská kotlina basin. In the recent years the theoretic questions of syntaxonomic position of the shrub communities were studied by VALACHOVIČ (2002, 2007). KOPRDA (2008) described in his diploma thesis the shrub communities of the part of Žitavská pahorkatina hills and the newest paper about hazel communities of the Veľká Fatra Mts was published by KLIMENT et JAROLÍMEK (2011).

Problem of classification of the Black Locust communities in Slovakia was examined by ŠčEPKA (1982, 1985), JURKO (1963), JURKO ET KONTRIŠ (1982), in the recent period by ŠIMONOVIČ et al. (2002), BENČAŤOVÁ et BENČAŤ (2005, 2008), BENČAŤOVÁ et al.157, (2008), KOPRDA (2008). The study area is located in the cadastres of villages Vieska nad Žitavou and Slepčany and because of the fact that the paper follows the papers by BENČAŤOVÁ et BENČAŤ (2005), BENČAŤOVÁ et al. (2008), characteristic of the territory is described in the mentioned references.

Material and methods

Phytocenological research was performed during the growing seasons of the years 2006–2007. Within the field research and vegetation synthesis was followed the Zürich-Montpellier School method with 7 degree scale abundance and dominance (BRAUN-BLANQUET, 1964). Nomenclature of plants is given according to MARHOLD et HINDÁK (1998), nomenclature of syntaxa according to the actual vegetation units of Slovakia by JAROLÍMEK et ŠIBÍK (2008).

Phytocenological records were saved in the database program TURBO(VEG) (HENNEKENS, 2005). Output numeric matrix of the program with the phytocenological records were used as an input data for the next management in the program JUICE (TICHÝ, 2002) for the following purposes: differentiation of the syntaxonomic units with the program TWINSPAN (HILL, 1979), indirect unimodal gradient analysis DCA and ecological analysis of the communities.

Results and discussion

The following syntaxonomic units were selected with the numeric classification methods in the study area:

o The scrubland communities

Studied scrubland communities syntaxonomically belong to *Rhamno-Prunetea* Rivas-Goday and Borja-Carbonell 1961 family and to the two alliances. *Berberidion vulgaris* Br.-Bl. 1950 alliance includes *Ligustro-Prunetum* R. Tx. 1952 association and *Arctio-Sambucion nigrae* Doing 1962 alliance includes *Anthrisco-Lycietum halimifolii* Jurko 1964 association (Fig. 1).

- Ligustro-Prunetum R. Tx. 1952 association

The scrubland communities of the association are spread in the whole study area. The most significantly they are represented on the slight slopes, mainly with the western or south-western exposition. Common sign of most of the records of the community is their occurrence on the sites that were in the past intensively used mostly like pastures, alternatively like mown meadows or they are developed like narrow stripes of shrubs among vineyard areas. The sites are affected by intensive human acting and so they were supplied with sufficient supply of nutrients, mainly with nitrogen in the initial phase of their creation.

Association is represented by the poorest scrubland community according to species diversity in the area. The tree layer is negligible, created mainly with the stronger individuals of the shrubs *Prunus spinosa*, *Crataegus monogyna*, or the fruit tree *Cerasus avium*. In the case of older shrubs in the studied area phytocenoses there are *Quercus cerris* and *Robinia pseudoacacia* that infiltrate to the shrubs from the surrounding black locust stands.

The cover of the shrub layer is large, averagely it reaches the value 90%. In the layer there occur mainly three types of shrubs which create typical, nearly impenetrable structure – dominant *Prunus spinosa* species, with associated *Crataegus mono-gyna* and *Rosa canina* species.

Herb layer is in the most of the records very poor, with the average cover 15%. Herbs are spread especially in the marginal parts of the communities where they are particularly represented with nitrophilous species with high demand for nutrients; *Galium aparine, Geum urbanum, Glechoma hederacea, Anthriscus cerefolium, Urtica dioica* and others. From the grass species there is constantly



Fig. 1. Dendrogram created from the three groups of records in the scrubland communities representing syntaxonomic units (1st association *Anthrisco-Lycietum halimifolii*, 2nd variant with *Prunus spinosa*, 3rd variant with *Vitis vinifera*).

occurred *Poa nemoralis* species. We also recorded quite high occurrence of the juvenile stages of *Ligustrum vulgare* species, *Prunus spinosa* and *Rosa canina* woody species.

On the basis of certain differences in the habitat conditions, in the floristic composition there were selected two variants within the association: phytocoenoses with predominant *Prunus spinosa* species as an ecological variant with *Prunus spinosa* of the association *Ligustro-Prunetum* R. Tx. 1952. and phytocoenoses with the floristic composition affected with the higher addition of cultural plant species (*Vitis vinifera*), as an ecological variant of association *Ligustro-Prunetum* R. Tx. 1952 association with *Vitis vinifera* which suggests big anthropic influence in these localities.

- Anthrisco-Lycietum halimifolii Jurko 1964 association

The shrubs of the association occupy very small area in the locality as they are located at the top parts of moderate slopes sufficiently warmed by the sun. They create differently wide stripes (2–15 m) at the interface of fields and vineyards ensuring favourable habitat conditions and additional nutrient supplementation from the fertilization of agricultural land.

The cover of the tree layer is negligible and we can find there not high specimens of *Acer campestre* and *Juglans regia*. Average cover of the shrub floor reaches 95 % and the layer is characteristic with mono-dominance of introduced *Lycium barbarum* species which creates dense and impenetrable stands. *Prunus spinosa* and *Rosa canina* species are characterised with high stability with lower cover.

Herb layer is very sparse with the average cover 20%. Inside the shrubs, there only rarely occur herbaceous species, mostly thermophilous ruderal and nitrophilous species of therophytes – *Ballota nigra, Anthriscus cerefolium, Fallopia convolvulus, Galium aparine, Geum urbanum, Urtica dioica,*

Arum alpinum and before the foliage of the shrubs *Veronica hederifolia* and *Lamium purpureum*. From the grass species occurs in the layer mesophyte *Poa nemoralis*.

• The Black Locust communities

The studied Black Locust communities syntaxonomically belong to *Robinietea* Jurko ex Hadač and Sofron 1980 family and to the two alliances. *Chelidonio-Robinion* Hadač and Sofron 1980 alliance includes *Chelidonio-Robinietum* Jurko 1963 association and *Balloto nigrae-Robinion* Hadač and Sofron 1980 alliance includes *Bromo sterilis-Robinietum* Jurko 1963 association (Fig. 2).

- Chelidonio-Robinietum Jurko 1963 association

In the studied territory it is the second most spreading and with the number of species the richest Black Locust community. It occurs especially on the slopes with western exposition and slight tendency. Common feature of all plots is sufficiency of soil moisture and increased mineral content of the soil. The tree layer is created with the dominant Black Locust completed in some cases with the native oaks (Quercus cerris, Q. robur). Average cover of the layer is 75% whilst average cover of the shrub layer is 10-55%. Within the species composition there dominates Sambucus nigra, another dominant species are Ligustrum vulgare and Robinia pseudoacacia. The differential species which differentiate one association from the other are represented by introduced Mahonia aquifolium and Prunus cerasus species which penetrate to the stands from the scrubland communities. Mahonia aquifolium species is nowadays considered to be an invasive species and probably it got to the community from the neighbouring Arborétum Mlyňany.

Physiognomy of the herb layer is largely identified with nitrophilous species with the dominant *Chelidonium majus*. The layer is also rich on *Galium aparine*, *Allium vineale*, *Urtica dioica* and others.



Fig. 2. Dendrogram created from the four groups of records within the Black Locust communities representing syntaxonomic units (1 *Chelidonio-Robinietum* association, 2 facia with *Rubus caesius*, 3 facia with *Vinca minor*, 4 *Bromo sterilis-Robinietum* association).

In the community there is noticeable influence of the time aspect and therefore there is conditional species occurrence on the basis of growing season as well. Early in spring there is visibly increased occurrence *Ficaria bulbifera* species and *Veronica hederifolia* species, but later there dominates *Chelidonium majus* species with the mixture of grasses, especially *Bromus sterilis* species. During the summer period the herb layer becomes dry.

On the basis of certain differences in the phytocenological, ecological and habitat conditions and dominant representation of *Hedera helix* species, but also on the basis of the highest similarity with this association we selected ecological variant with *Hedera helix* with facias with *Rubus caesius* and *Vinca minor* within the association.

Bromo sterilis-Robinietum Jurko 1963 prov. association

In the studied area it is the most spreading association but regarding the species number it is poorer than previous association. It occurs on the similar habitats regarding exposition and slope tendency but generally on the bright and drier places with sandy and mineral-poor soils.

The tree layer is created with the dominant Black Locust which is, however, a bit lower in its growth. The layer is also rich on *Acer campestre*, *Carpinus betulus*, *Quercus cerris* and *Q. robur* species.

The shrub layer has lower cover (1–50%) and again it is characterised with the domination of *Sambucus nigra* species, constantly also occurs *Robinia pseudoacacia* species and significantly is also represented *Euonymus europaeus* species.

Within the herb layer there is dominant *Bromus* sterilis species, with the lower cover occur nitrophilous Stellaria holostea, Galium aparine, Chelidonium majus, Arum alpinum, Geum urbanum and Urtica dioica species. Compared to the previous association, this association especially differs with Anthriscus sylvestris, Arrhenatherum elatius, Ballota nigra, Geranium robertianum, Lamium purpureum and Viola hirta species. Also in this association there is noticeable seasonal character of the herb layer, significant spring aspect at the beginning of summer (after fading of Bromus sterilis species) changes and understorey becomes poor.

Ecological analysis of the study communities

For the purposes of ecological analysis was used the selection into the shrub and Black Locust communities, the communities were compared with each other regarding their demands on different factors of the environment and we found out the following facts.

Demands of both community groups regarding their light requests are quite equal. The numerical values are in the range 5.5-6.4 and they can be evaluated as the half-shade-like or even half-light-like communities. With respect to the temperature they are thermophilic communities, relatively more thermophilic seem to be the shrubs. In the question of continentality both groups can be classified as oceanic or even sub-oceanic, i.e. containing the species that occur in most parts of Central Europe. Demands of both groups regarding their requests on soil moisture are equal, most of the phytocoenoses' species inclines to the dry or even freshly wet soils. In the question of soil reaction there are no differences between the groups. Eco-index 7 characterizes the species of acidic to neutral soils. Regarding the content of nitrogen substances in the soil, both groups can be described as nitrophilous communities.



Fig. 3. Comparison of the scrubland and Black Locust communities on the basis of average eco-indexes (L, light; T, temperature; C, continentality; M, moisture; R, soil reaction; N, soil nitrates).

Ecological analysis of the communities confirmed that the scrubland and also Black Locust communities are ecologically similar in the studied area and differentiate with each other only slightly in the individual ecological indicators (Fig. 3).

Conclusion

In the paper we focused on geobotanical and ecological characteristics of the scrubland and Black Locust communities in the Arborétum Mlyňany surroundings which cover quite large areas on this territory. We confirmed the fact that besides aesthetic function the shrubs in the agricultural land also have all-round biological and economic importance. Acquired phytocoenological data represent only a fragment of vegetation diversity of the shrub and Black Locust stands on the territory of Central Požitavie. Despite we hope that the information will contribute to the knowledge about the real state of scrubland and Black Locust communities on the whole territory of Slovakia and we also believe that the paper will help to the deepening of public awareness about the studied area.

Acknowledgements

The work was supported by projects of VEGA Slovakia No. 1/0551/11 and 2/0059/11.

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Received December 6, 2012 Accepted April 14, 2013

Diversity in honey locust (*Gleditsia triacanthos* L.) seed traits across Danube basin

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Abstract

FERUS, P., BARTA, M., KONÔPKOVÁ, J., TURČEKOVÁ, S., MAŇKA, P., BIBEŇ, T. 2013. Diversity in honey locust (*Gleditsia triacanthos* L.) seed traits across Danube basin. *Folia oecol.*, 40: 163–169.

Honey locust (Gleditsia triacanthos L.), in the past planted as ornamental, technical or forest tree, is presently considered as casually invasive tree in Danube basin. Since plant invasiveness is usually tightly associated with its reproduction biology, in this work we focused on characterization of seeds from honey locust populations across this area. Analysing seed coat colour, thousand seeds weight (TSW), seed projection area, seed thickness, percentage of germinated seeds and their germination energy, as well as portion of seeds infested by honey locust seed beetle (Megabruchidius tonkineus), consumed part of seeds and their germination ability in relation to seed characteristics, local temperature means and precipitation sums during vegetation period, we came to the following conclusions: seed coat colour diversity decreases with geographical latitude; TSW, seed projection area and thickness were negatively correlated to mean temperature and positively to precipitation sum; between percentage of naturally germinated seeds and TSW as well as seed thickness we found positive correlations; germination energy showed positive relation to mean temperature and a negative one to precipitation sum; and the same relations were observed for infested seeds percentage and consumed seed part. No infested seed was able to germinate. From these results we can conclude that in colder and wetter conditions higher seed germinability, and in warmer and drier conditions enhanced germination energy of seeds supports spreading of this tree species. However, honey locust seed beetle can significantly affect seed germinability in regions with warm and dry summers.

Keywords

Danube basin, honey locust (*Gleditsia triacanthos* L.), honey locust seed beetle (*Megabruchidius tonkineus*), invasiveness, seed traits,

Introduction

Honey locust (*Gleditsia triacanthos* L., Caesalpinaceae) is a leguminous tree originating in the middle and eastern part of North America, which was in Southern Slovakia and Hungary widely planted in parks as ornamental species, round vineyards, gardens and fruit groves as thorn-hedge, along roads and fields as wind barrier, and as a component of floodplain forests (CHRTKOVÁ and JASIČOVÁ, 1988; GYÖRGY, 2007; HARASZTHY, 2001). Into Europe it was introduced in 1700, and its first plantation

in the area of present Slovakia was established in 1806 in Dolná Krupá castle (BENČAŤ, 1982).

However, presently this species is ranked as often escaping from culture (GOJDIČOVÁ et al., 2002) or newly as naturalized in Slovakia (MEDVECKÁ et al., 2012) and casually invasive in Hungary (BALOGH et al., 2004). It causes extreme complications in Argentinian Pampa grasslands and central natural forests, where a large effort in ecological research has been paid (CHANETON et al., 2004; DE VIANA and SPERONI, 2003; MARCO and PAEZ, 2000). Finally, in 1993 Autralian Queensland

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spent 400,000 \$ for eradication campaign 'search and destroy', stimulated by honey locust infestation of 1,000 ha of Brisbane valley (CSURHES, 2004).

BENČAŤ (1982) classified this tree as very tolerant to industrial and transport emissions; SCHINDELBECK and RIHA (1988) described its relatively high resistance to low soil reaction (up to pH 5). More recent works focus on its extraordinary tolerance to high temperature (GRAVES et al., 1991; GRAVES and WILKINS, 1991; GODOY et al., 2011) and drought (GRAVES and WILKINS, 1991; BURTON and BAZZAZ, 1991; BURTON and BAZZAZ, 1995), very helpful for its expansion.

PyšEK and RICHARDSON (2007) summarizing results of 59 studies in 64 alien plant species ordered traits associated with their invasiveness. As expected, features connected to generative reproduction appeared among them. In honey locust, combined clonal and sexual reproduction, short juvenile period, high seed production and high seed germinability were described (MARCO and PAEZ, 2000).

However, there is no reference about changes in this kind of traits with climatic conditions in literature. Therefore, in this work we analysed honey locust seeds from populations across the Danube basin.

Material and methods

Honey locust (*Gleditsia triacanthos* L.) pods from different regions of Slovakia and Hungary (locations with GPS coordinates are listed in Table 1) were collected at the end of November 2011. From each locality three trees were sampled. Trees were determined using determination keys of REHDER (1990) and KRÜSSMANN (1960).

Table 1. Collecting sites of honey locust pods with GPS coordinates

Locality	GPS coordinates
Vieska n/Ž. (SK)	N 48°19′00.6′′ E 18°22′05.4′′
Szirák (HU)	N 47°49′49.4′′ E 19°30′36.4′′
Gyöngyös (HU)	N 47°45´56.4´´ E 19°56´42.3´´
Debrecén (HU)	N 47°34′06.3′′ E 21°35′50.3′′
Mezőtúr (HU)	N 47°00′11.4′′ E 20°38′41.0′′
Békéscsaba (HU)	N 46°40′50.1″ E 21°02′50.1″
Szeged (HU)	N 46°22´38.3´´ E 20°02´25.5´´

Pods were stored at 3 °C till seeds were shucked (3 days). Then we let them dry at room temperature (21/17 °C during the day/night). In the course of four months lasting storage in these conditions, adults of honey locust seed beetles (*Megabruchidius tonkineus* Pic, 1904) have enclosed and flied out from seeds. Injured seeds were collected and analysed for weight reduction related to seed beetle life cycle (%) and germination ability (%).

Thereafter, hundred healthy seeds from each tree sample were submitted to multiple analyses:

- o Seed coat colour
- Thousand seeds weight (g)
- Seed projection area (mm²)
- Seed thickness (mm)
- o Germinated seeds (%)
- \circ Germination energy (mm d⁻¹).

Seed coat colour was determined visually. Thousand seeds weight (TSW) we calculated from weight of hundred seeds, dried at room temperature. Seed projection area was defined by scanning and area analysis using ImageJ software (ver. 1.46). For seed thickness measuring we utilized a slide calliper. For the latter two tests, seeds have been swelling in water for 1 week, continuously transferred into transparent plastic boxes lined with a water-soaked tissue and analysed for natural germinability. Thereafter, non-swelled seeds were scarified in 93% sulphuric acid for one hour, washed thoroughly in water (AsL et al., 2011), let swell for 24 h and transferred into plastic boxes, as described above. Germination energy was analysed after 4 days of germination as hypocotyl length growth rate. All these procedures were done at 25 °C in constant diffuse light.

Hungarian Central Statistical Office provided us meteorological data (monthly temperature means (°C) and monthly precipitation sums (mm) for year 2011) for most of the analysed collecting sites in Hungary. Mlyňany Arboretum SAS (in Vieska nad Žitavou) dispose of its own meteorological station. From these data we calculated mean temperatures and precipitation sums for a period June–October 2011 (period of flowering, pod/seed establishment, pod/seed growth and ripening; see Table 2).

Experimental data were submitted to statistical analysis of variance (ANOVA) using Statgraphics Plus v.4.0 software. LSD tests at the 95% confidence level were performed to thousand seeds weight (TSW), seed

Table 2. Average meteorological data for the period June-October 2011 in most of the collecting sites

Locality	Nearest meteo station	Average temperature [°C]	Precipitation sum [mm]
Vieska n/Ž. (SK)	Vieska n/Ž. (SK)	17.4	307
Gyöngyös (HU)	Budapest (HU)	19.5	154
Debrecén (HU)	Debrecén (HU)	18.4	249
Mezőtúr (HU)	Kecskemét (HU)	18.9	212
Szeged (HU)	Szeged (HU)	19.1	116

projection area and thickness as well as germination parameters. Correlation analysis between respective parameters was accomplished using application MS Excell 2010.

Results

Seed coat colour of seeds from two northern collecting sites in Hungary (Szirák and Gyöngyös) showed larger variability than samples from Debrecén, Mezőtúr and Békéscsaba (Table 3). Only trees from alley near Szeged produced seeds uniform in colour. In Vieska nad Žitavou we found green-brown and medium brown seeds.

In weight of thousand seeds (TSW) LSD test distinguished three homogenous groups of samples: 1. with TSW round 220 g (Vieska nad Žitavou, Szirák and Békescsaba), 2. round 175 g (Gyöngyös and Szeged) and 3. intermediate seeds of TSW about 200 g (Debrecén and Mezőtúr). There was no statistical difference in projection area of seeds from respective collection sites. By seed thickness, localities can be ordered this way: 1. Debrecén, 2. Szirák, Gyöngyös, Szeged, 3. Vieska nad Žitavou, Mezőtúr and 4. Békéscsaba.

Naturally germinating seeds represent only a negligible portion from the total number of analysed seeds (Table 4): in Vieska nad Žitavou and Gyöngyösup up to 0.5%, in Debrecén, Mezőtúr and Szeged round 1%, in Szirák 1.35%, and in Békescsaba the portion slightly exceeded 1.5%. One hour pre-treatment by concentra-ted sulphuric acid almost completely released the germination process of any seed sample. We found values ranging from 95 to 98.5%. Only in samples from Debrecén and Békéscsaba less than 95% germinated seeds was identified. Germination energy of most of the samples showed similar level (approximately 9 mm d⁻¹). Extremes were observed only in Vieska nad Žitavou (8.09 mm d⁻¹) and Mezőtúr (10.56 mm d⁻¹).

In samples from Szeged we revealed the highest (9.15%) seed infestation by honey locust seed beetle (Table 5). In those from Békéscsaba almost 7.7%, in Gyöngyös and Debrecéna little more than 4%, in Szirák and Mezőtúr near 2.5% and in Vieska nad Žitavou no infested seeds were found. However, although ranging

Table 3. Parametrization of seeds from respective collecting sites. Abbreviations: TSW, thousand seeds weight; gb, greenbrown; mb, medium brown; db, dark brown; lb, light brown. Letters indicate a statistically significant difference at P = 0.05

Locality	Analyzed pods	Analyzed seeds	Seed coat colour	TSW [g]	Seed projection area [mm ²]	Seed thickness [mm]
Vieska n/Ž. (SK)	34	562	gb, mb	$*221.75 \pm 20.51b$	$57.57 \pm 3.75a$	$4.04\pm0.26bc$
Szirák (HU)	41	575	gb, lb, mb	$225.92\pm29.27b$	$59.64 \pm 7.84a$	$3.79\pm0.26ab$
Gyöngyös (HU)	33	463	gb, lb, mb	$173.49\pm14.90a$	$48.90 \pm 6.97 a$	$3.64\pm0.26ab$
Debrecén (HU)	33	519	gb, mb	$193.61\pm23.42ab$	$57.07 \pm 9.20a$	$3.48 \pm 0.37a$
Mezőtúr (HU)	43	848	gb, lb	$202.86 \pm 26.37 ab$	$51.91 \pm 5.92a$	$4.05\pm0.31bc$
Békéscsaba (HU)	31	598	lb, mb	$217.07 \pm 1.06b$	$49.47 \pm 3.87a$	$4.35\pm0.24c$
Szeged (HU)	44	973	mb	$175.96 \pm 3.99a$	$50.84 \pm 0.84a$	$3.58\pm0.08ab$

*Average \pm SD.

Table 4. Germination characteristics of honey locust seeds from respective collecting sites in Slovakia and Hungary. Letters indicate a statistically significant difference at P = 0.05

Locality	Germinated seeds [%]		Germination energy [m d ⁻¹]
	Non-pretreated	Pre-treated	
Vieska n/Ž. (SK)	0.50	*96.94 ± 4.33 <i>a</i>	$8.09 \pm 1.68a$
Szirák (HU)	1.34	$97.97 \pm 2.02a$	$9.30 \pm 0.30 ab$
Gyöngyös (HU)	0.33	$97.29 \pm 2.05a$	$9.23 \pm 0.35 ab$
Debrecén (HU)	1.07	$93.78 \pm 7.54a$	$9.00 \pm 1.21 ab$
Mezőtúr (HU)	1.00	$95.29 \pm 4.55a$	$10.56 \pm 1.89b$
Békescsaba (HU)	1.68	$92.54 \pm 11.13a$	$8.89 \pm 0.39 ab$
Szeged (HU)	1.01	$98.59 \pm 1.22a$	$9.22 \pm 0.30 ab$

*Average \pm SD.

from 24.5% (Vieska nad Žitavou) to 41.37% (Szeged) in average, no statistically significant difference in consumed part of endosperm across Danube basin was detected. And finally, no infested seed was able to germinate.

Performing correlation analyses, we found that TSW was strongly determined both by seed projection area and seed thickness (r values were a little higher than 0.6; Table 6). However, there was a weak negative relation between the latter two seed characteristics (r = -0.188). We also observed a strong negative correlation between TSW and mean temperature for June–October period (r = -0.897) and a strong positive one between this parameter and precipitation sum for the same period (r = 0.917). Seed projection area was strongly correlated with mean temperature (r = -0.918) and precipitation sum (r = 0.885). For seeds thickness we only observed moderate correlations to these meteorological parameters (r = -0.459 and r = 0.507, respectively). Despite of no relation of the percentage

of non-pretreated germinated seeds to seed projection area and mean temperature, moderate positive correlations of this characteristics to TSW (r = 0.449) and seed thickness (r = 0.389) as well as a weak one to precipitation sum (r = -0.117), were revealed. Percentage of pretreated germinated seeds was strongly related to precipitation sum (r = -0.544), moderately related to seed thickness (r = -0.467) but only weakly to the rest of parameters. Germination energy showed a strong correlation with mean temperature (r = 0.633) but a moderate one with precipitation sum (r = -0.434). However, it was not correlated with seeds characteristics. Except of seed thickness (r = -0.105), percentage of seeds infested by honey locust seed beetle was strongly related to all seed/weather characteristics. In the case of seed part consumed by beetle(s) we observed a strong negative correlation to TSW (r = -0.728), seed projection area (r = -0.676) and precipitation sum (r = -0.742). It was moderately related (r = 0.486) to mean temperature and weakly to seed thickness (r = -0.236).

Table 5. Percentage of infested honey locust seeds, consumed part of seeds and germinated infested seeds as influenced by collection locality. Letters indicate a statistically significant difference at P = 0.05

Locality	Seeds infested by seed beetle [%]	Consumed part of seeds [%]	Germinated infested seeds [%]
Vieska n/Ž. (SK)	0.00	_	_
Szirák (HU)	2.61	$*24.90 \pm 5.73a$	0
Gyöngyös (HU)	4.54	$39.53 \pm 4.45a$	0
Debrecén (HU)	4.05	34.11 ± 3.99 <i>a</i>	0
Mezőtúr (HU)	2.71	$30.32 \pm 21.79a$	0
Békescsaba (HU)	7.69	$37.76 \pm 9.05a$	0
Szeged (HU)	9.15	$41.37 \pm 1.49a$	0

 * Average \pm SD.

Table 6. Correlation coefficients (r) of relations between respective seed/weather parameters in honey locust populations for year 2011

	TSW	Seed projection area	Seed thickness	Mean June–October temperature	June–October precipitation sum
TSW	_	0.6***	0.655***	-0.897***	0.917***
Seed projection area	-	_	-0.188*	-0.918***	0.885***
Seed thickness	_	_	_	-0.459**	0.507***
Non-pretreated germinated seeds	0.449**	0.049	0.389**	0.056	-0.117*
Pre-treated germinated seeds	-0.251*	0.172*	-0.467**	0.24*	-0.544***
Germination energy	-0.01	0.03	0.051	0.633***	-0.434**
Infested seeds	-0.512***	-0.659***	-0.105*	0.69***	-0.894***
Consumed seed part	-0.728***	-0.676***	-0.236*	0.486**	-0.742***

*** - strong $(1 \ge r \ge 0.5)$, ** - moderate $(0.5 \ge r \ge 0.3)$ and * - weak linear regression $(0.3 \ge r \ge 0.1)$.

Discussion

Within following locations ordered by lowering geographic latitude we found decreasing diversity of seed coat colour (from green-brown, light brown and medium brown to medium brown). SCHOEPS (2002) defined that for this pigmentation chlorophylls, carotenes and xanthophylls are responsible. Since sampled trees were components of alleys, and since this species has polygamous character and it is fertilized by insects (CHRTKOVÁ and JASIČOVÁ, 1988; KOBLÍŽEK, 1995), gene flow between neighbouring individuals was expected. In the work of SCHNABEL and HAMRICK (1995) we can read about 17-30% minimum estimates of pollen gene flow, depending on maternal trees distance (round 100 and 200 m). However, in general seed coat colour is a trait, which is highly influenced by environmental conditions (SOUZA and MARCOS-FILHO, 2001).

ERTEKIN and KIRDAR (2010) studied effect of different seed coat colour on other seed characteristics of honey locust. They observed higher hundred seeds weight, seed coat weight, endosperm weight and embryo weight as well as germination in yellow coloured seeds compared with light and dark brown seeds. Despite of different seed coat colour scale, we can see similar trend in TSW but not in the percentage of germinated seeds. According to ASIEDU and POWELL (1998), slow rates of imbibition, caused by seed shrinkage and greater seed coat adherence to cotyledons during maturation, are associated with pigmentation. As indicated by GENEVE (2009), after dormancy release lens and micropyle function as a primary water gap for seed imbibition in honey locust as well as water locust (Gleditsia aquatica Marsh.).

BARNABÁS et al. (2008) summarize that high temperature and water deficit can impair ovary and embryo sac development, cause pollen sterility as well as fruit/ seed abortion or reduce their growth by restricted allocation of storage materials in cereals. This is in agreement with a general view of FENNER (2010) and MARTRE et al. (2011) on seed morphogenesis. Therefore, it is not a surprise that we found negative correlations of TSW, seed projection area and seed thickness with mean temperature as well as positive correlations of these seed parameters with precipitation sum in respective locations of Danube basin for a period from June till October, when reproductive cycle of honey locust has been accomplished. However, even in the warmest and driest regions stress was not enough intense to endanger seed germinability (GUTTERMAN, 1991). BORGES et al. (2005) focused on seed maturation process in Caesalpinia echinata Lam., a species relative to honey locust, at one place but in two years differing in rainfall during reproduction period. At the full fruit ripeness, seeds differed only in thickness (lower in drier year 2002).

MARCO and PAEZ (2000) present honey locust as a plant species conferring high potential to become invasive: fast growth, clonal and sexual reproduction, short juvenile period, high seed production and high seed germinability. This coincides with a general ranking of plant features associated with their invasiveness (Pyšek and Richardson, 2007). Compared with Acacia aroma Hook. et Arn., a native to Argentina, invasive honey locust disposes of higher seed production per plant, percentage of scarified seed germination and density of seedlings around the focal individuals (FERRERAS and GALETTO, 2010). HERRERA and LATERRA (2009) show in an ecological study from flooding Pampa grassland that addition of seeds of invasive species promoted seedling emergence, and this effect was higher for large than for small-seeded species. Similar results obtained EISENHAUER and SCHEU (2008) stating that established grassland community and invader seed size significantly affected the number of invader plants, while invader biomass was only affected by the established community. JAKOBSSON and ERIKSSON (2000) also found that the relative recruitment in undisturbed sward increased with increased seed size, and both recruitment success and seedling size were positively related to seed size. Although it was less strong, we revealed a correlation between the percentage of naturally germinated seeds and their TSW and thickness, as well. It can be explained by rising seed coat impermeability for water, associated with increasing adversity of environmental conditions during later stages of seed maturation (Souza and Marcos-Filho, 2001), responsible also for reductions in seed weight. But since germination energy was positively correlated to mean temperature and negatively to precipitation sum, seedlings of larger vigour can be expected in warmer and drier conditions. It is interesting that dark brown seeds, more frequently produced in such conditions, need much less water for germination than seeds of more lightly toned seed coats (ERTEKIN and KIRDAR, 2010). However, warmer and dried environment was associated with higher seed infestation by honey locust seed beetle (GYÖRGY, 2007; MAJZLAN, 2011; JERMY et al., 2002), which exclude them from the pool of potentially germinating seeds. So, this is not the case of relative Bruchidius dorsalis Fahraeus, considered as a crucial bio-agent providing germination of Gleditsia japonica Miq. seeds (TAKAKURA, 2002).

Thus, we can distinguish two different strategies supporting honey locust spreading: i) through higher germinability of larger seeds associated with lower temperatures and higher precipitation, and ii) through higher germination energy of smaller seeds connected with higher temperatures and lower precipitation. These results support the knowledge about high environmental plasticity of honey locust described by Godoy et al. (2011).

Acknowledgements

This work was supported by research projects Vega 2/0156/11, Vega 2/0085/09, Vega 2/0076/09 and SK-RO-0013-10. Special thanks to Dr. Pavol Eliáš for technical help.

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Received December 6, 2012 Accepted March 22, 2013

Diversity of flora in historical parks on example of Sokolow Podlaski Region in Poland

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Abstract

FORNAL-PIENIAK, B. 2013. Diversity of flora in historical parks on example of Sokolow Podlaski Region in Poland. *Folia oecol.*, 40: 170–175.

The paper is focused on diversity of flora in historical parks on example of Sokolow Podlaski Region. The research was done in 20 historical parks. Parks were established in XVIII–XIX centuries (landscape historic style) on oak-hornbeam habitat. Nowadays these parks are without anthropic pressure since II World War. The methodology included two research stages: field research and indoor studies. Research assumed flora and syntaxonomic analysis. The field research was done in years 2010–2012, including 70 phytosociological records on the area 500 m². Plant species are represented by natural, semi-natural and synantropical vegetation. There was observed impact for migration species from surroundings areas in study areas.

Keywords

flora, historical parks, oak-hornbeam habitat, Sokolow Podlaski Region

Introduction

Agricultural landscape is as a 'sea' with 'green islands' (BUČEK et al., 1996). Manor parks are sometimes the last sites for existing forest plants in agricultural landscape (OLACZEK, 1972). Many plants and animal species typical for agricultural landscape are rare and on the verge of extinction (ROBINSON and SUTHERLAND, 2002; HERMY and STIEPERAERE, 1981). Studies concerning historical parks mostly include issues related to a dendrological inventory, the history of the manor parks' ownerships, objects' condition and parks' cultural values and functions alongside with a proposal of their protection and restoration. There are no many studies about flora in these objects. The aim of the study is diversity of flora in historical parks on example of Sokolow Podlaski Region.

Material and methods

Sokolow Podlaski Region (137.18 km² surface) is located on east part of Poland. This area belongs to Mazowiecko-Poleski section according to MATUSZKIEWICZ (1993).

The research was done in 20 historical parks in Sokolow Podlaski Region (Fig. 1). Parks were established in XVIII and XIX centuries (landscape historic style) on oak-hornbeam site.



Fig 1. Location of study area.

The methodology included two research stages: field research and indoor studies. The field research was done in years 2009–2011. 70 phytosociological records on the area 500 m² (BRAUN-BLANQUET, 1964) were done on afforested areas of parks. Plant species were grouped by phytosociological system following MATUSZKIEWICZ (2001). The next stage of work was vegetation evalua-

tion of manor parks. Vegetation evaluation included 11 criteria as number of tree species in tree layer, origin of trees, types of plantings, tree-covered areas (%), health of plantings, number of shrub species, origin of shrubs, shrub-covered areas (%), number of herb species, origin of herb species, herb-covered areas (%) and bonitation from 0 to 3 points (Table 2). Parks were grouped

Criteria		Bonitation points
	Tree layer	
	Above 6 tree species	3
Number of tree species	4–5 tree species	2
	1-3 tree species	1
	n – native species	3
Origin of trees (dominated in park)	c – cultivator species (planted trees)	2
(dominated in park)	ex-exotic species (introduced artificial)	1
	Avenues, group of trees, individual trees	3
Types of plantings	Group of trees, individual trees	2
	Only group of trees	1
	Above 50%	3
Tree-covered areas [%]	25%-50%	2
	1%-24%	1
	Good (mostly without canopy losses, tree hollows, diseases)	3
Health of plantings	Medium (sometimes with canopy losses, tree hollows, diseases)	2
	Bad (many canopy losses, tree hollows, diseases)	1
	Shrub layer	
Number of shrub species	Above 6 plant species	3
	4–5 plant species	2
	1-3 plant species	1
	n – native species	3
Origin of shrubs (dominated in park)	c – cultivator species (planted shrubs)	2
(dominated in park)	ex-exotic species (introduced artificial)	1
	Above 50%	3
Shrub-covered areas [%]	25%-50%	2
	1%-24%	1
	Herb layer	
	Above 6 plant species	3
Number of herb species	4–5 pant species	2
	1–3 plant species	1
Origin of herb species	n – native species (apophytes, spontanophytes)	3
(dominated in park)	c – alien species (antropophytes)	1
	Above 50%	3
Herb-covered areas [%]	25%-50%	2
	0%-24%	1

Table 1. Vegetation evaluation of historical parks

into fours groups: parks with high vegetation values (from 25 to 33 points), parks with medium vegetation values (from 18 to 24 points), parks with low vegetation values (from 8 to 17 points) and parks with very low vegetation values (from 0 to 7 points) (Table 1).

 Table 2. Plant species in tree layer in manor parks in Sokolow Podlaski Region

Latin name	Syntaxonomic class
Acer campestre L.	Querco-Fagetea
Acer platanoides L.	Querco-Fagetea
Acer pseudoplatanus L.	Querco-Fagetea
Acer pseudoplatanus Atropurpureum L	Companion species
Aesculus hippocastanum L.	Companion species
Alnus glutinosa L.	Salicetea purpureae
Betula pendula Roth.	Companion species
Carpinus betulus L.	Querco-Fagetea
Corylus avellana L	Querco-Fagetea
Euonymus verrucosa Scop.	Querco-Fagetea
Fagus sylvatica L.	Querco-Fagetea
Fraxinus excelsior L.	Querco-Fagetea
Malus domestica Borkh.	Companion specie
Padus avium Mill.	Querco-Fagetea
Populus alba L.	Salicetea purpureae
Prunus avium (L.) Moench.	Querco-Fagetea
Prunus domestica L.	Companion species
Prunus mahaleb (L.) Mill	Companion species
Quercus robur L.	Companion species
Quercus rubra L.	Companion species
Robinia pseudoacacia L.	Companion species
Sambucus nigra L.	Epilobietea angustifolii
Tilia cordata Mill.	Querco-Fagetea
Ulmus laevis Pall.	Querco-Fagetea

Results and discussion

Plant species recognized in manor parks were represented by 7 plant communities of eutrophic forest community (Querco-Fagetea), riparian forest and brush of river valley (Salicetea purpureae) coniferous forest communities (Vaccinio-Piceetea), cut-over communities (Epilobietea angustifolii), bush communities (Rhamno-Prunetea), meadow and pasture communities (Molinio-Arrhenatheretea), margin communities, ruderal communities (Artemisietea vulgaris) and companion plant species (Tables 2–4). There have occurred plant species from all 3 syntaxonimic classes in tree layer. In shrub, there were distinguished plant species from 5 plant communities and 6 plant communities in herb layer.

Table 3.	Plant species in shrub layer in historical	parks
	in Sokolow Podlaski Region	

Latin name	Syntaxonomic class
Abies alba Mill.	Vaccinio-Piceetea
Acer campestre L.	Querco-Fagetea
Acer platanoides L.	Querco-Fagetea
Acer pseudoplatanus L.	Querco-Fagetea
Aesculus hippocastanum L.	Companion species
Betula pendula Roth	Epilobietea angustifolii
Carpinus betulus L.	Querco-Fagetea
Corylus avellana L.	Querco-Fagetea
Crataegus monogyna Jacq.	Rhamno-Prunetea
Euonymus verrucosa Scop.	Querco-Fagetea
Fagus sylvatica L.	Querco-Fagetea
Fraxinus excelsior L.	Querco-Fagetea
Padus avium Mill.	Querco-Fagetea
Philadelphus coronarius L.	Companion species
Populus alba L.	Salicetea purpureae
Quercus robur L.	Companion species
Ribes rubrum L.	Companion species
Robinia pseudoacacia L.	Companion species
Rubus idaeus L.	Epilobietea angustifolii
Sambucus nigra L.	Epilobietea angustifolii
Sorbus aucuparia L.	Companion species
Syringa vulgaris L.	Companion species
Taxus baccata L.	Companion species
Thuja occidentalis L.	Companion species
Tilia cordata Mill.	Querco-Fagetea
Ulmus laevis Pall.	Querco-Fagetea

Table 4. Plant species in herb layer in historical parks in Sokolow Podlaski Region

Latin name	Syntaxonomic class
Acer platanoides L.	Querco-Fagetea
Acer pseudoplatanus L.	Querco-Fagetea
Aegopodium podagraria L.	Querco-Fagetea
Aesculus hippocastanum L.	Companion species
Ajuga reptans L.	Companion species
Allium ursinum L.	Querco-Fagetea
Anemone nemerosa L.	Querco-Fagetea
Anemone ranunculoides L.	Querco-Fagetea
Asarum europaeum L.	Querco-Fagetea
Carex pilosa Scop.	Querco-Fagetea
Carex umbrosa L.	Querco-Fagetea
Carpinus betulus L.	Querco-Fagetea
<i>Cerastium sylvaticum</i> Waldst. & Kit	Artemisietea vulgaris
Chelidonium majus L.	Artemisietea vulgaris

Convallarja maialis L	Companion species
Corydalis cava L.	Querco-Fagetea
Corylus avellana L.	Querco-Fagetea
Dactylis glomerata L.	Molinio-Arrhenatheretea
Dactylis polygama Horv	Querco-Fagetea
Euonymus verrucosa Scop	Querco-Fagetea
Fagus sylvatica L.	Querco-Fagetea
Ficaria verna Huds.	Querco-Fagetea
Fragaria vesca L.	Epilobietea angustifolii
Gagea lutea L.	Querco-Fagetea
Galeobdolon luteum Huds.	Querco-Fagetea
Galium schultesii L.	Querco-Fagetea
Galium sylvaticum L.	Artemisietea vulgaris
Geum urbanumL.	Artemisietea vulgaris
Glechoma hederacea L.	Artemisietea vulgaris
Hedera helix L.	Companion species
Impatiens noli-tangere L.	Querco-Fagetea
Impatiens parviflora L.	Artemisietea vulgaris
Lamium album L.	Artemisietea vulgaris
Lamium maculatum L.	Artemisietea vulgaris
Lamium purpureum L.	Artemisietea vulgaris
Lathyrus vernus (L.) Bernh.	Querco-Fagetea
Luzula pilosa (L.) Willd.	Companion species
Lysimachia nummularia L.	Molinio-Arrhenatheretea
<i>Maianthemum bifolium</i> (L.) Schmidt	Companion species
Milium effusum L.	Querco-Fagetea
Oxalis acetosella L.	Companion species
Plantago major L.	Molinio-Arrhenatheretea
Poa nemoralis L.	Querco-Fagetea
Polygonatum multiflorum L.	Querco-Fagetea
Prunella vulgaris L.	Molinio-Arrhenatheretea
Prunus avium L.	Querco-Fagetea
Pulmonaria officinalis L.	Querco-Fagetea
Quercus robur L.	Companion species
Robinia pseudoacacia L.	Companion species
Rubus caesius L.	Rhamno-Prunetea
Sambucus nigra L.	Epilobietea angustifolii
Scilla bifolia L.	Querco-Fagetea
Sorbus aucuparia L.	Companion species
Stachys sylvatica L.	Querco-Fagetea
Stellaria holostea L.	Querco-Fagetea
Taraxacum officinale L.	Molinio-Arrhenatheretea
Tilia cordata L.	Querco-Fagetea
Trientalis europaea L.	Vaccinio-Piceetea
Urtica dioica L.	Artemisietea vulgaris
Vinca minor L.	Querco-Fagetea
Viola mirabilis L	Companion species

Most of the species were represented by the community from eutrophic forest community (Querco-Fagetea) in all layers (tree layer – 55%, shrub layer – 40% and herb layer – 55%). Percentage of cover plant species from Rhamno-Prunetea and Vaccinio-Piceetea classes was not very high in all parks (Figs 2–4).

There were distinguished many native plant species such as: Acer pseudoplatanus, Carpinus betulus, Fagus sylvatica, Euonymus verrucosa, Corylus avellana, Galeobdolon luteum.

Plant species of eutrophic forest community like: *Carpinus betulus, Milium effusum* which are typical for oak-hornbeam habitat. *Tilia cordata, Fraxinus excelsior, Anemone nemerosa, Gagea lutea, Galeobdolon luteum, Corydalis cava,* dominated in all parks. Plant species from Epilobietea angustifolii class are represented by: *Sambucus nigra, Betula pendula* and *Rubus idaeus*. Percentage of cover plant species from Rhamno-Prunetea and the other synthaxonomic classes was not very high in all parks.



Fig. 2. Percentage cover of plant species in different syntaxonomic unities in tree layer in historical parks.

Vegetation evaluation included eleven criteria: number of plant species in tree layer, origin of trees, types of plantings, tree-covered areas, health of plantings, number of plant species in shrub layer, origin of shrubs, shrub covered areas, number of plant species in herb layer, origin of herbs, herb covered areas (Table 5).

High diversity of native plant species in tree, shrub and herb layer was observed in the park objects. Individual trees and groups of trees were typical plantings on study parks. Eight parks with high vegetation values and twelve parks with medium vegetation values were distinguished.

Flora of manor parks is still modified by human and nature processes (SIKORSKI and WYSOCKI, 2003). Woody plant species were noticed in parks by many scientists e.g. DZWONKO and LOSTER (2001), FORNAL-PIE-NIAK and WYSOCKI (2006, 2009). Plants from Querco-Fagetea occur in Sandomierska Basin park's herb layer (FORNAL-PIENIAK, 2007) and Sokolow Podlaski Region. There were also observed plant species from Molinio-Arrhenatheretea and Trifolio-Geranietea sanguinei.







Fig 4. Percentage cover of plant species in different syntaxonomic unities in herb layer in historical parks.

radie 5, i funding evaluation of manor parks in bollot i outable region	Table 5.	Planting	evaluation	of manor	parks in	Sokolow	Podlaski	Region
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Criteria/number of parks	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
							А	Tree la	iyer											
Number of plant species	2	2	2	1	3	2	2	3	2	3	2	2	2	2	2	3	2	3	2	3
Origin of trees (dominated)	1	2	3	1	3	1	1	1	2	3	3	2	2	1	2	1	2	3	1	1
Types of plantings	2	3	1	2	3	3	2	1	2	3	3	2	2	2	3	1	2	3	2	1
Tree-covered areas	2	2	2	2	3	3	2	2	2	3	3	2	2	2	2	2	2	3	1	1
Health of plantings	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
Sum	9	11	10	8	14	13	9	9	10	14	13	10	10	9	11	9	10	14	8	8
							В	Shrub	ayer											
Number of plant species	2	2	2	1	2	2	2	1	2	2	2	2	2	2	2	2	2	2	2	2
Origin of shrubs (dominated)	3	2	2	2	3	2	3	2	2	3	3	3	3	3	3	3	3	3	3	3
Shrub covered areas	1	1	1	1	1	2	2	1	1	1	1	1	1	1	1	1	1	1	1	1
Sum	6	5	5	4	6	6	4	4	5	6	6	6	6	6	6	6	6	6	6	6
							С	Herb 1	ayer											
Number of plant species	3	3	3	3	3	2	3	3	2	3	3	3	3	3	3	3	3	3	3	3
Origin of herbs (dominated)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
Herb covered areas	3	3	2	3	3	3	2	3	3	3	3	3	3	3	3	3	3	2	3	3
Sum	9	9	8	9	9	8	8	9	9	9	9	9	9	9	9	9	9	8	9	9
Sum A + B + C	24	25	23	21	29	27	21	22	23	29	28	25	25	24	26	24	25	28	23	23

33-25 points: parks with high vegetation values; 24-18 points: parks with medium vegetation values; 17-8 points: parks with low vegetation values; 7-0 points: parks with very low vegetation values.

Nowadays we have very little information about condition of vegetation in historical parks in Poland. Many dendrological field researches were done but without list of plant species in herb layer. This pilot study is to show the vegetation evaluation diversity of historical parks in Sokolow Podlaski Region in Poland.

Acknowledgement

This research project would not have been possible without the support of prof. Czesław Wysocki and prof. Jan Supuka.

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Received December 6, 2012 Accepted May 13, 2013

Differentiation of some interspecific hybrids of firs (*Abies* sp.) according to the length of primary branches and number of their secondary branches

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Abstract

GALGÓCI, M., MAŇKA, P., KORMUŤÁK, A., ČAMEK, V., GÖMÖRY, D. 2013. Differentiation of some interspecific hybrids of firs (*Abies* sp.) according to the length of primary branches and number of their secondary branches. *Folia oecol.*, 40: 176–180.

During 2011, the length of primary branches was measured in individual seedlings of firs representing 15 crossing variants. Measured branches were divided into separate groups according to the number of secondary twigs. Our data indicate the possibility for differentiation between the hybrid combinations based on length of their primary branches. The interspecific combinations *A. pinsapo* \times *A. alba* and *A. alba* \times *A. pinsapo* were more similar to mother species in this trait rather than to paternal parent. A given combination of different age differed primarily by the number of primary branches with a higher number of secondary branches in older seedlings. Comparison involving both primary and secondary branches appears to be more efficient in discriminating between hybrid combinations than comparison primary based on secondary branches alone.

Keywords

Abies, branches, differentiation, hybrids, somatic heterosis

Introduction

The genus *Abies* Mill. with its 49 species and about 126 known interspecific hybrids demonstrates the importance and potential opportunities for interspecific hybridization in forest tree breeding (GREGUSS, 1995; KORMUŤÁK, 1994, 2004).

Generally, it is concluded that extensive crossability between representatives of the genus *Abies* is a result of their high genetic relatedness due to specific pattern of speciation (GREGUSS, 1995). KLAEHN and WINIENSKI (1963) consider this to be result of geographic rather than genetic isolation. Recent studies also point to the fact that crossability in the genus is the result of geographical isolation, not genetic isolation (KORMUŤÁK, 2004; KORMUŤÁK et al., 2012; KORMUŤÁK et al., 2013). A high level of genetic diversity in interspecific hybrids is the result of heterozygous gene loci which leads to various forms of heterosis in hybrids (ROHMEDER and SCHÖNBACH 1959; MERGEN et al., 1964).

Increased resistance to pests and diseases (MÜLLER, 1989; ROHMEDER and SCHÖNBACH, 1959) along with increased vitality and adaptability of the hybrids to changing conditions of the environment is generally referred to as adaptive heterosis (GREGUSS, 1995; AUSENAC, 2002). It may also involve a higher proportion of surviving individuals, especially in relation to either immission load of the environment (ČÍTKOVÁ, 1988; RIŠKOVÁ, 1982; EVANS and MÜLLER, 1972) or increasing annual temperature (AUSENAC, 2002).

Reproduction or seed heterosis is the ability of the offspring to produce increased amount of viable seeds

compared to the parental species. This applies in seed orchards and natural sites where adverse environmental conditions reduce the number of viable seeds (ULJUKINA and DERJUTKIN, 1981; GREGUSS, 1995; KORMUŤÁK, 1994, 2004).

Somatic heterosis of interspecific fir hybrids is the most commonly observed phenomenon which is directly related to the enhanced production potential of forest trees (HAWLWEY and DE HAYES, 1985a, 1985b; KORMUŤÁK, 1994, 2004; ROHMENDER and EISENHUT, 1961; KLAEHN and WINIENSKI, 1963; MERGEN et al., 1964).

The papers dealing with somatic heterosis in hybrid firs have been focused preferentially on height growth neglecting other growth characteristics of the hybrids. Considerable variation of morphological traits in fir hybrids makes a problem with their taxonomic identification (GREGUSS, 1995; KORMUŤÁK, 1994, 2004). In present work we have tried to create a system of taxonomic differentiation of selected interspecific fir hybrids based on morphometric features of their primary branches.

Material and methods

In summer 2011, the length of primary branches in seedlings of some interspecific combinations of firs and in control variants of the parental species from open pollination and controlled cross pollination were measured. We used hybrids of the following species: Abies alba Mill., Abies pinsapo Boiss., Abies numidica de Lannoy ex Carrière., Abies nordmanniana (Steven) Spach, Abies procera Rehder, Abies holophylla Maxim. We have analyzed 4-year old seedlings of A. pinsapo \times A. numidica, A. pinsapo \times A. alba, A. alba \times A. pinsapo, A. alba - controlled crossing, A. alba - open pollination and A. pinsapo - open pollination along with 6-year old seedlings of A. nordmanniana × A. numidica, A. nordmanniana \times A. procera, A. nordmanniana \times A. alba, A. alba \times A. numidica, A. alba – open pollination, A. nordmanniana - open pollination and 7-year old seedlings of A. nordmanniana × A. holophylla, A. nordmanniana \times A. alba and A. nordmanniana – open pollination. Seedlings were grown in nurseries of Mlyňany Arboretum SAS in spaced 20×20 cm. They were also



Fig. 1 Branches of the first and second degree.

grown in standard light conditions without shading. Length measurement was carried out with the help of a ruler Fig. 1. Measured branches of the first degree were divided into groups according to the number of their secondary branches. These groups were statistically evaluated and tested by nested ANOVA (SAS, 1988).

Results and discussion

There were applied various approaches in characterization of intra- and interspecific hybrids of firs involving height growth parameters, morphology and anatomy of needles (LARSEN, 1934; KLAEHN and WINIESKI, 1962; CRITCHFIELD, 1988), pollen viability and seed quality (GALgóci, 2010), photosynthesing pigments, isoenzymes, terpenes and DNA (GAUDLITZ, 1983; ZAVARIN et al., 1977; WAGNER et al., 1987; DONG and WAGNER, 1994). The length of branches as morphological trait has not been used so far. Using Duncan grouping we were able to differentiate between some crossing variants according to the primary branches length and number of their secondary twigs (Table 1). It is of interest to mention that testing of individual groups of primary branches in 6- and 7-year old seedlings has not affected the descending order of the variants given in Table 1. Even a change of the order has not any statistical impact on differentiation of the crossing variants according to the type of their branches. It follows, therefore, that it does not matter which length parameter is used as a criterion for differentiation. In 6- and 7-year old seedlings both primary and secondary branches length characteristics may be used. It was not possible to discriminate between 7-year old seedlings of the crossing variants on the basis of length characteristics of the first three groups of the primary branches with 0-2 secondary twigs. Efficient in this respect were only the primary branches with three secondary twigs which had clearly differentiated the crossing variant A. nordmanniana \times A. alba. In the category of primary branches with four secondary twigs, we have distinguished crossing variants A. nordmanniana \times A. holophylla and A. nordmanniana - open pollination. Similar discrimination was also possible between 6-year old seedlings. The only exceptions in this respect were the variants A. alba – open pollination and A. nordmanniana \times A. procera which had not complied discrimination criteria mentioned above (Table 1). Comparison of our data with those published by GALGÓCI et al. (2011) which refer to the height growth characteristics of the 6- and 7-year old progenies of the same crossing variants revealed a high degree of correlation with respect to the 7-year old seedlings only. At the 6-year old seedlings level the authors detected statistically significant differences between the pair of crossings A. nordmanniana \times A. numidica – A. nordmanniana \times A. alba and A. $nordmanniana \times A$. numidica A. nordmanniana – open pollination. Quite different situation was observed at

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the stage of 4-year old seedlings. At each group of the analyzed primary branches with corresponding number of twigs, the order of crosses has changed in this case at statistically significant level starting from the longest primary branch to the shortest one. The findings in the 5 groups scale of primary branches were those which had enabled to differentiate the 4 year old seedlings of all the analyzed crossing variants based on their primary branches. Comparison of the length characteristics in two different age categories of A. nordmanniana - open pollination seedlings have not revealed statistically significant differences in average lengths of the primary branches in 6- and 7-year old progenies. Both these age categories were represented by the seedlings with primary branch containing maximum 2 twigs. In the crossing variant A. nordmanniana × A. alba statistical differences between 6- and 7- year old seedlings exist only in the group of primary branches without and/or with one secondary twig. Seven year old seedlings are involved in the group of primary branch with 3 and 4 secondary twigs. Six year old seedlings have not occurred in this group. Among 6- and 4-year old seedlings of A. alba - open pollination and controlled crosses of A. alba maternal species statistically significant differences were revealed between seedlings of the same age but not between seedlings of different age categories. We have not observed such phenomenon in other groups of primary branches.

Mutual comparison of the growth characteristics in the crossing variants A. pinsapo - open pollination 2007, A. alba – open pollination 2007, A. pinsapo \times A. alba and A. alba \times A. pinsapo resulted in the conclusion that variants A. *pinsapo* – open pollination and A. *pinsapo* \times *A. alba* differ significantly in several groups of the primary branches from the variants A. alba open pollination and A. alba \times A. pinsapo. As a rule, the hybrids have exhibited the tendency to be similar in this respect to mother tree. The phenomenon may be ascribed to the matrilinear inheritance of growth characteristics. A typical feature of crossing variants with arid species A. numidica and A. pinsapo involved the parental specimen in the formation of secondary twigs in relatively young plants as well as in plants with short primary branches. It is supposed that this feature of the hybrids is related to the ability of the parental species to invade new localities.

Acknowledgement

This study was supported by the VEGA Grant Agency, projects no. VEGA 2/0076/09, and 2/0110/13.

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Combining crossing – controlled variant /year of planting	ΤN	NB0	$\overline{x}0$	DG0	DGI	NB1	$\overline{x}1$	NB2	$\overline{x}2$	DG2	NB3	<u>x</u> 3	DG3	NB4	<u>x</u> 4	DG4
A. nordmanniana × A. holophylla (2004)	40	212	6.54	A	51	13.38	A	48	15.11	A	14	19.19	A	11	22.42	A
A. nordmanniana – open pollination (2004)	12	64	7.23	A	5	13.3	A	10	14.95	A	Ι	I	I	Ι	I	I
A. nordmanniana – open pollination (2005)	15	54	7.04	AB	13	12.29	AB	14	14	AB	I	I	I	I	I	I
A. $alba - open pollination (2005)$	57	152	7.02	AB	71	11.59	AB	83	13.93	AB	32	16.54	BC	6	15.3	в
A. $alba \times A$. $alba$ (2007)	28	130	6.97	AB	15	7.89	DC	I	I	I	I	I	I	I	I	I
A. nordmanniana \times A. alba (2004)	29	163	6.78	ABC	58	11.23	AB	73	13.04	ABCD	16	17.43	AB	13	18.55	В
A. pinsapo \times A. alba (2007)	33	624	6.46	ABCD	56	11	В	114	13.21	ABCD	6	12.13	EFG	9	15.65	В
A. nordmanniana × A. procera (2005)	47	229	6.08	BCD	78	11.18	AB	117	12.16	BCD	20	16.62	ABC	12	16.51	В
A. pinsapo $-$ open pollination (2007)	28	224	5.8	CDE	27	9.93	BC	41	13.46	ABC	17	16.07	BCD	×	16.22	В
A. alba × A. numidica (2005)	27	134	5.78	DE	41	10	BC	24	11.24	Ð	8	13.54	DEF	Ζ	16.14	В
A. nordmanniana × A. numidica (2005)	30	169	5.46	Е	34	9.86	BC	11	11.25	8	9	14.32	CDE	I	I	I
A. nordmannniana $ imes$ A. alba (2005)	29	189	4.84	FE	29	9.45	C	19	10.98	D	Ι	I	I	Ι	I	I
A. $alba - open pollination (2007)$	18	74	4.43	FG	22	6.17	DE	10	11.25	Ð	I	I	I	I	I	I
A. alba \times A. pinsapo (2007)	4	26	3.78	GH	6	4.9	Щ	I	I	I	4	11.4	FG	I	I	I
A. pinsapo × A. numidica (2007)	10	87	3.23	Н	4	6.48	D	L	8.12	Щ	9	9.85	Ð	I	I	I

Table 1. The differentiation of interspecific hybrids of firs on the basis of number of secondary twigs on the primary branches

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Received December 6, 2012 Accepted March 27, 2013

Study of the richest gene pool of trees and shrubs in Slovakia, in the Mlyňany Arboretum SAS

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Abstract

HOŤKA, P., BARTA, M., BIBEŇ, T. 2013. Study of the richest gene pool of trees and shrubs in Slovakia, in the Mlyňany Arboretum SAS. *Folia oecol.*, 40: 181–187.

An inventory of the gene pool of woody plants in the Arboretum Mlyňany SAS was carried out in years 2001–2011. The results were summarized in 2012 to provide a data base for complete digitalisation of the living collections. This work discusses the history of introduction activities in the Arboretum, aged 120 years to this date. There are compared the results of introduction among the essential phases of building the woody plant collections. We discuss the characteristics of introduction of evergreen woody plants by the count Ambrózy-Migazzi (1892–1914), the phases of development of the research area of this academic institution from the year 1953 to the climax in the last 1990s, as well as the current state of its living collections. There are outlined possibilities for introducing new species into this park object.

Key words

Inventory of Living Collections, Mlyňany Arboretum SAS

Introduction

The woody plant collections in the Arboretum Mlyňany date their origin 120 years ago. Established in conditions of the historical Austro-Hungarian Monarchy, later they represented one of the most important sources of the gene pool of Central European woody plants in frame of the former Czechoslovak Republic. Their importance has been maintained until today. Currently, the woody plant collections in the Arboretum Mlyňany of the Slovak Academy of Sciences belong to the leading ones of this type in Slovakia. Their relevance is primarily for science. They provide the source of the study material for investigation of the acclimation process in exotic woody plants and they also serve for educational and recreational purposes. Thank to the considerable diversity of the plant material assorted on individual original plots, the park subject is very attractive, visited by a large number of visitors, all the year around.

The collections in the Arboretum were progressively extended. The present state is the result of the philosophy applied at their establishment, and the following intensive and high-quality management. The Arboretum Mlyňany was built as an evergreen park by the count Štefan Ambrózy-Migazzi (with the slogan Semper Vireo - I am ever green), intended to assemble as much as possible evergreen and semi-deciduous woody plants in the understorey of the original forest stand consisting of the Turkey oak and hornbeam. It was an unprecedented idea - to introduce sempervirent species in the foothills of the Carpathians. Later, after the management of the collections which had been transformed to the Slovak Academy of Sciences, the project of sempervirent species introduction was extended with research of introduction and adaptation of all promising exotic woody plants and their assortment on so called phyto-geographic plots. The principal goal was to enrich the gene pool of domestic woody plants primarily with woody plants suitable for use in forestry and with woody plants suitable for use in settlement greenery and landscape creation, thanks to their high aesthetic values. The plant material in the Arboretum Mlyňany was used in study of a number of scientific projects dealing with issues of taxonomy, ecology, physiology, genetics, phytopathology, garden and park architecture, landscape architecture and settlement greenery.

The overall inventories of the living collections of trees and shrubs carried out in the course of history of the Arboretum Mlyňany, mainly in occasions of its anniversaries, reflected the actual state of collections at the given moment and the results the institution had recorded in woody plant introduction to that moment. They outlined next possibilities for introduction activities. The results of the inventories should serve for creation of a comprehensive database for the cultivated assortment. There should also be carried out complete digitalisation of the data gathered in the field, supplementing the list of the grown woody plant taxa. For the upcoming years in the Arboretum Mlyňany SAS, such synthetic knowledge is critically important as it will facilitate the access to the collections for scientific purposes as well as for educational activities.

Material and methods

In the years 2001-2011, a series of inventories, were carried out in the individual departments of the Arboretum Mlyňany SAS, on the area of nearly 67 ha. The Arboretum consists of the original Ambrózy evergreen park (ca 40 ha, Departments P1 to P56), phyto-geographic plots and supplementary plots. The phyto-geographic plots comprise: Plot of East-Asian woody plants (ca 14 ha, Departments A1 to A23), Plot of North-American woody plants (ca 7.5 ha, Departments S1 to S7) and Plot of Korean woody plants (ca 4.5 ha, Departments K1 to K7), the other plots are the Plot of autochthonous woody plants of Slovakia (ca 1.5 ha, in parts of Departments P42, P43, P44, P45, P46, P48 and P51), Rosarium (ca 1 ha, a part of plot K6) and smaller representative plots (collection of decorative woody plant cultivars, a patch of conifers, a patch of sempervirent species and a bed of ever-flowering taxa).

The aim of the inventory was not only to identify the taxa on individual plots but also to identify their origin: agenda *Index seminum* (free exchange of seed material among botanical institutions at the global level), expedition activities, gift or purchase from decorative plant nurseries.

The assessment of the current condition of the woody plants in individual departments was carried out with using the results of the previous inventories, the works by NÁBĚLEK (1958), BENČAŤ (1967), TÁBOR and TOMAŠKO (1992), background data from the seed boxes (since 1959) and plant charts (HOŤKA, 2004; HOŤKA and FOGADOVÁ, 2008). The accent was also put on assessment of the results of introduction of individual woody plant groups (conifers, evergreen and semi-deciduous broadleaf and deciduous broadleaf species)

in the individual periods of the history of the Arboretum Mlyňany: i) the period of the founder of the Arboretum and the period after his departure when the development of collections of sempervirent species was managed by the horticulturist Mr. Mišák (1894–1925), documented by TABOR and TOMAŠKO (1992), ii) the period between WWI and WWII before incorporating the Arboretum in SAS (1926–1953), documented by BENČAŤ (1967), iii) inventory of the flora in the Arboretum documented by NÁBĚLEK (1958), iv) the results of inventory at the 75th anniversary of the establishment of the Arboretum Mlyňany documented by BENČAŤ (1967), v) the results of inventory at the 100-th anniversary of the starting of the Arboretum Mlyňany documented by Tábor and TOMAŠKO (1992), vi) results of the current inventory reflecting the development of collections after year 1993.

In this paper we present, apart from basic data (overall state of gene pool of woody plants), the results concerning the introduction of sempervirent and hiemirent taxa (evergreen and semievergreen woody plants).

In the text and tables, the plant categories are labelled with the following abbreviations: species - sp., subspecies - ssp., variety - var., forma - f., and cultivar - cv.

The number of taxa means the number of species and their infraspecific categories.

The botanical nomenclature used in this paper, during the recent inventory as well as during the preceding inventories, mostly follows Rehder (REHDER, 1949) and Krüssmann (KRÜSSMANN, 1976, 1977, 1978 and 1983). The plant names in the text and in the tables are without abbreviations of their authors.

Results and discussion

The results of the recent inventory of the gene pool of trees and shrubs finished in 2012 are listed in Table 1.

 Table 1. Taxonomic profile of the living collections of the

 Arboretum Mlyňany SAS in 2012

Category	Number	%
Species	1,107	57.3
Subspecies	11	0.6
Variety	180	9.3
Forma	18	0.9
Cultivar	617	31.9
Total	1,933	100.0

Comparisons of these results with the results of former inventories carried out in 1967 and 1992 show that the current number of taxa grown in the Arboretum today is by 250 less than in year 1992 (TABOR and TOMAŠKO, 1992), however by 275 more than in year 1967 (BENČAŤ, 1967). The number of species (botanical species and hybrids obtained by cultivation) in year 2012 was by 360 less than in year 1992 (TÁBOR and TOMAŠKO, 1992) and by 65 species less than in 1967 (BENČAŤ, 1967) (Table 2).

Table 2. Comparison between results of current inventoryand previous ones published in 1967 nad 1992

Year	1967	1992	2012
Family	78	93	83
Genus	272	294	235
Species	1,172	1,467	1,107
Subspecies	-	6	11
Variety	191	78	180
Form	58	5	18
Cultivar	237	627	617
Total	1,658	2,183	1,933

From the total number, the currently grown cultivars represent almost 32% of the woody plants assortment, while in 1992 it was 29% and in 1967 a little more than 14%. The number of cultivars in the Arboretum Mlyňany SAS has increased mainly thanks to the extension of the rosarium plot and introduction of new rose cultivars in 2004. The numbers of cultivars exhibit distinct increasing trends also in several other botanical collections in Slovakia and abroad thanks to constantly increasing numbers of ornamental cultivars that were given preference in many collections with the aim to give the expositions as much attractive look as possible. The raised number of cultivars in the recent inventory also reflects the changes in taxonomical classification of botanical items – today many of former va-

rieties and forms are recognised as clones or cultivars. However, we have identified a number of botanical species. Today, the collections of the Arboretum Mlyňany SAS comprise 180 varieties of woody plants.

Since 1992, there have been several distinct changes, primarily concerning presence of families and selected genera (Table 2). Until 1992, the collections in the Arboretum contained in overall 93 woody plant families, by 10 more than their present number (83). The number of genera showed a decrease by 59 compared with the year 1992. This may be due to several fragile, mostly evergreen species experimentally introduced to the Arboretum from plant nurseries in 1992. Many of these species were extinct because they could not tolerate less favourable climatic conditions in the Arboretum. Another negative phenomenon was growing several, mainly monotypic genera in only a few exemplars. Such genera were the most vulnerable against adverse ecological conditions and also against flaws in the cultivation process. From the total number of 27 of very rarely cultivated woody plant taxa in the collections of the Arboretum, 20 are represented each only by one single living exemplar today (HoŤKA, 2011).

In terms of woody plant groups – coniferous, evergreen, semi-deciduous and deciduous – dominant are deciduous broadleaf species, representing 1,333 taxa (69%, Table 3). As for the species, there are 822 from this group, representing 74% of the overall number of the species. The proportion of cultivars is almost 56% of the total number of growing cultivars.

The Arboretum Mlyňany SAS is extraordinary important, owing to the collection of evergreen and semideciduous woody plant taxa. Table 4 illustrates the presence of taxa in collections and provides historical data about introduction of this group of woody plants. The records from the period 1894–1925 need not fully correspond to the state at that time as they seem to be

Table 3. Categories of trees and shrubs growing in the Arboretum Mlyňany SAS in 2012

Group	sp.	ssp.	var.	f.	cv.	Total	%
Conifers	131	4	14	7	171	327	16.9
Broad-leaved							
(Semi-) Evergreens	154	0	15	3	101	273	14.1
Deciduous trees and shrubs	822	7	151	8	345	1,333	69.0
Total	1,107	11	180	18	617	1,933	100.0

Table 4. Summary of broad-leaved evergreens and semievergreens introduction in the Arboretum Mlyňany

Period	1894–1925	1926–1952	1958	1953–1967	1968-1992	1993–2012
Families	48	22	28	39	38	26
Genera	104	44	53	90	85	56
Species	236	74	106	225	251	154
Taxa	248	123	126	291	408	273

only records made by the founder of the Arboretum, the count Ambrózy-Migazzi and the gardener Mr. Mišák, concerning the purchase of woody plants from nurseries in Europe and transport from nurseries in Bohemia by gardener Mišák.

Today, the plant collections in the Arboretum contain altogether 273 taxa of sempervirent and hiemivirent taxa of woody plants, which makes 14% of the total number of the taxa cultivated in the Arboretum. In year 1992, there were 408 taxa of this group representing more than 18% of the total. The decrease by 135 units compared with the year 1993 was probably due to the changes in the management of collections after 1993. The decrease in the number of cultivated genera was considerable – by 29, the decrease in the species number represented 97. The number of cultivars grown in this group of woody plants is very low compared to the number of coniferous and deciduous broadleaf cultivars – only little more than 16%, which is not favour-able for the collections in 2012.

The history of building the woody plant collections has recorded several trials with introduction of a wide range of evergreen and semi-deciduous woody plants. From the families and genera grown in the individual periods of the collections history but not recorded (present) in the next inventories, there are worth of noting (in bold):

a) in the period 1894–1925: **Bignoniaceae** (*Bignonia*), Caprifoliaceae (*Linnaea*), Compositae (*Cassinia*, *Olearia*), Ericaceae (*Arctostaphyllos*, *Epigaea*, *Leiophyllum*), Flacourtaceae (*Azara*, *Idesia*), Garryaceae (*Garrya*), Iteaceae (*Itea*), Labiatae (*Phlomis*), Leguminosae (*Ulex*), Philadelphaceae (*Carpenteria*), Polygalaceae (*Polygala*), Rosaceae (*Cercocarpus*), Rutaceae (*Choisya*), Trochodend-raceae (*Trochodendron*), Violaceae (*Hymenan-thera*)

- b) in the year 1958 (one species): Myricaceae (*Myrica cerifera*)
- c) in the period 1953–1967: Araliaceae (× Fatshedera), Casuariniaceae (Casuarina), Cistaceae (Helianthemum), Cornaceae (Corokia), Hamamelidaceae (Loropetalum), Labiatae (Teucrium), Lauraceae (Cinnamomum, Persea), Leguminosae (Ceratonia), Loranthaceae (Viscum), Myrtaceae (Acca, Eucalyptus, Myrtus), Oleaceae (Olea), Palmae (Chamaerops), Ranunculaceae (Clematis), Rosaceae (Rhaphiolepis), Schisandraceae (Schisandra), Scrophulariaceae (Penstemon, Phygelius)
- d) in the period 1968–1992: Berberidaceae (× Mahoberberis), Compositae (Santolina), Daphniphyllaceae (Daphniphyllum), Elaeagnaceae (Elaeagnus), Empetraceae (Empetrum), Ericaceae (Andromeda, Arbutus, Arcterica, Bruckenthalia, Calluna, Cassiope, Chamaedaphne, Daboecia, Gaulnettya, Kalmiopsis, Pernettya, Phyllodoce, Vaccinium), Escalloniaceae (Escallonia), Graminae (Shibataea), Hamamelidaceae (Distylium), Lauraceae (Danae), Myrsinaceae (Ardisia), Myrtaceae (Callistemon), Rhamnaceae (Rhamnus), Rosaceae (Dryas, Rubus), Schisandraceae (Kadsura), Theaceae (Camellia).

Table 5. Genera of broad-leaved evergreens and semi-evergreens successfully cultivated in the Arboretum Mlyňany up to the present time

	C		Number of tax	a in the period	
Family	Genus	1894–1925	1953–1967	1968–1992	1992-2012
Apocynaceae	Vinca	2	2	5	6
Aquifoliaceae	Ilex	10	10	40	23
Araliaceae	Hedera	2	2	28	6
Derhanderer	Berberis	13	7	36	21
Вегрепцасеае	Mahonia	11	3	6	6
D	Buxus	4	4	19	18
Buxaceae	Pachysandra	1	1	2	2
	Abelia	1	1	1	1
Caprifoliaceae	Lonicera	8	12	9	12
	Viburnum	5	8	9	9
Celastraceae	Euonymus	3	6	13	15
Cistaceae	Cistus	1	3	1	1
Cornaceae	Aucuba	1	3	3	3
Cruciferae	Iberis	1	1	5	1

	0		Number of tax	a in the period	
Family	Genus	1894–1925	1953–1967	1968–1992	1992-2012
Ericacea	Erica	8	12	1	1
	Kalmia	3	2	3	2
	Leucothoe	2	1	3	1
	Pieris	3	2	7	6
	Rhododendron	21	52	39	39
Fagaceae	Quercus	6	5	9	5
Hypericaceae	Hypericum	6	5	9	3
Labiatae	Lavandula	1	1	4	1
	Salvia	1	1	1	2
Liliaceae	Ruscus	2	2	3	2
	Үисса	6	1	4	2
Oleaceae	Ligustrum	6	8	7	6
	Osmanthus	2	2	2	3
	Phillyrea	2	4	3	2
Rosaceae	Cotoneaster	9	20	24	15
	Prunus	2	10	16	11
	Pyracantha	3	5	11	8
	Stranvaesia	2	3	4	3
Rutaceae	Skimmia	3	3	3	5
Thymelaeaceae	Daphne	5	4	2	1

Table 5. Genera of broad-leaved evergreens and semi-evergreens successfully cultivated in the Arboretum Mlyňany up to the present time – continued

On the other hand, over the whole history of the Arboretum, the introduction of sempervirent and hiemivirent species was successful with representatives of 19 families and 34 genera, mostly taxa of the genera *Ilex*, *Berberis*, *Buxus*, *Lonicera*, *Euonymus*, *Rhododendron*, *Cotoneaster* and *Prunus* (Table 5).

The potential of the development of the collections in the future is huge. The diversity of assortment grown in the leading arboretums is substantially higher. For comparison: in 2012, the Arboretum Mlyňany SAS, with its area of ca 67 ha comprised in summary 1,933 woody plant taxa (1,107 species), while several years ago, in the Arnold Arboretum of the Harvard University (USA) with an area of 132 ha there were 3,926 taxa (1,937 species) (ANONYMUS, 1999) and in an arboretum in Washington (Washington Park) with an area 81 ha even 4,605 taxa (MULLIGAN, 1977).

The high potential for introduction of new species in the Arboretum Mlyňany is also evident from the data according Krüssmann (KRÚSSMANN, 1976, 1977, 1978 and 1983) who provides summary of the high taxonomic diversity of the gene pool of woody plants suitable for introduction in conditions of the moderate climatic zone (Fig. 1).

In the number of genera of coniferous species, the Arboretum Mlyňany SAS currently manifests only one

half of their introduction potential. There are possible to introduce more than 470 additional species, the current proportion of conifers in the Arboretum is a bit more than 21%. Even more possibilities are for introduction of broadleaf woody plants with 590 additional possible genera with more than 4,500 species. The current state in the Arboretum is only cca 18% of potentially suitable broadleaf woody plants. This potential introduction does not include varieties of the two groups – coniferous and broadleaf woody plants (evergreen, semi-deciduous, deciduous).

Clearly, it is also necessary to keep in mind that individual species and lower-than-species level taxa in the individual genera differ in their acclimation capacity. This means that the actual numbers of the species and of infra-specific taxa suitable for the Arboretum Mlyňany SAS may be somewhat lower. Nevertheless, the potential of future introduction maintains huge.

Acknowledgements

This publication was supported by the Slovak Grant Agency VEGA, Project No. 2/0085/09 Climate changes and prospects of introduced taxa of East-Asian den-



Fig. 1. Comparison between the recent numbers of genera and species of coniferous and broad-leaf trees and shrubs cultivated in the Arboretum Mlyňany SAS and the potential for introducing new plants according to KRÜSSMANN (1976, 1977, 1978 and 1983).

droflora in Mlyňany Arboretum SAS and Project VEGA No. 2/0159/11 Adaptability of selected evergreen woody plants and their possible uses in garden and landscape architecture.

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Received December 6, 2012 Accepted April 19, 2013

Coniochaeta prunicola – causal factor involved in health state decline of selected trees of the genus Prunus

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Abstract

IVANOVÁ, H. 2013. *Coniochaeta prunicola* – casusal factor involved in health state decline of selected trees of the genus *Prunus*. *Folia oecol.*, 40: 188–191.

The record of *Coniochaeta prunicola* Damm & Crous (Coniochaetales, Sordariomycetes, Ascomycota) as a pathogen of host trees was described and illustrated from Nitra. This pathogen was isolated from symptomatic twigs and leaves of *Prunus laurocerasus* L. as well as from symptomatic leaves of *Prunus persica* Mill. and based on morphological attributes identified as a causative agent of this trees damage. *C. prunicola* is characterized by dark brown ascomata clothed with setae, the fasciculate, unitunicate, cylindrical asci and broadly almond-shaped, ellipsoidal ascospores with a longitudinal germ slit.

Keywords

Ascomycota, Coniochaeta prunicola, morphological attributes, Prunus laurocerasus, Prunus persica, Sordariomycetes

Introduction

Prunus laurocerasus L. (syn. *Laurocerasus officinalis* L.), evergreen shrub or a small tree in Rosaceae family, was frequently planted as an ornamental plant in temperate regions worldwide. It is often used as a mass landscape and ground cover plant in urban green areas.

Prunus persica L. (Batsch.) (syn. *Persica vulgaris* Mill.) is a deciduous tree, native to China, where it was first cultivated. It bears an edible juicy fruit called a peach. The species name of persica refers to its widespread cultivation in Persia, whence it was transplanted to Europe. It belongs to the family of Rosaceae.

These shrubs and trees are susceptible to various pathogens, which caused discoloration, brown spots, blight symptoms and necroses, affecting their aesthetic value. The symptoms of infection, which are observable from spring to autumn, increase when the plants are in bloom, resulting in dieback and leaf drop. The damage is caused by fungus *Coniochaeta prunicola* Damm & Crous.

The genus *Coniochaeta* (anamorph: *Lecythophora*) includes ascomycetous fungi known as pathogens of woody plants, but some species can also cause human infections. *Coniochaeta* contains more than 80 species occurring mostly on wood and bark, leaves and leaf litter of different trees, in dung of various animals, and in soil and water. Species of the genus *Coniochaeta* and their *Lecythophora* anamorphs occur on different plant material (on wood or bark of different trees, on leaves and leaf litter), in dung of various animals, in soil and in water with extremely low pH and high concentrations of heavy metals (ERIKSSON, 1992; KAMYIA et al., 1995; LÓPEZ-ARCHILLA et al., 2006; ASGARI et al., 2007). Some *Coniochaeta* species have been found to exhibit useful biochemical properties. Species of *Coniochaeta* have been isolated from different plant parts of the representative genus *Prunus*.

During an investigation on mycoflora of cherry laurel trees and peach trees growing in urbanized area, besides the fungi of the classes Hyphomycetes and Coelomycetes isolated from affected cherry laurels (BERNADOVIČOVÁ and IVANOVÁ, 2011), the ascomycetous fungus *Coniochaeta prunicola* (Coniochaetaceae, Coniochaetales) that affects leaves and twigs of the host trees was noticed. Although the incidence of disease was sporadic, the infected trees showed relatively severe damage.

This study aims for identification based on morphological attributes which the microscopic fungus isolated from symptomatic cherry laurel and peach trees in connection with the new disease noticed recently, and to describe the distinctive morphological features for the isolated *Coniochaeta* species as a causal factor involved in health state decline and vitality weakening of *Prunus laurocerasus* and *Prunus persica*.

Material and methods

From spring to autumn 2009–2011, leaves and twigs of *Prunus laurocerasus* and leaves of *Prunus persica* (Redhaven) with blight symptoms were sampled from plants growing in private gardens and public greenery of the town of Nitra. The material was collected at several locations from the diseased *Prunus* trees in the areas of Nitra - Chrenová and Nitra - Zobor. Altogether 25 trees were studied (17 trees of *Prunus laurocerasus*, 8 trees of *Prunus persica*). The age of evaluated trees was between 15–35 years. The samples of biological material were deposed in herbarium at the Institute of Forest Ecology of the Slovak Academy of Sciences, Branch for Woody Plant Biology in Nitra.

Classical phytopathological methods - cultivation on nutritive medium in test chamber with constant temperature and humidity were used to isolate and obtain pure cultures. The leaf and twig parts cut from the diseased plants were surface-sterilized by immersion in sodium hypochlorite solution (1% available chlorine) for 20 minutes, rinsed twice or three times in sterile distilled water and then dried carefully with filter paper. After that, the plant samples were cut to fragments of 3-5 mm which were placed on 3% potato-dextrose agar (PDA) in Petri dishes. This was followed by cultivation at $24 \pm$ 1 °C and 45% humidity in dark conditions in a versatile environmental test chamber MLR-351H (Sanyo) and subsequent isolation on the 3% PDA medium. Pure fungal cultures were obtained by using multiple purifications. The obtained isolates were transferred on 3% PDA medium to induce sporulation. Study of fungal structures was performed with a clinical microscope BX41 (Olympus) under a $400 \times$ and $1,000 \times$ magnification.

The isolated fungus was identified by microscopic analyses based on morphological characteristics of the fruiting bodies (perithecia), spore bearing organs (asci) and reproduction organs (conidia and ascospores). The identification was performed using morphological keys assembled by HAWKSWORTH and YIP (1981), ELLIS and ELLIS (1987), CHECA et al. (1988), ROMERO et al. (1999), ASGARI et al. (2007) and morphological studies in MA-HONEY and LAFAVRE (1981), HANLIN (1990), WEBER (2002) and DAMM et al. (2010).

Results and discussion

Concerning all morphological characteristics and determined differences, the fungus under investigation in our study isolated from branches showing necrosis symptoms and blighted leaves of cherry laurel trees and from blighted leaves of peach trees was identified as *Coniochaeta prunicola*.

Anatomical-morphologically characteristics of fungus *Coniochaeta prunicola* Damm & Crous on *P. laurocerasus* and *P. persica* are in Table 1.

Review of the literature shows that although the characteristics of asci and ascospores are very important, setae are still the prominent feature of the most Coniochaeta species. Most of the described setae are dark brown to black rigid hairs, straight or bent, unbranched with a sharp apex. They may be scattered over the perithecial wall or concentrated in its upper portion (MAHONEY and LAFAVRE, 1981). Some species are described as lacking setae (ROMERO et al., 1999). According to DAMM et al. (2010) fungus C. prunicola isolated from branches of stone fruit (Prunus sp.) produced subglobose to pyriform ascomata, 200-250 µm in diameter, neck 50-60 µm long. Peridium was pseudoparenchymatous, 20-25 µm (5-8 layers), outer wall consists of dark brown textura angularis, with setae. Setae were brown (or hyaline), straight, cylindrical, tapering to a round tip, smooth-walled or granulate, 2-3.5 µm wide, up to 80 µm long. Results of our study are in Table 1.

The key provided in ASGARI et al. (2007) leads our results to Coniochaeta velutina, except that the ascospores of this species have guttules, and isolates of Coniochaeta prunicola produce larger ascospores compared to Coniochaeta velutina. These ascospore features correspond to those provided by MUNK (1957), where isolates from Prunus sp. produced ascospores $6-8 \times 4-6 \times 3-4 \ \mu m \text{ or } 9-10.5 \ (12.5) \times 5 \ (7.5) \ \mu m$ in size (IVANOVÁ, not published yet) and by description in DAMM et al. (2010) and another authors. The other species (Coniochaetidium sp., Ephemeroascus sp. and Poroconiochaeta sp.) transferred into Coniochaeta by GARCÍA et al. (2006) differed from Coniochaeta prunicola by displaying ornamental ascospore walls, or by lacking Lecythophora anamorphs. Most of the Coniochaeta species exhibit different ascospore sizes: Coniochaeta leucoplaca (Berk. & Ravenel) 7-10 × $5-9 \times 4-8 \ \mu m$ and Coniolariella ershadii (Zare, Asgari & W. Gams) Zare, Asgari & W. Gams (basionym Coniochaeta ershadii Zare, Asgari & W. Gams) 16 × 18×9.5 –10 µm isolated from twigs of *Pistacia vera* L. (ASGARI et al., 2007; ZARE et al., 2010), Coniolariella gamsii (Asgari & Zare) Dania García, Stchigel & Guarro (basionym *Coniochaeta gamsii* Asgari & Zare) $16-19 \times 6-11 \ \mu m$ isolated from leaves of Hordeum vulgare L. (ZARE et al., 2010; ASGARI and ZARE, 2006), Coniochaeta ligniaria (Grev.) Massee $9-20 \times 8-15 \times$ 4-8 µm (MAHONEY and LAFAVRE, 1981), Coniochaeta rhapalochaeta sp. nov. (Romero & Carmarán) 10-14 × $7.5-9 \times 5-6 \,\mu\text{m}$ isolated from wood of *Bulnesia retama* (Gillies ex Hook. & Arn.) Griseb. (ROMERO et al., 1999), Coniochaeta prunicola Damm & Crous 9-10.5 (12.5)

Host plant	Prunus persica	Prunus laurocerasus
Plant part	Leaves	Twigs, leaves
Causal agent	C. prunicola	C. prunicola
Ascomata	Perithecial, solitary, subglobose to pyriform, $125-173 (265) \times 95-145 (229) \mu m$, with a central ostiole, neck $31-42 \mu m$	Perithecial, solitary, 162–221 \times 119–159 $\mu m,$ subglobose to pyriform, with a central ostiole, neck 38–42 μm
Setae	Hyaline or brown setae, smooth walled, 3–4.5 \times 21–29 μm	Hyaline or brown setae, smooth walled, 3–4.5 \times 35–51 μm
Paraphyses	Hyaline, septate, $63 \times 3-4 \ \mu m$	Hyaline, septate, $7478\times34~\mu m$
Asci	Fasciculate, unitunicate, cylindrical with truncate apex, obtuse end, small apical ring 4–5 μm long, 8 ascospores/ ascus, 58–68 (94) \times 8–10 μm	Cylindrical, unitunicate with obtuse end, with a small apical rings 4–5 $\mu m,$ 8 ascospores/ ascus, 68–81 \times 8–10 μm
Ascospores	Uniseriate, 1-celled, ellipsoidal, smooth-walled without ornamentation wall, green to brown with granular contents, 9 (10)–12 × 5 (6) μ m, longitudinal germ slit 5 × 8 μ m	Uniseriate, 1-celled, ellipsoidal to almond shaped, brown, smooth-walled with granular content, $9(10-)13 \times (5-)6-7(-8) \mu m$, without ornamentation of the ascospore wall, longitudinal germ slit $7 \times 6 \mu m$
Guttules	Absent	Absent
Hyphae	_	Hyaline, 2–3 µm wide
Conidia	Hyaline, 1-celled, smooth walled, cylindrical to ovoid, (2–)3–6(–7) \times 1–2 μm	Hyaline, 1-celled, smooth walled, cylindrical to ovoid, sometimes allantoid $(2-)3-4(-7) \times 1-2$ µm formed on hyphal coil
Conidiophores	Directly on hyphae	Directly on hyphae
Colarette	Distinct, cylindrical, 2-3 µm long	Inconspicuous
Colonies on PDA	Pale saffron, pale buff to white, flat, with sparse aerial mycelium	Pale buff to white, flat, with sparse aerial my- celium
Chlamydosp.	Lacking	Lacking

Table 1. Comparison of morphological characteristics of Coniochaeta prunicola Damm & Crous identified in genus Prunus

 \times 5 (7.5) μm isolated from leaves of *Prunus domestica* (IVANOVÁ, not published yet).

Causal organism was systematically isolated from leaf and twig tissue showing rusty to brown coloured blight symptoms. Growth on PDA was slow. Colonies appeared white at first, than turned on pale buff to white or pale saffron. Conidia were produced in great numbers in culture media. Perithecia developed on PDA after about 4–5 (*P. laurocerasus*) or 8–10 (*P. persica*) weeks. Cultures of *Coniochaeta prunicola* do not turn dark as *Coniochaeta velutina* cultures (WEBER, 2002; DAMM et al., 2010). This fact was also confirmed in our study with isolates of fungus *C. prunicola* from peach trees (IVANOVÁ and BERNADOVIČOVÁ, 2012) and cherry laurel shrubs (IVANOVÁ and BERNADOVIČOVÁ, 2013), (Table 1).

In anamorph stage of *Coniochaeta velutina* described from various tree and shrub hosts in *Lecythophora* genus, sizes of conidia obtained from pure cultures varied: $3-6 \times 2-4 \mu m$ (TAYLOR, 1970), 2.5–3.5 $\times 1.5-2 \mu m$ (UDAGAWA and HORIE, 1982), $2-4 \times 1-2.5 \mu m$ (HUTCHINSON and REID, 1988), and $3-8 \mu m$ long (KIRSCHNER, 1998). According to DAMM et al. (2010), the anamorph of *Coniochaeta prunicola* is also similar

to that of *Coniochaeta velutina*, but the collarettes in the latter are shorter, up to 1 μ m in length, and the conidia are wider and not regularly allantoid. This fact has also been confirmed in our study (Table 1).

The fungus *Coniochaeta prunicola* was found in the examined samples relatively uncommonly. Our studies and morphological identification have shown that *Coniochaeta prunicola* was a new pathogenic fungus associated with affected branches and leaves of *P. persica* and *P. laurocerasus* in Slovakia. This preliminary identification, however, needs using methods of molecular biology for confirmation, since the morphological characteristics alone may not be fully reliable for this purpose. Further studies are required for determination of pathogenicity and relevance of *Coniochaeta* infection in connection with peach trees and cherry laurel damage. The planned molecular analysis based on large subunit nuclear ribosomal DNA sequences is required for detailed study of the discussed pathogens.

Acknowledgement

This study was conducted thanks to financial support of the project No. 2/0149/10 of scientific grant agency of

the Ministry of Education of the Slovak Republic and Slovak Academy of Sciences VEGA.

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Received December 6, 2012 Accepted March 27, 2013

Propagation of two selected species of the genus Pieris D. Don.

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Abstract

KONÔPKOVÁ, J., BIBEŇ, T. 2013. Propagation of two selected species of the genus *Pieris* D. Don. *Folia oecol.*, 40: 192–200.

This work evaluates the results of propagation experiments of Mountain Pieris (*Pieris floribunda* /Pursh/ Benth. & Hook.) and Japanese Pieris (*Pieris japonica* /Thunb./ D. Don) we carried out in the Arboretum Mlyňany SAS. The material was sampled from the two exemplars of these species growing in the Arboretum. The methods used were auto-vegetative propagation by cuttings and *in vitro* micropropagation. The response of the studied woody plant species varied according to the species and the method used. In Japanese Pieris, better results were achieved by vegetative propagation by cuttings; in Mountain Pieris, much more effective propagation method was micropropagation. We also studied the effect of climatic variables on the physiological conditions of the parent plants, and the overall rooting success in primary cultures obtained by micropropagation of Mountain Pieris. The data were recorded on each sampling event in the growing seasons 2011 and 2012. The process of micropropagation in Mountain Pieris was evaluated based on the production characteristics of the regenerants after the 3rd sub-cultivation. The results confirmed statistically significant differences in the number of shoots/explants and in the concentration of chlorophyll *a* between the dates of the primary culture establishment. The maximum number of shoots/explants (10.9) was obtained in variant B (primary culture established on 07/21/2012) and the highest concentration of chlorophyll *a* 6.66 mg g⁻¹ on dry matter was found in variant C (primary culture established on 08/24/2011).

Key words

climatic conditions, chlorophylls, micropropagation, propagation by cuttings

Introduction

Pieris species are evergreen shrubs or small trees belonging to the family Ericaceae. Their leaves are alternate, often gathered at the ends of branches, elongated, lanceolate, 2-8 cm long, matt or glossy. Some cultivars in the group variegata have yellow or white variegated leaves; young leaves of some improved cultivars are bronze to flame-red coloured. The pieris species are characterised with attractive flowers, some cultivars display spectacular foliage colours in spring. The flowers are 5s, with white, rosy to light-red bell-shaped crowns, 4-7 mm long, with 10 stamens. The flowers are arranged in terminal panicles long 5-18 cm. Upright inflorescences consisting of green-white to reddish buds are formed already in October. They maintain their bright colouring over the whole winter until bursting in flower in March or April. The fruits are inconspicuous five-valve spherical capsules. There are about ten pieris species growing in the North America, East Asia and the Himalayas (HORÁČEK, 2007).

The climate conditions in Europe, however, are favourable for only four pieris species and several hundreds of cultivars have been improved in nurseries of decorative woody plants, mostly in England and in Germany. The first trial with pieris introduction in Slovakia was made in 1899, in the Arboretum Mlyňany, by the founder of the Arboretum Dr. Štefan Ambrózy-Migazzi and his gardener Jozef Mišák. In 1899, there were imported several exemplars of the Mountain Pieris (Pieris floribunda) from the plant nursery of Peter Smith in Germany, and several individuals of Japanese Pieris (Pieris japonica) were planted in the original evergreen Semper vireo park eight years later in 1907. Today, the living woody plant collections in the Arboretum Mlyňany SAS contain: Pieris floribunda /Pursh/ Benth. & Hook., Pieris japonica /Thunb./ D. Don, P. japonica cv. Debutante, P. japonica cv. Purity, Pieris polita W.
W. Sm. & Jeffey and *Pieris taiwanensis* Hayata (HoŤKA and BARTA, 2012).

Pieris species require soils, climate and management types similar to most of heath land plants. They need protected sites with full sun; the original species, however, thrive also in partial shade. The necessary conditions for annual flowering are sufficient air humidity and soil moisture content. The representatives of these species respond sensitively to mineral fertilizers and to pruning. In their native area, some species as *Pieris japonica* reach into high altitudes with winter temperature below -20 °C. In our climatic conditions, these species grow relatively well in protected sites, but they cannot resist black frosts in higher situated ones. The critical period is generally the end of February and the beginning of March when the buds in panicles may be damaged by spring frosts.

Pieris are propagated mainly by seeds, some cultivars, however, are propagated in auto-vegetative way, with summer cuttings separated from semi-wood twoyear-old shoots (KAMENICKÁ et al., 2004). Today, the propagation also uses *in vitro* methods (STARETT et al., 1993). Plant biotechnologies are rapid and effective tools for propagation of a number of decorative and forest woody plants, and as such, they are focused appropriately in Slovakia (KAMENICKÁ and VÁĽKA, 1997; KAMENICKÁ et al., 2005; GAJDOŠOVÁ et al., 2007; Os-TROLUCKÁ et al., 2007; ĎURKOVIČ, 2008).

This work compares the propagation of the Mountain Pieris and the Japanese Pieris by several propagation methods.

Material and methods

The plant material for the experiments was sampled from a 44-year-old exemplar of Mountain Pieris (*Pieris floribunda* /Pursh/Benth. & Hook.) (Fig. 1) and a 70-year-old exemplar of Japanese Pieris (*Pieris japonica* /Thunb./ D. Don) (Fig. 2), growing in the original evergreen *Semper vireo* park in the Arboretum Mlyňany SAS, both in almost identical site conditions (GPS coordinates – Mountain Pieris 48°19'11.2" N, 18°22'13.9" E; Japanese Pieris 48°19'11.7" N, 18°22'13.0" E).



Fig. 1. Flowering Mountain Pieris (*Pieris floribunda* /Pursh/Benth. & Hook.)



Fig. 2. Flowering Japanese Pieris (*Pieris japonica* /Thunb./ D. Don).

Mountain Pieris – native in moist forest hill slopes in North America. It is a high resistant, slow growing, round-shaped shrub, with an ultimate height of 1.2–2 m. The flowers appear in March to April (casually in May) clustered in terminal panicles by five. The greenishwhite buds are attractive over the whole winter.

Japanese Pieris – is a native species in light open forests in mountains of the Japanese islands Shikoku, Kyushu and Honshu. It is a medium-sized shrub with attractive glossy foliage and white waxy flowers arranged in straight panicles. The flowering period is March–April. There exists a large number of cultivars of this species, such as 'Bisbee Dwarf' (syn.: P.j. 'Bisbee'), 'Blush', 'Brouwer's Beauty', 'Chaconne', 'Compacta', 'Debutante', 'Dorothy Wyckhoff', 'Fuga', 'Little Heath', 'Nocturne', 'Prelude', 'Purity', 'Pygmaea'(syn.: P.j. 'Nana Compacta'), 'Sarabande', 'Snowdrift', 'Toccata', 'Valley Rose', 'Variegata', 'White Cascade' (HORÁČEK, 2007).

From the two parent plants, Mountain Pieris and Japanese Pieris, there was sampled material for propagation, at regular monthly intervals over the growing seasons 2011 and 2012. The material was of two types: cuttings used for auto-vegetative propagation and explants from axillary vegetative buds for in vitro propagation. In this work are evaluated 4 experimental variants described in Table 1. The table also contains climatic data, namely mean daily temperatures and precipitation sum from the beginning of the growing season to the first sampling date (variants A, D) and between the samplings (variants B, C). These values were measured at the Meteorological Station of the Arboretum Mlyňany SAS. In individual experimental variants, there was also sampled plant material for determining concentrations of chlorophylls (chlorophyll a, chlorophyll *b*, chlorophyll a + b, ratio a/b).

Table 1. Description of experimental variants

Variant	Sampling	Mean daily	Precipitation
	date	temperature [°C]	sum
			[mm]
А	June 21, 2011	16.02	110.0
В	July 21, 2011	19.71	117.4
С	August 24, 2011	19.41	74.2
D	June 28, 2012	15.92	101.8

Auto-vegetative propagation by cuttings – in each experimental variant (A, B, C, D) were cut terminal cuttings (50 ps in each variant) from the donor plants. The cuttings were reduced to a length of 5 cm, treated with 4 different growth stimulants (R1 – Rhizopon A – 0.5% 3-indolyl acetic acid, R2 – Rhizopon A – 1% 3-indonyl acetic acid, R3 – Rhizopon AA – 0.5% 3-indonyl butyric acid, and R4 – Rhizopon AA – 0.5% 3-indonyl butyric acid) and rooted in substrate KLAS-MANN TS 5 in propagation rooms of the greenhouse. The terminal cuttings were covered with a plastic sheet to ensure sufficient moisture content. Simultaneously with these cuttings were also rooted control cuttings untreated with stimulants – control (CN).

The rooting success (%) in the individual experimental variants was evaluated after 60 days of cultivation.

Propagation by the method in vitro - plant explants sampled from axillary vegetative buds of the donor plants, Mountain Pieris and Japanese Pieris, by 20 ps in each variant, were washed in water, cut into shorter segments and sterilised by 5 min immersion in a 0.3% light agar solution supplemented with 25 ml l⁻¹ PPM (PPMTM, Plant Cell Technology, Inc., Washington, DC USA) and then by immersion for $1-2 \min in a 0.1\%$ solution of mercury chloride - to prevent exogenous contamination. After a thorough rinsing (3 times with redistilled sterile water), the shoots were shortened to 1-2 cm long explants and transported in sterile conditions onto a modified WPM cultivation medium (LLOYD and McCown, 1980; STARETT et al., 1993) enriched with cytokinin N⁶-[2-Isopentenyl]adenine (2iP) at a concentration of 8 mg l⁻¹, pH values of cultivation media were adjusted to 5.2 either with 1M KOH or with 1M HCl, and supplemented with 20 g l^{-1} sucrose, 8 g l^{-1} agar, poured in cultivation dishes and sterilised in an autoclave for 20 min at a temperature of 121 °C and a pressure of 120 kPa.

The explants were cultivated in controlled conditions, for a 16-hour cultivation period, at temperatures of 24 °C ± 1 °C during day and 20 °C ± 1 °C at night, and illumination intensity 40–50 µmol s⁻¹ m⁻². After 10 weeks of cultivation, there was evaluated the number of vital primary explants and the differentiated shoots were used repeatedly for sub-cultivation. The micropropagation process in the experimental variants A, B, C was evaluated after the 3rd sub-cultivation, on the background of production characteristics of the regenerating material (number and length of shoots, biomass production). At the same time, chlorophyll concentrations were determined in the regenerating segments (chlorophyll *a*, chlorophyll *b*, chlorophyll *a* + *b*, ratio *a/b*).

Determining of chlorophyll concentrations

The chlorophyll concentrations were determined by spectrophotometry, as proposed by LICHTENTHALER (1987). Chlorophylls in plants occur in form of chlorophyll-protein complexes, consequently, their extraction from plant material requires using non-polar solvents (ethanol, acetone, benzene). The extraction from plant material discussed in this work was carried out with 80% solution of acetone. The pigment extracts were prepared from material taken from aboveground parts of the tissue cultures. From each culture, there were taken 10 discs, each with a diameter of 5 mm. The discs were homogenised in a grinding mortar with a small amount of quartz sand, waterless magnesium carbonate and 3 ml of 80% acetone. The obtained homogeneous substance was filtered through a glass porous filter (S_2) . From the filtered substance, there were sampled 20 ml amounts (DYKYJOVÁ et al., 1989) whose absorbance was measured in a spectrophotometer V-600 (Jasco, Japan), at wave lengths $\lambda_{a} = 663.2$ nm and $\lambda_{b} = 646.8$ nm corresponding to the absorption maxima of chlorophyll a and chlorophyll b. The measured absorbance values were substituted in the equations proposed by LICHTENTAHLER (1987) for calculating concentrations of photosynthetic pigments of chlorophylls a, b, total chlorophylls a + band chlorophyll ratio a/b. Finally the results were converted to dry-mass corresponding values.

The biomass production was assessed based on dry biomass, gravimetrically, after drying out the specimens to the constant weight at 105 $^{\circ}$ C.

Results and discussion

The auto-vegetative propagation of Japanese Pieris by cuttings treated with various growth stimulants exhibited the best mean establishment rate (53.5%) in variant B. The lowest percent of rooted shoots (14.5%) was obtained in variant A (sampling date 21 June 2011). Over the study period, there were not recorded any strong fluctuations in the mean daily temperature, with the lowest values at the beginning of the growing season. As for the precipitation sum, the most distinct drop was found in variant C (Table 1). In variant B (21st of July 2011 sampling date), when the rooting of cuttings was the best, both temperature and precipitation reached the highest values what suggests good physiological stage of the parent plant from which the cuttings have been taken for rooting. This finding is in agreement with the results of WALTER (2011) which recommends for the propagation of genus Pieris D. Don mature terminal shoots cut from July to September.

Among the growth stimulants, 1% 3-indolyl acetic acid (R2) was the most effective – with 40.4% average rooting rate. With 1% 3-indolyl butyric acid (R4), the average rooting success was by 5% lower. The rooting

success values obtained with using growth stimulants in lower concentrations (R1, R3) were changed only a little (R1 = 29.7%, R3 = 28.9%) compared to R2 and R4 (Fig. 3). Application of auxins affected considerably not only the root quality but also the speed of root system development. In case of shoots without growth stimulants, the first roots appeared with a two-week delay and in lower abundance.

According to SPETHMANN (1990) the success in propagation by cuttings is determined decisively by the age and fitness of the plant and, consequently, by the physiological viability of the cuttings. Some woody plants better propagate with green - non-lignified (summer, soft cuttings), the other exhibit more success with winter (hard) cuttings. Physiological fitness of parent plants is also considerably affected by the stand microclimate. From this point of view, the sampling date is important factor affecting root development in shoots. Therefore, we also investigated the influence of selected microclimatic variables (air temperature, precipitation) on assimilatory pigments concentrations in parent woody plants. We found the concentration of chlorophyll a in the parent plant of Japanese Pieris in the individual experimental variants in the range 3.26-5.82 mg g^{-1} (Table 2). The highest concentrations occurred in variant B during intensive biomass creation, the lowest in variant C – probably due to the precipitation deficit in this period. Similar trends were found for the rate of chlorophyll a to chlorophyll b, a/b, with the values ranging from 2.24 to 2.94. Analysis of variance (Table 3) and consecutive Duncan test confirmed statistically

significant differences in assimilatory pigments concentrations in all experimental variants (Table 2). Strong influence of precipitation sum on chlorophyll a concentration was also confirmed by linear regression analysis (Fig. 4) with a high correlation coefficient (r = 0.9148). For temperature, no similar influence was detected. The effects of environmental factors on assimilatory pigments contents were studied by DEMMING-ADAMS et al. (1996), SEIFERMANN-HARMS (1994) and KIRCHGESSNER et al. (2003). The last author suggests the following rank of climatic factors affecting pigments: air temperature, solar radiation, global radiation, atmospheric precipitation. Exploration of the dependence of rooting success of Japanese Pieris cuttings on climatic conditions and on chlorophyll content in parent woody plant (Table 4) by correlation analysis resulted in a conclusive relation for the precipitation sum under using stimulant R2 (r = 0.5204) and for control (r = 0.6455). The correlation between the shoot rooting success and mean daily temperature during sampling was only weak. Significant to highly significant was dependence of rooting success on concentration of chlorophyll a, total concentration of chlorophylls *a* and *b* and concentration ratio a/b with using growth stimulants R1, R2, R3, R4. The closest dependence was found in the control variant (r values: chlorophyll a 0.8987; chlorophyll a + b 0.8660; chlorophyll *a/b* 0.9965) (Table 4). Considerable differences in rooting success of cuttings of Japanese Pieris among individual variants of experiments were also due to insufficiently stable temperature and moisture conditions in the propagation compartment in the greenhouse in the Arboretum.



Experimental variant

Fig. 3. Rooting success in cuttings of Japanese Pieris in individual experimental variants.

Donor plants	Experimental variant	Chlorophyll <i>a</i> $[mg g^{-1}] \pm SE^1$	Chlorophyll b [mg g ⁻¹] ± SE ¹	Chlorophyll $a + b$ [mg g ⁻¹] ± SE ¹	Chlorophyll a/b [mg g ⁻¹] ± SE ¹
	А	$4.56\ \pm\ 0.372\ b$	$1.94 \pm 0.367 a$	$6.50 \pm 0.722 \text{ b}$	$2.37 ~\pm~ 0.256 ~ab$
Dionia inconica	В	$5.82 \pm 0.317 \text{ c}$	$2.03 \pm 0.287 \text{ a}$	$7.85 \pm 0.221 \text{ c}$	$2.94 \pm 0.579 c$
Pieris japonica	С	$3.26 \pm 0.287 \text{ a}$	$1.52 \pm 0.399 \text{ a}$	$4.78 \pm 0.675 a$	$2.24 \pm 0.433 \text{ a}$
	D	$5.09 \pm 0.845 c$	$1.84 \pm 0.393 \text{ a}$	$6.93 \pm 1.227 \text{ bc}$	$2.79 \pm 0.191 \text{ bc}$
	А	$4.06 ~\pm~ 0.829 ~b$	$1.76 \pm 0.619 \text{b}$	$5.82 \pm 1.404 \text{ b}$	$2.41\ \pm\ 0.431\ b$
Pieris floribunda	В	$5.27 \pm 0.707 c$	$1.60 \pm 0.328 \text{ ab}$	$6.86 \pm 1.023 \text{ bc}$	$3.34 \pm 0.292 \text{ c}$
	С	$1.71 \pm 0.315 a$	$1.05 \pm 0.389 \text{ a}$	$2.75 \pm 0.642 \text{ a}$	$1.77 \pm 0.486 \text{ a}$
	D	$5.47 \pm 0.320 \text{ c}$	$2.13 \pm 0.184 \text{ b}$	$7.60 \pm 0.466 c$	$2.58 \pm 0.159 \text{ b}$

Table 2. Chlorophylls concentrations in parent plants of Japanese Pieris and Mountain Pieris in individual experimental variants

SE¹, standard error of arithmetic mean.

Differences among values labelled with the same symbols (a)-(d) are not statistically significant at 95% significance level (Duncan test).

Table 3. Analysis of variance (ANOVA) of chlorophylls concentrations in parent plants of Japanese Pieris and Mountain Pieris in individual experimental variants

					F-1	test			
Course	Pieris japonica						Pieris floribunda		
of variance	of freedom	Chlor. <i>a</i>	Chlor. b	Chlor. a+b	Chlor. a/b	Chlor. a	Chlor. b	Chlor. a+b	Chlor. a/b
Among treatments	3	22.55*	1.87	13.09*	3.55*	39.33*	5.21*	22.47*	14.95*
Residual (within treatments)	16								
Total	19								

*statistically significant differences at 95% level (P < 0.05).

Table 4. Correlation between rooting success in cuttings of Japanese Pieris, vitality of primary explants in Mountain Pieris, climatic conditions and chlorophylls concentrations in parent plants

Woody plant	Symbol	Precipitation sum	Mean temperature	Chlorophyll a	Chlorophyll b	Chlorophyll $a+b$	Chlorophyll <i>a/b</i>
	R1	0.2811	0.3147	0.6236*	0.2945	0.5744*	0.8371*
	R2	0.5204*	0.3155	0.8119*	0.5234*	0.7719*	0.9581**
Pieris	R3	0.2250	0.2779	0.7631*	0.5629*	0.7378*	0.8416*
јаропіса	R4	-0.0238	-0.0150	0.5315*	0.1486	0.4718*	0.7995*
	CN	0.6455*	0.0592	0.8987**	0.6457*	0.8660**	0.9965**
Pieris floribunda	In vitro	0.5955*	-0.9106**	0.7261*	0.9535*	0.7910*	0.3083

|r| < 0.40 - poor correlation (very weak relation); 0.40 < |r| < 0.85 - good correlation (significant)*; 0.85 < |r| < 1.0 - strong correlation (highly significant)**.

The successful micropropagation of Mountain Pieris (Fig. 6), contrarily to the Japanese Pieris, was consistent with the former knowledge according to which the morhopgenetic response of explants can exhibit considerable differences, even intraspecific or among cultivars of the same species (SALAJ and BLEHOVÁ, 2006). Evaluation of establishment success in primary cultures revealed (Fig. 5) that the highest percentage of vital primary explants (80% and 85%) was obtained in case of juvenile shoots (variants A, D). Similarly as in Japanese Pieris, assimilatory pigments were also investigated in Mountain Pieris. These pigments showed statistically significant differences among the variants (Table 2). The highest concentration of chlorophyll



Fig. 4. Linear regression analysis of precipitation sum's effect on chlorophyll *a* concentrations.

a (5.47 mg g⁻¹) was recorded in variant D, the lowest one (1.71 mg g⁻¹) in variant C. The Mountain Pieris exhibited more pronounced differences in the values of *a/b* ratio, ranging from 1.77 to 3.34. Linear regression analysis (Fig. 4) confirmed a strong correlation between the precipitation sum and chlorophyll *a* concentration in parent plants (r = 0.8577). The correlation analysis also confirmed that the percentage of vital shoots of Mountain Pieris was significantly negatively correlated (r = -0.9106) with the mean daily air temperature in the individual experimental variants (Table 4). Evidential correlation in Mountain Pieris and chlorophyll *a* concentration

tration (r = 0.7261) and between this culture initiation and chlorophyll a/b ratio (r = 0.7910). In case of chlorophyll *b*, the correlation was highly significant – r = 0.9535 (Table 4).

The same results were obtained by ŠEDIVÁ (1998) working with propagation of evergreen plants in the family Ericaceae who succeeded in establishing primary cultures for 6 from 14 taxons. The most serious problems seemed to be connected with Japanese Pieris and its cultivars. For the Mountain Pieris, Šedivá obtained the best results on WPM cultivation medium supplemented with 5 mg l^{-1} 2iP.



Fig. 5. Vitality of primary explants of Mountain Pieris in individual experimental variants.

The micropropagation process of Mountain Pieris was evaluated based on the production characteristics in the regenerants (variants A, B, C) after the 3^{rd} sub-cultivation. The results of variance analysis showed statistically significant differences in the number of vital shoots, dry biomass amount (Table 5) and concentrations of chlorophylls *a*, *a* + *b*, *a/b* (Table 7) dependent on the date of establishment of the primary culture (Fig. 6a). The maximum number of 10.9 shoots/explants with ultimate length of 19.88 mm was obtained in variant B (primary culture established on July 21, 2011) (Fig. 6b). In this variant was also ob-

tained the highest value of dry biomass 0.0632 g (Table 6). The highest concentration of chlorophyll *a* making 6.66 mg g⁻¹ in dry mass (Table 8) was found in variant C (primary culture established on August 24, 2011), which was also reflected in vitality and leaf area of shoots (Fig. 6c). Synthesis of assimilatory pigments in tissue cultures is also controlled by amounts of cyto-kinines in the cultivation media (KAUL and SABHARWAL, 1971; SALAJ and BLEHOVÁ, 2006).

Differences among values labelled with the same symbols (a)–(d) are not statistically significant at 95 % significance level (Duncan test).

Table 5. Analysis of variance (ANOVA) of growth characteristics of tissue cultures of Mountain Pieris after the 3rd sub-cultivation, in three experimental variants

Variance	Degrees	F-test				
Variance	of freedom	Number of shoots	Shoot length	Dry mass		
Among treatments	2	8.72*	1.03	2.24*		
Residual (within-treatments)	22					
Total	24					

*statistically significant differences at 95% significance level (P < 0.05).

Experimental variant	Number of shoots / explants \pm SE ¹	Shoot length $[mm] \pm SE^1$	Dry mass / explants $[g] \pm SE^1$
Α	7.60 ± 3.253 a	17.53 ± 4.391 a	$0.0322 \pm 0.0013 a$
В	$10.90 \pm 3.253 \text{ b}$	19.88 ± 2.229 a	$0.0632 \pm 0.0039 \text{ ab}$
С	$5.35 \pm 4.600 a$	16.81 ± 7.572 a	$0.0399 \pm 0.0049 c$

Table 6. Average values of growth characteristics of tissue cultures of Mountain Pieris after the 3rd sub-cultivation

SE¹, standard error of arithmetic mean.

Table 7. Analysis of variance (ANOVA) of chlorophylls concentrations in regenerants of Mountain Pieris after the 3rd subcultivation, in individual experimental variants

Variance	Degrees	F-test				
variance	of freedom	Chlorophyll a	Chlorophyll b	Chlorophyll $a + b$	Chlorophyll a/b	
Among treatments	2	5.73*	1.04	39.28*	2.93*	
Residual (within treatments)	22					
Total	24					

*statistically significant differences at 95% significance level (P < 0.05).

Table 8. Chlorophylls concentrations in regenerants of Mountain Pieris after the 3rd sub-cultivation, in individual experimental variants

Experimental variant	Chlorophyll a [mg g ⁻¹] ± SE ¹	Chlorophyll b [mg g ⁻¹] ± SE ¹	Chlorophyll $a + b$ [mg g ⁻¹] ± SE ¹	Chlorophyll a/b [mg g ⁻¹] ± SE ¹
Α	$3.86 \pm 1.691 a$	$1.76 \pm 0.815 a$	5.82 ± 2.497 a	$2.23 \pm 0.278 \text{ a}$
В	4.94 ± 1.609 a	$2.16 \pm 1.001 \text{ a}$	$7.10 \pm 2.524 \text{ ab}$	$2.47 \pm 0.623 \text{ ab}$
С	$6.66 \pm 0.582 \text{ b}$	$2.36 \pm 0.267 \text{ a}$	$9.02 \pm 0.508 \text{ b}$	$2.87 \pm 0.471 \text{ b}$

SE¹, standard error of arithmetic mean.

Differences among values labelled with the same symbols (a)–(d) are not statistically significant at 95% significance level (Duncan test).



Fig. 6. Propagation of Mountain Pieris by the method *in vitro* – a) primary culture; b) multi-shoot tissue culture after the 3^{rd} sub-cultivation – experimental variant B; c) multi-shoot tissue culture after the 3^{rd} sub-cultivation – experimental variant C.

Propagation of plants in the family Ericaceae *in vitro* was also studied by NORTON, M. E. and NORTON, C. R. (1985) who compared the effects of cytokinines: N⁶-benzyladenine (BA) and 2iP. These authors as well as most of the others obtained better results with using 2iP, as BA was toxic for many species and the shoots became necrotic. MALÁ and ŠíMA (2000) report that the success in micropropagation depended primarily on the appropriate cultivation environment (appropriate cultivation medium, temperature, moisture, light). However, important are also so called indirect factors – sampling date of primary explants, age and physiological fitness of donor plants, surface sterilisation and preparation of explants.

Acknowledgement

This work was supported from the Projects VEGA No. 2/0159/11 and VEGA No. 2/0085/09.

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> Received December 6, 2012 Accepted March 27, 2013

Introgressive hybridization between Scots pine and mountain dwarf pine at two localities of northern Slovakia

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Abstract

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Introgressive hybridization within hybrid swarm populations of *Pinus sylvestris* and *P. mugo* was analyzed in the localities Medzi Bormi and Sokolie, northern Slovakia, using species-specific *trnV-trnH/Hinf* I restriction profiles of chloroplast DNA of needles and zygotic embryos. The presence of trees with both *P. sylvestris* and *P. mugo* haplotypes indicates hybrid nature of the swarms. Molecular analysis revealed a relatively high rate of introgressive hybridization between putatively hybrid swarm individuals and the parental trees of the pure species *P. sylvestris* and *P. mugo* growing in the same localities. The proportion of hybrid seeds with *P. sylvestris* pollen donor averaged at 30.3%, whereas those with pollen donor of *P. mugo* at 19.8%. The results are discussed within context of the established crossability relationship between *P. sylvestris* and *P. mugo*.

Keywords

introgressive hybridization, P. mugo, Pinus sylvestris

Introduction

Scots pine and mountain dwarf pine represent the two principal species of pines indigenous to Slovakia with predominantly allopatric distribution in the country. The altitudinal range of the former extends from the lowlands of 200 m a.s.l. up to 1,465 m (KRIPPEL, 1986), whereas that of the latter between 1,400 m and 1,800 m (SOMORA, 1981). However, in some places of northern Slovakia the natural habitats of these species overlap creating conditions for their spontaneous hybridization. The hybrid swarms generated in this way involve the hybrid individuals along with the trees of Scots and mountain dwarf pines. The hybrid nature of these swarms exerts profound effect on reproductive behaviour of their trees. Except for hybridization of hybrid individuals with each other, the back-crossing of the hybrids with one or both parental species is supposed to take place in a variable extent. The process is called introgressive hybridization and is believed to contribute substantially to the increased variability in participating species (ANDERSON, 1949).

In present study, we followed this process in two hybrid swarm populations of Scots pine and mountain dwarf pine in northern Slovakia using DNA molecular markers.

Material and methods

Species and populations

The hybrid swarm population of Scots pine (*Pinus sylvestris* L.) and mountain dwarf pine (*P. mugo* Turra)

growing in the peat-bog locality "Medzi Bormi" near Zuberec (alt. 815 m) together with the hybrid swarm growing on calcareous rocks of the locality "Sokolie" near Terchová (alt. 1,172 m) were subjected to molecular analysis of introgressive hybridization. As a control, the pure species populations of *P. mugo* in Roháče (alt. 1,500 m) and *P. sylvestris* in Hruštín (alt. 800 m) were used.

The principle according to which introgressive hybridization assessment was made involved comparison of the chloroplast DNA (cpDNA) haplotypes of both needles and zygotic embryos of a given tree. Owing to the paternal inheritance of cpDNA in conifers (WAGNER et al., 1987), the identical haplotype of the maternal tree (haplotype P. sylvestris) and its zygotic embryo (haplotype P. sylvestris) was taken for intraspecific crossing. On the contrary, the opposite haplotypes of the maternal tree (haplotype P. sylvestris) and its embryo (haplotype P. mugo) was taken as an evidence for introgressive hybridization between hybrid individual of P. sylvestris haplotype and P. mugo. Molecular analysis of the hybrid swarms Medzi Bormi a Sokolie was carried out twice, in the years 2009 and 2010 involving 23 trees in the locality Medzi Bormi in 2009 and 10 trees in 2010. At the locality Sokolie, 20 trees were subjected to analysis in 2009 and 19 trees in 2010. Control populations of P. mugo and P. sylvestris were represented in the experiment with 10 trees each subjected to analysis in 2009 only. The extent of experiment performed with hybrid swarm populations depended on the number of trees bearing cones in a given year.

DNA extraction

Total DNA was separately extracted from needles of individual trees using CTAB method by MURRAY and THOMPSON (1980). The number of seeds from corresponding trees which were involved into study ranged between 9 and 10 sound seeds. In *P. sylvestris* from Hruštín 15 seeds per tree were used.

Following a 3–5 day embibition of seeds on wet paper wool in Petri dishes, the embryos of individual seeds were excised from surrounding female gametophyte tissue and used in DNA extraction. Each embryo was extracted in Eppendorf tube using rotatory homogenizer with a pestle. More advanced germinating embryos were extracted with NucleoSpin Plant Kit (Macherey-Nagel), those which had not protruded from the seed coat with Simax[™] Genomic DNA Extraction Kit (Beijing SBS Genetech Co., Ltd.).

PCR-RFLP analysis

The *trn*V-*trn*H region of cpDNA was PCR amplified as described earlier (KORMUŤÁK et al., 2008). The obtained PCR products were digested with restriction enzyme *Hinf* I, which was found to discriminate the cpDNA of

P. sylvestris from that of *P. mugo* (WACHOWIAK et al., 2006a). The generated fragments were electrophoretically separated in 2 % agarose gels with EtBr and $1 \times$ TBE buffer.

Results

Amplification of cpDNA *trnV-trn*H region of needles and embryos resulted in PCR-product of approximate size of 2,300 bp in both species (Fig. 1, lane-a). Its digestion with restriction enzyme *Hinf* I generated four fragments in *P. sylvestris* needles and three fragments in *P. mugo* needles, respectively (Fig. 1, lanes b–h; Fig. 2). No individual variation in restriction profiles of *P. sylvestris* and *P. mugo* was observed.



Fig. 1. PCR-product of *trnV-trn*H gene region of cpDNA (lane a) and its restriction fragments generated by *Hinf* I in *P. sylvestris* individuals (lanes b–h); M, molecular size marker.

This indicates necessarily the species-specific nature of the differences scored in these species. Of particular importance there was in this respect the DNA fragment of 700 bp size characteristic for *P.mugo* along with the 680 bp size fragment characteristic for *P. sylvestris* which have enabled to discriminate reliably between the parental species and to score efficiently the gene flow between the trees of hybrid swarms. The results of this study are summarized in Table 1.

As expected, all the trees of *P. sylvestris* in Hruštín shared *P. sylvestris* haplotype and the same was true of their embryos which exclusively exhibited *P. sylvestris* haplotype. Similar figure was characteristic for *P. mugo* population in Roháče with *P. mugo* haplotype shared uniformly by the scored trees and their embryos.



Fig. 2. *trnV-trnH/Hinf*I restriction profiles of eight *P. mugo* individuals (lanes a–i) and five *P. sylvestris* individuals (lanes j–n); M, molecular size marker.

Locality	Year	Individ	luals	Number of	Embryo h	aplotype	Hybrid
		Haplotype	Numb.	anal. seeds	P. sylvestris	P. mugo	seeds [%]
Hruštín	2009	P. sylvestris	10	153	153	0	0
Roháče	2009	P. mugo	10	100	0	100	0
Medzi Bormi	2009	P. sylvestris	16	170	123	47	27.6
		P. mugo	7	76	41	35	53.9
	2010	P. sylvestris	2	20	11	9	45.0
		P. mugo	8	80	9	71	11.2
Sokolie	2009	P. sylvestris	8	84	56	28	33.3
		P. mugo	12	114	32	82	28.0
	2010	P. sylvestris	18	175	170	5	2.8
		P. mugo	1	10	3	7	30.0

Table 1. Frequency occurrence of introgressive hybrids in localities Medzi Bormi and Sokolie in 2009 and 2010

Contrary to the control populations mentioned above, the hybrid swarm populations are much more heterogeneous involving the trees of both *P. sylvestris* and *P. mugo* haplotypes. As it follows from Table 1, their proportions varied not only in the tested localities but also annually in a given locality. Contrasting differences in proportions of the two haplotypes in 2009 and 2010 in the localities Medzi Bormi a Sokolie were due to the preferable sampling of those trees which bore mature cones in the respective years. For example, there were 8 trees of *P. sylvestris* haplotype and 12 trees of *P. mugo* haplotype on Sokolie scored in 2009 but the next year the reverse figure was obtained in the locality involving 18 trees of *P. sylvestris* haplotype and 1 tree of *P. mugo* haplotype only.

Considerably variable was also the rate of introgression on individual localities, including annual variation. Among 170 embryos scored in 2009 in 16 trees of *P. sylvestris* haplotype in the locality Medzi Bormi, 123 embryos were found to originate from pollination with P. sylvestris pollen and 47 embryos from pollination with P. mugo pollen. The amount of hybrid embryos P. sylvestris \times P. mugo has accordingly attained the level of 27.6%. Even higher was the rate of introgression between the trees of *P. mugo* haplotype in the locality with P. sylvestris pollen, as evidenced by the 53.9% proportion of hybrid embryos *P. mugo* \times *P. sylvestris* in 2009. The reverse situation was ascertained in 2010 with 45% proportion of *P. sylvestris* \times *P. mugo* hybrid embryos and 11.25% share of the embryos of reciprocal combination detected in the same locality. Though lower, the rate of introgression in the locality Sokolie has exhibited the same tendency as described in the locality Medzi Bormi, with profound differences in proportions of P. sylvestris \times P. mugo and P. mugo \times p. sylvestris embryos in individual years (Table 1).

Summarily, the rate of introgression between the putative hybrid individuals of both localities investigat-

ed and the parental species *P. sylvestris* and *P. mugo* differs considerably. Among the total number of 449 seeds originating from 44 individuals of *P. sylvestris* haplo-type, 89 hybrid embryos of *P. sylvestris* × *P. mugo* were detected representing 19.8% share. On the contrary, the proportion of hybrid embryos *P. mugo* × *P. sylvestris* averaged at 30.3%, based on 85 hybrid embryos revealed among 280 seeds from 28 putative hybrid individuals of *P. mugo* haplotype. The obtained data indicate different intensities in gene flow between individuals of the putative hybrid swarms of *P. sylvestris* and *P. mugo* on one hand and respective parental species on the other hand.

Discussion

Of the two hybrid swarm populations investigated, only population Medzi Bormi was subjected to a detailed morphometric analysis. Based on needle morphology and needle anatomical traits, the hybrid nature of the population was postulated by MUSIL (1975) and VIEWEGH (1981). In case of the population Sokolie only indirect evidence of its hybridity has recently been provided involving intermediate habitus of the trees, reduced size of the cones and reduced number of seeds per cone (KORMUŤÁK et al., 2009). All these characteristics are also shared by the three additional hybrid swarms in northern Slovakia, i. e. those in Tisovnica, Suchá Hora and Medzi Bormi (KORMUŤÁK et al., 2011). Homogeneous nature of the controlled populations uniformly represented by the P. sylvestris haplotype in Hruštín and/or by the P. mugo haplotype in Roháče along with a heterogeneous nature of investigated hybrid swarm populations which involve the trees of both the above mentioned haplotypes may be taken for a strong hint indicating hybridity of the swarms. It is not clear whether tested trees of the hybrid swarms represent F_1 generation of the true hybrids between *P*. sylvestris and P. mugo or if they are back-crosses of the F₁ generation individuals with the parental species already. However, their ability to intercross with P. sylvestris and P. mugo provides a strong support for introgressive hybridization taking place in both investigated localities with hybrid swarm populations. It follows from the data obtained so far that hybrid swarm individuals intercross more efficiently with P. sylvestris than with P. mugo. This finding is in contradiction with the results of artificial hybridization between P. sylvestris and P. mugo carried out by WACHOWIAK et al. (2006b) as well as with the data of the authors referring to the lack of evidence for the hybrid swarm in sympatric population of P. mugo and P. sylvestris in the locality "Bór na Czerwonem" at the northern foothills of the Tatra Mountains in Poland. In both cases the authors observed a rare hybridization between the parental species with only unidirectional gene flow from P. mugo to P. sylvestris. The reciprocal crosses P. $mugo \times P$. sylvestris

have not been identified so far. The only explanation for a relatively high hybridological afinity between hybrid swarm individuals in the tested localities and the parental species *P. sylvestris* and *P. mugo* is a weakening of reproductive barriers between participating individuals due to the hybrid nature of the involved hybrid swarm individuals. However, this assumption needs to be verified further with the emphasis on proving hybridity of the hybrid swarm individuals.

Acknowledgement

This study was financially supported by the VEGA Grant Agency, project no. 2/0076/09.

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Received December 6, 2012 Accepted March 26, 2013

Phenological season onsets in the Czech Republic

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Abstract

Kožnarová, V., Hájková, L., Bachanová, S., Richterová, D., Škvarenina, J. 2013. Phenological season onsets in the Czech Republic. *Folia oecol.*, 40: 206–219.

Phenological season onsets are defined according to the phenological stages onsets in typical plant species occurring in the Czech Republic. For processing phenological phases were chosen: beginning of flowering 10% of *Corylus avellana* L., *Alnus glutinosa* (L.) Gaert. and *Galanthus nivalis* L. (pre-early spring); beginning of flowering 10% of *Cerasus avium* (L.) Moench, *Betula pendula* Roth. and *Acer platanoides* L. (early spring); fully leaved *Betula pendula* Roth., *Acer platanoides* L. and *Cerasus avium* (L.) Moench. (full spring); beginning of flowering 10% of *Tilia cordata* Mill., *Sambucus nigra* L. and *Dactylis glomerata* L. (early summer); full ripeness of *Sambucus nigra* L. and *Sorbus aucuparia* L. (full summer); leaves colouring 10% of *Betula pendula* L., *Sorbus aucuparia* L. and *Tilia cordata* Mill. (early autumn); leaves fall 100% of *Sambucus nigra* L., *Sorbus aucuparia* L. and *Tilia cordata* Mill. (end of autumn). The season onsets were calculated from 40 phenological stations in the Czech Republic within 1991–2010. Phenological season onsets were executed by GIS methods (Clidata-DEM) into maps. The effect of altitude on the phenological phase onset and duration of selected phenological period (flowering and ripening of black elder) is described in phenotermopluviogram at selected stations (Lednice, 155 m a.s.l.; Měděnec 830 m a.s.l.).

Keywords

phenological phase, phenological season, GIS, phenotermopluviogram

Introduction

Commonly observed phenological events include the timing of sprouting and flowering of plants in the spring, colour changes of plants in the fall, bird migration, insect hatches, and animal hibernation. Because the occurrences of such seasonal phenomena are generally initiated and driven by climate, phenological record is a sensitive proxy for investigating climate change and its influences on ecosystems over time (HAJKOVÁ et al., 2012). Inevitably, climate and its variability, including the ongoing current changes at various spatial and temporal scales, must impact on agricultural activi-

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ties. Various indications for shifts in plant and animal phenology resulting from climate change have been observed in Europe. During recent years, phenology, the science of the timing of seasonal plant and animal activity, has had an increasing attention in the context of climate change (MENZEL et al., 2001).

The natural seasonality is described with phenological calendars (SCHNELLE, 1955), which are lists of annual sequence of phenological phases in the form of their starting dates, their duration and the intervals between the phases (AHAS et al., 2000).

The year can be divided into phenological season according to the reaction of nature to the actual course

of weather. Phenological seasons are determinated by typical development phases of plants which are in connection with the change in the weather course throughout the year (e.g. bud burst, inflorescence emergence, first leaves, flowering, ripeness, leaves colouring). Similarly, individual season could be described according to the behaviour of animals (e.g. arrival and departure of migratory birds, first flight of bees and occurrence of plant pests).

Phenological maps including the map of the arrival of spring according to IHNE (1885) were stated in the publication "Phenological observation in Moravia and Silesia in the years 1923 and 1924" (Novák and ŠIMEK, 1926). The authors declare: "It is necessary to use different seasons than astronomical or meteorological for knowledge phenological conditions in central European climate. The beginning and duration of phenological seasons significantly differ according to the latitude and elevation. Apart from the 4 basic seasons: spring, summer, autumn and winter, as a rule usually some other adjoining grades are distinguishable, which interpret the relation of the life phases to the atmosphere more exactly." We usually distinguish (according to IHNE, 1885) pre-early spring, spring, summer, autumn and winter (NEKOVÁŘ and HÁJKOVÁ, 2010; HÁJKOVÁ et al., 2011).

The phenological seasons are defined according to other authors (Kožnarová and Klabzuba, 2004; Petкік et al., 1986) subsequently: phenological pre-early spring is period onset of growing season (e.g. flowering of snowdrop, coltsfoot, snowflake and hepatica), trees and shrubs, e.g. hazel, sallow, cornel, common alder, efflorescent before foliage, and spring works start. Phenological spring is divided into early and full and it corresponds with onset of the main growing season. For phenological early spring is typical when trees (cherry, apple tree, pear-tree) have blooms and leaves at the same time, in full spring full leaved lilac and crane are flowering, and most grass too. Phenological summer classifies into early and full as well, period between summer and autumn is so-called Indian summer. Gradual colouring of leaves and harvest root-crops are characterized for autumn. Defoliation is typical for end of autumn, the main growing season is finishing and winter is coming.

Onsets and durations of phenophase are influenced by weather in each year and weather conditions are expressed usually by daily temperature and precipitation. Other characteristics described air circulation and components of radiation balance are often disregarded even if it is possible to express them by synoptic weather situation.

As explained EITZINGER et al. (2010), there are several seasonal climate variations, such as the El Nino Southern Oscillation or the North Atlantic Oscillation, which are important in determining seasonal weather patterns and can impact on crop yields. Studying how current climatic variability influences crop yields is not only important in providing a baseline for estimation of future impacts, but it can also give insights into potentially useful crop prediction methods. Climate factors represent one of the main inputs for plant growth and have a direct effect on many plant physiological processes such as the onset and duration of phenological stages.

The large-scale circulation of the atmosphere is one of the principal components determining the regional variation in the climate, including wind, temperature and precipitation. The increasing interest in the development synoptic classifications allows interpretation of surface environmental processes and patterns in the synergistic effects of atmospheric characteristics (Bower et al., 2007). Traditional manual subjective methods such as those proposed by LAMB (1950) for British Islands, or HESS and BREZOWSKY (1977) for Europe, are today replaced by or combined with objective or semi-automated techniques permitting the analysis of large amounts of data using less time and effort (KožNAROVÁ et al., 2009).

The classifications were created for varied purposes, and they differ in the concepts, manifested in the differing spatial and time scale of arrangement of the categories. CHMI (Czech Hydrometeorological Institute) classification, set up for short-term and mediumterm regional forecasts, enables to include an integrated circulation process in its categories, which occurs over a large part of Central Europe; it is thus more generous from the systematic point of view, and also uniform for Bohemia and Slovakia (STEHLIK, 2002).

The aim of this study was to investigate the annual and spatial variability of the phenological season onset and phenological season duration in the Czech Republic with respect to the weather variability (air temperature and precipitation) between 1991 and 2010.

Material and methods

The CHMI phenological network is consisting of 50 wild plants phenological stations. Voluntary observers monitor the onsets of phenophase following CHMI methodological instructions (Number 2, 3, 10). Patterns of phenophase are in the Phenological atlas (COUFAL et al., 2004). The observer carries out observations each lasting two days during the vegetation season, and outside the vegetation period once or twice a week. The vegetation period is defined from March to October. The observed data (expressed in a day of year) are checked and transferred to application Oracle Phenodata (official phenological database of CHMI).

Selected phenological stations with comprehensive database of three plant species were included into the processing. The only exception is obtained phenological stages full onset of summer, which was determined on the basis of a combination of two plant species.

The onsets of individual phenological seasons are defined with some combinations of plants and phenophase. Pre-early spring is recommended to be determined according to the beginning of flowering 10%, i.e. according to the appearance of the first blossoms in the following plants: hazel (Corylus avellana L.), common alder (Alnus glutinosa (L.) Gaert.) and snowdrop (Galanthus nivalis L.). For the determining the phenological early spring, the phenophase beginning of flowering 10% was chosen in wild cherry (Cerasus avium (L.) Moench), silver birch (Betula pendula Roth.) and Norwegian maple (Acer platanoides L.) - flowers and leaves are developed with a short time lag. For the phenological full spring the phenological stage full foliage was chosen. Into the execution were selected silver birch (Betula pendula Roth.), Norwegian maple (Acer platanoides L.) and wild cherry (Cerasus avium (L.) Moench). The period of early summer is characterized by flowering of lime tree (Tilia cordata Mill.), black elder (Sambucus nigra L.) and flowering of grasses e.g. cocks foot (Dactylis glomerata L.). The period of phenological full summer is typical by ripening of black elder (Sambucus nigra L.) and rowan (Sorbus aucuparia L.). The phenological early autumn is characterized by leaves colouring of silver birch (Betula pendula Roth.), rowan (Sorbus aucuparia L.) and lime tree (Tilia cordata Mill.), the end of autumn coincides with leaf fall.

The phenological data of the selected plants were evaluated in the environment of MS Excel, but especially a space analysis in the environment of geographical information systems was carried out. In total, data from 40 stations with MASL (mean above sea level) ranging from 155 m (Doksany, 50° 27' N, 14° 10' E) to 830 m (Měděnec station, 50° 26' N, 13° 08' E) were used for creating the maps. The mean value of three (or two in case of full summer onset) plant species and one phenological stage (e.g. beginning of flowering 10%, fully leaved, full ripeness, leaves fall) were used as an input data into map processing. The data for determination of phenological season were used from the same phenological stations with complete time series. For the depiction of maps, the method Clidata-DEM was used with a horizontal differentiation of 500 m and the distance between two neighbouring phenological stations at similar conditions was 40 km. This method is based on a local linear regression between the measured value (average data of the onsets of the selected phenophase in the period 1991-2010) and a digital model relief. For each station, regressive coefficients from the nearest stations by means of the method of the smallest squares were calculated, which were later consequently interpolated in the space distribution, and by means of map algebra, and a straight line equation, a space distribution of the given phenophase was acquired (TOLASZ et al., 2007). The maps are processed from the observed data of the phenological stations; in the area above the boundaries of the present occurrence the map expresses potentially possible values.

From statistical characteristics we chose these parameters: average, median, lower and upper quartile, standard deviation, minimum and maximum.

In case of a lack of climatological stations with phenological observations, therefore were used socalled "technical" series for meteorological elements in this paper. For the data processing ("technical" series), the software packages AnClim (Štěpánek, 2011a), LoadData (ŠTĚPÁNEK, 2011b) and ProClimDB (ŠTĚPÁNEK, 2011c) were created. They offer complex solution, from tools for handling databases, through data quality control to homogenization of time series, as well as time series analyses, extreme value evaluation and model output verification. Thanks to the "technical" series, we have gained a sufficiently large number of climatological series for subsequent analysis, with equal spatial distribution in the territory of the Czech Republic (Štěpánek et al., 2011). The results of the onsets of phenological phases of plant species of the corresponding phenological station in the given year were associated with "technical" series for geographical coordinates of phenological stations in the period 1991–2010 for further processing.

For a detailed analysis of the impact of different weather conditions on phenophase onsets of black elder (*Sambucus nigra* L.) were chosen two phenological stations at different elevations (Lednice 155 m a.s.l., 50° 27' N, 14° 10' E; Měděnec 830 m a.s.l., 50° 26' N, 13°

08' E) in the monitored period. For climatic conditions description was opted modified Walter-Lieth diagram (Fig.1 and Fig. 2), in which we used agrometeorological year with cold half-year (from October to March) and warm half-year (April-September) for the expression. We completed it by more climatological characteristic. Abbreviations used in graph are:

t = average monthly air temperature (°C); r = average monthly total precipitation (mm); t_{year} = average year air temperature (°C); r_{vear} = average year total precipitation (mm); *abs* t_{max} = absolute maximum of air temperature; $t_{max VII}$ = average monthly maximum of air temperature of the warmest month; $t_{min I}$ = average monthly minimum of air temperature of the coldest month; $abs t_{min}$ = absolute minimum of air temperature; $t_{min XI-III < 0.0 °C}$ = months with average minimum of air temperature <0.0 °C; *abs* $t_{min IX-VI < 0.0 °C}$ = months with absolute minimum of air temperature < 0.0 °C; $t_{min > 0.0 °C}$ = average count of days with air temperature > 0.0 °C; abs r_{max} = absolute maximum of daily total precipitation; $r \ge$ 0.1 mm = average count of days with total precipitation ≥ 0.1 mm; $r \geq 1.0$ mm = average count of days with total precipitation $\geq 1.0 \text{ mm}$; $r \geq 10.0 \text{ mm} = \text{average count of}$ days with total precipitation ≥ 10.0 mm.



Lednice: 16°48' E; 48°48' N; 165 m asl; t_{year}: 10.1 °C; r_{year}: 541.7 mm

Fig. 1. Climagram of Lednice station.



Měděnec: 13°08' E; 50°26' N; 830 m asl; t_{year} : 5.7 °C; r_{year} : 899.0 mm

Fig. 2. Climagram of Měděnec station.

Relation between temperature and precipitation and length of the flowering and ripening of black elder (Sambucus nigra L.) at Lednice and Měděnec represents termopluviogram (KožNAROVÁ et al., 1997). It is constructed by average monthly air temperature, average monthly precipitation total and long-term mean of both these characteristics. On axis \underline{x} there is the temperature of total deviation and on axis \underline{y} there is precipitation of total deviation of long-term mean. The longterm mean of both characteristics is placed in the centre of this diagram. Limits of intervals (Table 1) used in the termopluviogram are calculated according to the World Meteorological Organization recommendation (KožNAROVÁ and KLABZUBA, 2002).

The termopluviogram was modified and final graph is called "phenotermopluviogram". Sums of temperature and precipitation are calculated from daily values within the period from the beginning of flowering 10% to the full ripeness. Black square is symbol for Lednice station and it is expressed by combination of both of these climatological characteristics (sums of temperature and precipitation). The position of the

square in the graph describes condition of temperature and precipitation during the observed period (beginning of flowering–full ripeness) in one year within the interval 1991–2010 (e.g. warm and very wet period). White squares represent Měděnec station.

Results and discussion

Figures 3–10 describe average onsets of phenological phase based on selected species.

Chart (Fig. 3) shows the duration of phenological stages, the vertical phenological gradients are added to the chart. The vertical phenological gradient expresses the time difference of phenological season onset per 100 m elevation; "spring and summer" phenological seasons appear later with increasing elevation, "autumn" phenological seasons occur earlier with increas-

Table 1. Limits of intervals

Temperature Σt [°C]	Precipitation Σr [mm]	Phenological period Σpp [days]	Percentiles [%]
Extraordinary warm	Extraordinary wet	Extraordinary long	<2.0
Very warm	Very wet	Very long	2.0–9.9
Warm	Wet	Long	10.0–24.9
Normal	Normal	Normal	25.0-75.0
Cold	Dry	Short	75.1–90.0
Very cold	Very dry	Very short	90.1–98.0
Extraordinary cold	Extraordinary dry	Extraordinary short	>98.0



Vertical phenological gradient (days/100 m)

Fig. 3. Extreme data of phenological seasons onsets (day of year).

ing altitude. The vertical phenological gradient shows the shift of onset in dependence on elevation, the number is given in an absolute value.

Pre-early spring (Fig. 4) begins at the earliest before the 1^{st} March (day 60). The latest onset of pre-early spring comes after 17^{th} March (day 76) in the border mountain range. The vertical phenological gradient is 4 days/100 m.

The German Weather Service (Abb. DWD) issued its phenological calendar in the year 2004 (relative to altitude of 440 m) and divided it into 10 periods. It is based on the observations by the DWD in the period from 1988 to 2002 inclusive (NEKOVÁŘ et al., 2008). The results of DWD for the pre-early spring onset are between 15th February and 25th March. It begins with the flowering in snowdrop and hazel, continues with the flowering in common alder, coltsfoot, also bud burst in gooseberry.

In the Czech Republic there was the pre-early spring defined as the combination of beginning of flowering 10% in hazel, snowdrop and common alder. The beginning of flowering 10% (median value) in these three species ranges between 5th March (*Galanthus ni-valis*) and 19th March (*Alnus glutinosa*) in the Czech Republic.

Early spring (Fig. 5) starts before 19^{th} April (day 109) and the latest onset is in the mountains regions after 5^{th} May (day 125). The vertical phenological gradient is 3 days/100 m. NEKOVÁŘ et al. (2008) mentioned

for Germany (altitude 440 m) the beginning of early spring 25th March and 29th April (it is limited with the flowering in Forsythia and leaves developing in oak and flowering in apple tree). The early spring was used as the beginning of main and large growing season. The mean value for the Czech Republic is 25th April (day 115). As a long-term average (1969–1998) the beginning of growing season (BGS) in Europe starts on 23rd April (CHMIELEWSKI and RÖTZER, 2002).

The early spring is determined by the beginning of flowering 10% in wild cherry, silver birch and Norwegian maple. The beginning of flowering 10% (median value) in these three species ranges between 22nd April (*Betula pendula*) and 14th May (*Cerasus avium*). According to NovAk and ŠIMEK (1926) the onset of spring in Moravia and Silesia was between 21st April (111 day) and 20th May (140 day) in the year 1923.

Full spring (Fig. 6) begins at the earliest in the lowlands before 4^{th} May (day 124) and in the highest elevations after 20^{th} May (day 140). The phenological gradient is 4 days/100 m.

DWD evaluated for the elevation of 440 m the onset of full spring between 29th April and 29th May – flowering in apple tree and black elder (NEKOVÁŘ et al., 2008). In the Czech Republic there was used for the determination full foliage in silver birch, Norwegian maple and wild cherry. The fully leaved onset (median value) in these three species oscillates between 8th May and 15th May.



Fig. 4. Average onset of pre-early spring.



Fig. 5. Average onset of early spring.



Fig. 6. Average onset of full spring.

Early summer (Fig. 7) comes at the earliest before 3^{rd} June (day 154) the latest onset is e.g. in the highest mountain elevations (after 24^{th} June – day 175). The vertical phenological gradient is 4 days/100 m.

According to the DWD results the early summer revealed between 29^{th} May and 21^{st} June (black elder

and lime tree flowering, ripening of red currant). We have used for determination of early summer beginning of flowering 10% in lime tree, black elder and cocks foot, the median value of these three species is between 25th May (*Dactylis glomerata*) and 26th June (*Tilia cordata*).



Fig. 7. Average onset of early summer.

Full summer (Fig. 8) begins firstly in lowlands and in the lower catchment of the Ohře and Dyje river (before 28^{th} July – day 209), the latest onset of full summer comes after 18^{th} August (day 230). DWD results show the occurrence of full summer between 21st June and 5th August (vine blossoms, gooseberry, late cherries, early plums ripen). Full summer was determinated in our study by fully ripeness of



Fig. 8. Average onset of full summer.

black elder and rowan; the median values are 7th August (*Sorbus aucuparia*) and 21st August (*Sambucus nigra*).

Early autumn (Fig. 9) starts at the earliest at mountain elevations usually before 16^{th} September (day 259) and the latest onset is later than 2^{nd} October (day 275). The vertical phenological gradient is 4 days/100 m. DWD results of early autumn vary between 28th August and 22nd September (dogwood, wild rose, chestnut, oak ripen). Early autumn in the Czech Republic has been set by leaves colouring 10% in silver birch, rowan and lime tree. Median values of these three species are between 17th September (*Sorbus aucuparia*) and 28th September (*Tilia cordata*).



Fig. 9. Average onset of early autumn.



Fig. 10. Average onset of end of autumn.

End of autumn (Fig. 10.) begins in the middle and higher elevations before 21st October (day 294) and the latest onset occurs after 6th November (day 310). The vertical phenological gradient is 3 days/100 m.

the end of autumn in the Czech Republic. The results (median value) of black elder, rowan and lime tree are between 23rd October (*Sorbus aucuparia*) and 30th October (*Tilia cordata*).

The end of autumn of DWD results begins after 15th October. The phenological stage leaf fall indicates

Detailed statistical characteristics of phenological season's onsets (average, median, lower and upper



Fig. 11. Box plots of phenological seasons of year.



Fig. 12. Box plots of spring phenological onsets.

quartile, standard deviation, minimum and maximum) of pre-early spring, early spring, full summer and end of autumn are presented in Figs 11, 12, 13 and 14. The standard deviation of these phenological seasons ranges between 5.4 (early spring) and 8.9 (full summer), the difference between average and median is 1 day.

The duration of phenological seasons is different during the year. The early spring has the shortest duration (19 days on average); on the contrary the early summer shows the longest duration (65 days on average). Pre-early spring takes 46 days on average, full spring lasts 27 days on average, full summer lasts 39



Fig. 13. Box plots of summer phenological onsets.



Fig. 14. Box plots of autumn phenological onsets.

days on average and early autumn takes 35 days on average. The main growing period lasts on average 149 days (early spring-early autumn) and the large growing period takes 184 days on average (early spring-end of autumn).

The duration of period from beginning of flowering to ripeness of black elder (*Sambucus nigra* L.) at stations in Lednice (155 m a.s.l.) and Měděnec (830 m a.s.l.) is described in Table 2. Table 2 presents years with the longest and the shortest duration of phenological phases of black elder.

The phenotermopluviogram on Fig. 15 represents relations between air temperature and precipitation totals in interval of beginning of flowering 10% to full ripeness at location in Lednice and Měděnec.

Long-term mean in the centre of graph (gray point) for Lednice station is $\sum t = 1,470.3$ °C (air temperature);

 $\sum r = 171.4$ mm (precipitation) and duration of flowering and ripening = 75 days. Long-term mean in the centre of graph (the same gray point) for Měděnec station is $\sum t = 1,134.5$ °C; $\sum r = 224.1$ mm and duration of phenophases = 76 days.

The period is shorter in colder and drier years; on the contrary this period is much longer in warmer years with higher amounts of precipitation. These years are expressed in the graph.

The longest period of flowering and ripening at location in Lednice was 89 days (1992), with the sum of air temperature (Σ t) 1,838.5 °C and the precipitation total (Σ r) 141.2 mm; at Měděnec location 103 days (2000), Σ t = 1,402.1 °C and Σ r = 237.8 mm. The shortest interval was at location in Lednice 62 days (1993); Σ t = 1,158.8 °C and Σ r = 173.6 mm, and at Měděnec station 47 days (2004), Σ t = 747.8 °C and Σ r = 161.6 mm.

Table 2. The longest and the shortest duration of flowering and ripening of black elder

Station	Flowering		F	Ripeness		Beginning of flowering – full ripeness	
	Maximum	Minimum	Maximum	Minimum	Maximum	Minimum	
Lednice	Year: 2007	Year: 2006	Year: 2009	Years: 1994, 2007	Year: 1992	Year: 1993	
(165 m a.s.l.)	47 days	21 days	66 days	41 days	89 days	62 days	
Měděnec	Year: 2001	Year: 2008	Year: 2000	Year: 2001	Year: 2000	Year: 2004	
(830 m a.s.l.)	48 days	16 days	79 days	19 days	103 days	47 days	



Fig. 15. Phenotermopluviogram.

Conclusion

It is very difficult to objectively compare phenological stages onsets at different regions and in various periods. Phenological data always show temporal variations of plant development and express the biological limits and their dependence on climate and weather. A significant role is also played by observation and evaluation methodology. There were several fundamental changes in the methodology of phenological observations in the Czech Republic in the recent years. Single procedure used in plant material, building of station network under the auspices of the Czech Hydrometeorological Institute, a new methodology of observation and assessment has begun in 1991 and analysed dataset has a relative short time period (1991–2010).

From our analysis and map processing phenological seasons are as follows: the average onsets of phenological seasons in the selected elevation zones in the Czech Republic are: pre-early spring (1st March–26th March, vertical phenological gradient: 4 days /100 m), early spring (16th April–7th May, vertical phenological gradient: 3 days /100 m), full spring (3rd May–26th May, vertical phenological gradient: 4 days /100 m), early summer (29th May–30th June, vertical phenological gradient: 4 days /100 m), full summer (29th July–30th August, vertical phenological gradient: 4 days /100 m), early autumn (4th September–3rd October, vertical phenological gradient: 4 days /100 m) and end of autumn (14th October–4th November, vertical phenological gradient: 3 days /100 m).

From the pre-early spring to the early spring elapse 46 days, from the early spring to the full summer 110 days and from the full summer to the end of autumn 74 days on average. If we define the growing period from the early spring to the end of autumn (it approximately corresponds with the main growing season), it lasts 184 days on average. MENZEL et al. (2001) have revealed a lengthening of the growing period (the growing period in 1974–1996 was up to 5 days longer than in the 1951–1973 period). SCHEIFINGER et al. (2002) have also found the vegetation period of many plant species has been increased through an advanced onset of spring phases and a forward shift of autumn phases in mid latitudes.

Acknowledgement

This study was supported by "S grant of MSMT CR", "SVV-2011-263202" and VEGA MŠ SR 1/0257/11 (APVV-0423-10).

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Received January 31, 2013 Accepted June 18, 2013

Pines dieback caused by *Cenangium ferruginosum* Fr. in Slovakia in 2012

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Abstract

KUNCA, A., LEONTOVYČ, R. 2013. Pines dieback caused by *Cenangium ferruginosum* Fr. in Slovakia in 2012. *Folia oecol.*, 40: 220–224.

Serious pine dieback was reported in early spring 2012 from several localities in Slovakia. Needle and bark necrosis turning to twig cankers were the most conspicuous symptoms. There were no or at least not significant damages caused by bark beetles, leaf eating insects, root rots neither tracheomycosis. *Sphaeropsis sapinea* (Fr.) Dyko & B. Sutton was also excluded as the main pest agent, which played an important role in *Pinus nigra* Arnold dieback from 2000 to 2007. Laboratory examination revealed *Cenangium ferruginosum* Fr. as the agent responsible for that dieback. The knowledge on the pine dieback based on the field investigation and laboratory studies are presented and the reasons of the predisposing factors are discussed in the following paper.

Key words

Cenangium, damage, dieback, drought, frost, pine

Introduction

Pines cover 7 % of forest land in Slovakia and belong to the most important forest trees (ANONYMUS, 2011). Scots pine *Pinus sylvestris* L. and Austrian pine *Pinus nigra* Arnold are planted mostly at poor stands such as sandy soils and shallow soils on limestones. However, they also occur on deep nutritional kambisoils and just there they have suffered from a severe damage caused by fungal pathogens in 2012.

In the past pines have overcome several episodes of dieback caused by fungal pathogens:

- 1. *Cenangium ferruginosum* dieback reported by LEONTOVYČ (1962) occurred in 1959 to 1960 (KUN-CA, 2004). It was mentioned that climatic extremes played an important role as predisposing factors.
- Invasive needle-cast fungus Mycosphaerella pini Rostr. ex Munk anamorph Dothistroma pini Hulbary was reported for the first time in Slovakia in 1996 (KUNCA and FOFFOVÁ, 2000). At present its oc-

currence is permanent and it is distributed all over Slovakia (KUNCA and LEONTOVYČ, 2002).

- 3. *C. ferruginosum* was found in Nové Mesto nad Váhom in 2001, but without serious dieback (recorded in the Forest Protection Service database in Banská Štiavnica).
- 4. Gremmeniella abietina (Lagerb.) M. Morelet was found in Veľká Fatra Mountains, locality Kráľova studňa, in 2003. Mountain pine (*Pinus mugo* Turra) trees were damaged, but without serious dieback (recorded in the Forest Protection Service database in Banská Štiavnica).
- 5. *Sphaeropsis sapinea* was a serious pathogen on Austrian pines since 2000 through 2007. It still occurs on *Pinus nigra* but after sanitary cuttings many localities recovered very well (KUNCA, 2004).

The aim of the paper is to describe symptoms of the present pines dieback, check involved pathogens, discuss predisposing factors and map the disease spreading.

Material and methods

The source of the information about pines dieback comes from practical foresters. Specialists from the Forest Protection Service settled in Banská Štiavnica were informed about a pine dieback occurrence and invited in order to determine the pest range, the reason of the dieback as well as to suggest the subsequent proper control measures. Several localities with the pines dieback were visited and marked in the map using Corel Draw 11 (Fig. 1). Samples from needles, twigs, bark, roots and soil were examined for pest agents in the field as well as in the laboratories.

In the field inspections were realized mostly to search for insect agents such as bark beetles (*Ips* spp., *Tomicus* spp.). On the roots *Armillaria* signs (syrrocium, and rhizomorphs) were looked for. Other signs of biotic agents and dieback symptoms were checked on other parts of trees as well.

In the laboratories samples were cultivated in wet chambers. Prior to isolation of fungal agents, the surface of samples was sterilized by dipping the sample into the 50 % methanol for 30 seconds. Then they were rinsed with distilled water and dried by filter paper. Samples were cultivated on carrot agar that was prepared from 400 g smashed fresh carrot roots, 18 g of agar powder and filled up to 1,000 ml of demineralized water. These ingredients were mixed and sterilized under 121 °C reaching 210 kPa during 15 min. in Systec DX-90 autoclave. Microscopic features of fungal structures were studied by stereomicroscope Leica S8 APO and microscope Zeiss Axio Scope.A1. Pictures of microscopic features were transmitted on the screen using software Micrometrics SE Premium. Pure cultures were cultivated in climatic chambers Climacell 707. Speed of

mycelial growth was measured under 14 °C, 20 °C and 25 °C with 50% of relative humidity and 40% of light intensity. Isolates of pure cultures were re-cultivated in flow box with horizontal air flow AURA HZ 48.

Samples were collected in all the studied localities where pine dieback with similar symptoms appeared (Fig. 1). The age of these stands ranged from 20 to 100 years. All together about 30,000 m³ of infected wood was assumed.

Pathogenicity of obtained isolates was tested on *Pinus nigra* twigs on 10 trees in Banská Štiavnica region in June and July 2012. Bark of twigs was cut in the square shape with 0.5 cm length of its side. A plug 0.5 \times 0.5 cm of the pure culture of the pathogen grown on carrot agar for 3–5 weeks was inserted into the wound and that was enwind with a parafilm. Bark necrosis was checked after 3 months.

Results

The first symptoms on pines were noticed at the end of the winter 2011/2012, in February 2012 and they showed up within a very short time. Damages were obvious due to brown needles of the whole crown of all or most trees in the stands. However, some trees among damaged ones had healthy twigs and even there were some completely healthy trees. Symptoms were located in the crown and were bound to the twig's diameter not exceeded 10 cm. So, roots, bark and conducted tissues of the trunk were healthy. Needle cast was found on the needles caused by *Mycosphaerella pini*, *Cyclaneusma* sp., and *Lophodermium* sp., but not in a large extent. Most needles were free from any signs of pathogens.



2012 🕚 LS Pukanec, Sv. Anton, LS Divín, LS Víglaš, ML Krupina, LS Partizánske, Zámutov (VT), OZ Nárnestovo, Krásnohorské Podhradie

Fig 1. Pine dieback in Slovakia in 2012.

The first symptoms were visible on needles of all ages and these needles were pale green. Later the base of needles turned brown which is very typical for *Gremmeniella abietina* (BUTIN, 1995; SINCLAIRE et al., 1987). However, microscopic characteristics of our spores did not fit the sizes of *G. abietina* described in common literature (Table 1). Tissues under the needles with the changed colour were at that time already dry. When the bark of twigs was cut in strips, black dots with pycnidia were found. The pycnidia were also visible on the bark, however, much easier on Scots pine than on Austrian pine. The bark necrosis often occurred in 10 to 100 cm long sections, ringing the twig alternating with healthy parts.

In twig samples cultivated in the wet chambers under laboratory conditions the spores production was stimulated. The spores came out in drops of grey slimy mass from the black 0.5 to 1.0 mm large pycnidia. Conidia were staminate with 2 to 3 vacuoles at the ends of conidia and sometimes with central vacuole. They measured $7.1 \pm 0.8 \times 1.9 \pm 0.3 \ \mu m \ (N = 30)$. The shape and the size of the conidia resembled *Phacidium coniferarum* (G.G. Hahn) DiCosmo, Nag Raj & W.B. Kendr. (PŘíHODA, 1959).

Sexual stage, apothecia, seldom occurred among black pycnidia in spring, but they were much more common in the summer. Anyway, the apothecia were first closed like the egg, then spread, with the diameter up to 2 mm, standing on a very short stalk. The hymenium was olive green to grey. It contained a lot of paraphysis and there were some asci with 8 ascospores. Clavate asci measured $92.6 \pm 13.9 \times 14.4 \pm 1.9 \ \mu m$ (N = 30), ellipsoidal ascospores were without septa 10.8 $\pm 1.1 \times 5.9 \pm 0.5 \ \mu m$ (N = 30), but with many small rounded elements. Ascospores were concentrated at the apical end of the asci, because of it that part was a bit dilated so ascospores were grouped in the pile.

Pure cultures were obtained from the slime coming out from pycnidia, not from apothecial tissue. Pure culture on carrot agar was brown in the center and white to transparent on the edge. The growth of the culture was very slow, up to 2 cm in 4 weeks under 25 $^{\circ}$ C, and even slower under 20 $^{\circ}$ C and 14 $^{\circ}$ C.

Pathogenicity of the obtained isolates was proved by pathogenicity tests. There were necrotic lesions under the bark but not visible on the bark. There was no resin and only dead needles above the infected part showed that infection proceeded. By reisolation it was confirmed that the same pathogen came out as was used in the pathogenicity test.

Regarding symptoms, signs of pathogens and the pure culture characteristics it is evident that *Cenangium ferruginosum* is the pathogen responsible for the pine dieback (Table 1).

Discussion

C. ferruginosum occurs all around the Northern Hemisphere (SINCLAIR et al., 1987). In the Czech Republic there was serious *C. ferruginosum* dieback in 2010 as well as in 2004 and the drought was considered as the most important predisposing factor (PEŠKOVÁ and SOUKUP, 2011). SINCLAIR et al. (1987) believe that severe winter frost is the predisposing factor that determines the following successful infection, especially if winter is preceded by unusually mild autumn weather. Similarly BUTIN (1995) considers drought following several months lasting wet period as the most important predisposing factor.

In Slovakia, the specific climatic conditions in certain regions occurred prior to the damage and could play an important role as the predisposing factors. There are some facts about the climate development:

• There was the extremely wet whole year 2010 through the mid-summer 2011, continuously for at least 19 months.

Table1. Microscopic characteristics of two possible fungal pathogens on pines

	Dimensions					
Europian analisa	Diameter of fruiting					
Fungai species	bodies	Ascospores	Conidia			
	[µm]	[µm]	[µm]			
Gremmeniella abietina	0.5-1.2	3–4 cells:	3–4 cells:			
by Butin (1995)		$14.0-20.0 \times 3.3-5.0$	$24.0 - 48.0 \times 2.5 - 3.5$			
Gremmeniella abietina	1.0	4 cells:	3–6 cells:			
by Ellis and Ellis (1985)		$15.0-20.0 \times 3.0-4.5$	$25.0-40.0 \times 3.0-3.5$			
Cenangium ferruginosum	1.0-2.0	1 cell:	1 cell:			
by Butin (1995)		$11.0-13.0 \times 5.0-7.0$	$5.0-6.0 \times 2.0-3.0$			
Cenangium ferruginosum	up to 3.0	1 cell:	not mentioned			
by Ellis and Ellis (1985)		$11.0-14.0 \times 5.0-6.0$				

- The second half of the summer 2011 through the end of winter 2011/2012 was dry.
- There was a severe frost ranging from -15 °C to -20 °C in the winter 2011/2012, lasting for about 20 days from the end of January to the end of February 2012 (ANONYMUS, 2012).

We believe that *C. ferruginosum* had survived in the pine stands as a saprophyte for decades at those stands. As there were good climatic conditions during supernormal wet 19 months (2010 through July 2011) *C. ferruginosum* population could multiply as a saprophyte on dead pine twigs.

According to KARADŽIĆ and MILIJAŠEVIĆ (2008) the infection of trees is possible throughout the year. However, SINCLAIR et al. (1987) describe that C. ferruginosum produces no infectious asexual spores, only ascospores can cause infection. Based on this fact incipient infections begin each year in summer and early autumn and are held in check by host defenses unless these are defeated by environmental damage or by other pests. This pattern would explain the sporadic appearance of symptoms (SINCLAIR et al., 1987). We assume that in Slovakia the infections occurred mostly in summer and early autumn 2011. Then dry autumn 2011 limited proper preparations of pine trees for the following winter hibernation. As later trees were again weakened by the severe permanent winter frost, bark and woody tissues could be colonized by the pathogen that ringed the twigs and top of stems. At the end of winter, the sun heated the bark, needles started to transpirate the rest of water supply and symptoms suddenly burst out.

KARADŽIĆ and MILIJAŠEVIĆ (2008) consider *C. ferruginosum* as one of the most dangerous pathogenic fungus on pine trees. By pathogenicity tests realized with mycelial plugs in the summer was confirmed that *C. ferruginosum* is not only a saprophyte but also a pathogen. In 3 months the twigs were ringed by necrosis and the above part was dead. There was no resin on the bark in the infection point (SINCLAIR et al., 1987) and this symptom differs from other bark canker pathogens such as *Sphaeropsis sapinea* (ZÚBRIK et al., 2008; ZÚBRIK et al., 2013).

Pine plantations are situated mostly in south-western Slovakia, in region named Záhorie. They grow on sandy soils and usually suffer from drought. In spite of severe growing conditions the *C. ferruginosum* dieback did not appear in that region. Diseased pine stands were located mostly in volcanic mountains of Štiavnické vrchy and Javorie and in crystalline mountains of Tribeč. These volcanic and crystalline basic rocks offer a good nutrition reserve of elements for soils developed there. The role of nutrition deficiency could be compared in the future analysis.

Once the trees are infected and colonized by pathogens causing bark necrosis in the crown, the trees are strongly stressed and soon can be infested by secondary invaders such as *Ips acuminatus* Gyll., *Ips sexden*

tates Börn, Tomicus piniperda L., Tomicus minor Htg., Buprestidae, or by other pathogens such as Armillaria sp., or Ophiostoma sp. (KUNCA et al., 2007; NOVOTNÝ and ZÚBRIK, 2004; ZÚBRIK and KUNCA, 2011; ZÚBRIK et al., 2008; ZÚBRIK et al., 2013) and later on some of them might become the primary pest agents.

C. ferruginosum is an important pathogen in some countries in southern Europe (KARADŽIĆ and MILIJAŠEVIĆ, 2008; TEJERINA et al., 2007). In Central Europe it used to be considered more as a saprophyte than a pathogen (PŘíHODA, 1959). Regarding climate change we may get in touch with the pathogenic behavior of *C. ferruginosum* much more frequently than in the past.

Conclusions

Specific climate conditions predisposed pine trees to infection and tissue colonization by *C. ferruginosum*. The disease occurred on deep nutrient soils in mountains in Central Slovakia, but did not occur on sandy soils in pine monocultures of Záhorie region. Pathogen determination was based mostly on microscopic morphological characteristics of ascospores and conidia. Pathogenicity was proven in infectious tests. There were no secondary pests such as *Ips* spp., *Tomicus* spp., *Armillaria* spp., which could accelerate the pine damage.

Acknowledgement

This paper was prepared thanks to the financial support of the Slovak Research and Development Agency based on the agreement No. APVV-0045-10, then thanks to OP Research and Development for the project "Centre of Excellence for Biological Methods of Forest Protection" ITMS 26220120008 and for the project "Advanced technologies of trees protection of the juvenile growth stages" ITMS 26220220120.

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Received December 6, 2012 Accepted April 24, 2013

A comparative analysis of image processing softwares to indirect estimation of leaf area index in forest ecosystems

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Abstract

PASTORELLA, F., PALETTO, A. 2013. A comparative analysis of image processing softwares to indirect estimation of leaf area index in forest ecosystems. *Folia oecol.*, 40: 225–236.

The leaf area of vegetation can be expressed in terms of leaf area index (LAI). This index depends on species composition, developmental stage, prevailing site conditions, seasonality and management practices. LAI is an important ecological parameter because vegetation-atmosphere processes of the canopy are controlled by the foliage. It can be estimated with hemispherical photographs using some commercial and free softwares. The choice of the software and the settings of parameters are two fundamental aspects to obtain suitable data. The paper focuses on the comparative analysis of the LAI data obtained with three image processing softwares (Spot Light Intercept Model, Gap Light Analyzer and WinScanopy) in a case study in Italy. The data were analyzed in a qualitative and quantitative way. The Wilcoxon signed rank test showed statistically significant differences among the three softwares. WinScanopy provides lower LAI values than the other two softwares. The same non-parametric test distinguishing per forest type and forest age showed statistically significant differences among softwares in three forest types (silver fir, Norway spruce and beech forests) and in the young stands. Instead, no statistically significant differences were found in the mature stands.

Keywords

forest canopy, hemispherical photographs, indirect optical methods, leaf area index (LAI)

Introduction

Canopy foliage amount controls many biological, physical and biogeochemical processes in the water, nutrient and carbon cycle (FASSNACHT et al., 1994; HEISKANEN, 2006). The amount of leaf area in the plant canopies influences primary production (or photosynthesis), transpiration, precipitation's interception, microclimate, and energy, water and carbon exchanges between vegetation and atmosphere (LEUSCHNER et al., 2006). A common measure of canopy foliage used in the ecological studies is the leaf area index (LAI) which can be defined as the amount of foliage one-sided area in a canopy per unit of ground surface area (m²/m²) (WAT-SON, 1947; CHEN and BLACK, 1992). LAI is a dynamic parameter that depends on several variables such as species composition, developmental stage, prevailing site conditions, seasonality, management practices and it expresses the photosynthetic and transpiration surface of trees canopies (JONCKHEERE et al., 2005a).

In literature, there are several methods for groundbased estimation of LAI (ASNER et al., 2003), such as: direct methods (destructive harvesting and direct determination of one-sided leaf area, collection and weighing of total leaf litterfall), indirect contact methods (allometry, plumb lines, point quadrats methods) and indirect optical methods (ceptometer, LAI-2000 and hemispherical photographs) (LIANG et al., 2012). Both direct and indirect methods are complementary but the calibration is still necessary for indirect methods. The cross-validations between direct and indirect methods have pointed to a significant underestimation of LAI with the indirect methods. The selection of the most appropriate method, taking into account the physiological process, and the application of new technical solutions are useful strategies to reduce bias or discrepancies in LAI estimation (BréDA, 2003).

The hemispherical photographs were introduced for the first time by ANDERSON (1964) in order to compute the light penetration through the forest canopy. After this first application, this indirect optical method for LAI estimation spreads especially since the development of high resolution digital cameras, which allow images to be rapidly processed after acquisition (THI-MONIER et al., 2010).

The hemispherical photographs analysis is applied in many fields in order to evidence the relationship between LAI and both forest site and stand characteristics such as: light and radiation regime (KUCHARIK et al., 1999; MACHADO and REICH, 1999; GODOY et al., 2010), forest water balance (VAN DER ZANDE et al., 2009), seedling survival and growth (PUERTA-PIÑERO et al., 2007; MARCHI and PALETTO, 2010), and stand response to thinning (DAVI et al., 2008; MAN et al., 2008). Moreover, the LAI is used as an indicator of site quality in closed canopy (COKER, 2006), as a part of data when running ecological models (PIETSCH, 2002), and for improving the accuracy of remote sensing techniques (SPRINTSIN et al., 2007).

From the technical point of view, in order to obtain LAI values useful to support scientists and forest managers two aspects are fundamental: (1) the choice of the software to be used in the image processing and (2) the parameters settings.

In order to contribute to these issues, the main objective of this paper is to provide useful information to scientists and forest managers through the comparison of the LAI values obtained using different softwares (Spot Light Intercept Model, Gap Light Analyzer and WinScanopy) and parameters settings.

Material and methods

Study area

The study was conducted in the Trentino province (North-East of Italian Alps). The climate of the zone is cool, temperate and mild continental. The mean yearly temperature is 11.5 °C, while the annual rainfall averages 883 mm with two main peak periods, in spring (May rainfall averages 94 mm) and autumn (October rainfall averages 110 mm).

The data were collected in the eastern part of Trentino (Alta Valsugana and Adige valleys) in four pure forest types (silver fir, Norway spruce, beech and European larch forests). For each forest type, 8 sample points were selected considering the different stand structures (horizontal and vertical structures) and age classes. According to the data of forest management unit plans, the horizontal structure was subdivided in two qualitative classes (high and low diameter differentiation), while the vertical structure classification distinguished the one-layer stands from the multi-layer stands (PAs-TORELLA and PALETTO, 2013a). The age of forest stands was classified in two classes: young forest stands and mature forest stands.

The sampling unit (plot) is represented by a circular area with a radius of 13 m (surface of 531 m²) according to the standard of the second Italian National Forest Inventory. In each plot, the main site and stand attributes (GPS coordinates, slope, number of trees, species, diameter at breast height and tree height) were collected in order to calculate basal area, average diameter and height, stand volume and density. Besides, a set of hemispherical photographs of the canopy was taken in the plot with the purpose to estimate LAI. The hemispherical photographs were taken using a Nikon Coolpix 990 camera and a fish-eye converter Nikon FC-E8 (Nikon Corporation, Tokyo, Japan) at 1.5 m above the ground. The camera was run in the "programmed auto" mode where it automatically adjusts shutter speed and aperture obtaining the best exposure and using the parameters fixed in Fishevel lens mode (focus fixed at infinity, widest zoom, metering center-weighted, circular frame). Moreover, the camera LCD side was set facing north using a compass so that the top of the hemispherical picture was directed to the north.

In each sample plot a set of 5 images was acquired, the first picture was taken in the central point of the plot and the others at 7 m from the center in correspondence of the four cardinal points (North, South, East and West). Consequently, the total number of pictures collected in the field was 160 (40 images for each forest type).

Software compared and parameters settings

LAI was estimated for each plot using a canopy analysis system developed by Régent Instruments Inc. The canopy analysis system analyzed the circular hemispherical pictures taken by fisheye lens converter with a 183° field of view (FOV). Recently, several commercial software packages as well as freeware programme were developed and used in a broad range of applications (JONCKHEERE et al., 2005b; JARČUŠKA, 2008). Considering the usability (user-friendly software) and the extent of use in the forestry sector, two free softwares – Gap Light Analyzer (GLA) 2.0 (FRAZER et al., 1999) and Spot Light Intercept Model (SLIM) 3.02a (COMEAU and MACDONALD, 2012) - and one commercial software - WinScanopy Pro 2003d - were chosen for this comparative analysis. The last available versions for the free software and the licensed version for WinScanopy (3.02) are used in the study.

WinScanopy is a widely used software designed for canopy hemispherical or rectangular image analysis. WinScanopy's standard system includes a Nikon Coolpix 990 camera, a fish-eye converter Nikon FC-E8 and a self-leveling system (BRÉDA, 2003). Camera and lens were calibrated together by Régent Instruments Inc., which provides also the calibration file for setting the program. Subsequently, to avoid softwares comparison, the parameters (lens manufacturer and properties) were set – whenever possible – in the other softwares on the basis of these calibrations.

GLA computes canopy and site openness, effective leaf area index, sunfleck-frequency distribution and daily duration, and the amount of above- and belowcanopy (transmitted) direct, diffuse, and total solar radiation incident on a horizontal or arbitrarily inclined receiving surface (FRAZER et al., 1999).

SLIM is a program designed to estimate LAI, gap fraction, and fractional transmittance from hemispherical photographs or Licor LAI-2000 Plant Canopy Analyzer data. It is part of a set of programs that model light distribution beneath a forest canopy designed by COMEAU and MACDONALD (2012).

For both free softwares, individual configuration settings can be created and saved to disk for later use such as camera lenses and orientations, topographic settings, regional climatic patterns and growing seasons.

The analysis of hemispherical photographs is composed of six steps (WALTER, 2009): 1) acquisition; 2) input, image editing and registration; 3) classification; 4) data extraction; 5) calculation; 6) output. For the purpose of this comparative analysis we investigated how the type of software and their parameters influence LAI estimation. In each software some parameters are set by default, while others can be set by an operator. In particular, the parameters that can be set manually are the following: image editing, image registration, image classification (thresholding), lens and camera setting, azimuth and zenith per sky region, and model of sky brightness.

The main characteristics of the software and camera in LAI estimation are resumed in Table 1. Software characteristics and specifications are usually comparable but the lens calibration, the equation degree and the elaboration method seem to be specific for each software.

Table 1. Software parameters settings

Parameters	SLIM	GLA	WinScanopy
Version	3.02a	2.0	Pro2003 d
Pixel classification	Automatic (Rider clustering method)	Manual	Manual
Threshold value	_	128	128

Default settings of the camera	No	No	No
Lens calibration	Polar projection	Polar projection	FC-E8
Equation degree	6	-	9
Elaboration method	-	_	LAI-2000 original
Sky Brightness model	SOC model	SOC model	SOC model
Number of sky regions (default)	480 ¹	324	144
Azimuth per region	24	36	18
Zenith per region	20	9	8
Focal length [mm]	8	8	8
View angle [°]	180	183	180

¹Standard BCR (Below Canopy Reading Resolution).

The images – taken in color and saved in the JPG format – were converted to grey scale at the blue channel by the software GNU Image Manipulation Program (GIMP) 2.6. The blue-filtered grey scale is widely considered the best to obtain maximum contrast between trees' crowns and background compared with red- and green-filtered grayscale images (FRAZER et al., 2001). The image registration was set manually, sampling photographs on the basis of its characteristics (image quality, sunflacks, etc.).

The image classification (thresholding) converts greyscale values for each pixel to black (representing foliage and also other non-sky elements) and white (representing sky) for later analysis. The goal of thresholding is to obtain a reasonably accurate discrimination between the background sky and the foliage. In the analyzed softwares, images may be processed setting the threshold manually; two of them can also make an automatic calculation. In particular, SLIM can apply two different models for the automatic thresholding (Nobis and Hunziker or Ridler clustering method), Win-Scanopy allows to calculate the color threshold by an automatic method or by a manual method, while GLA needs to set the threshold manually. For the purposes of this comparative analysis the threshold value was set manually at the default value (128) for GLA and Win-Scanopy, while for SLIM it was set automatically. As suggested by the software manuals, thresholding was evaluated and carefully monitored to obtain the best image (contrast, color, light environment).

The lens characteristics were set to "FC-E8" in WinScanopy (that has been designed to produce this type of projection, FRAZER et al., 2001) and to "polar projection" in the free softwares. In fact the fish-eye converter FC-E8 was designed to produce a simple polar (equiangular, equidistant) projection (HERBERT,

1987; WALTER, 2009) that is characterized by a linear relationship between the radial distance from a projected point to the image centre and the zenith angle between the lens' optical axis and the same point's location in the hemispherical region (Hu et al., 2009). However, it did not conform exactly to this design specification (Englund et al., 2000; FRAZER et al., 2001; INOUE et al., 2004) presenting spectral aberration. SLIM has been developed for use only with true fisheye lenses that utilize equiangular projection but require a sixthorder polynomial for lens calibration so the default calibration was used (Nikkor 8 mm lens). GLA supports four standard projections (polar, orthographic, stereographic, Lambert's Equal Area) and any number of user-defined custom lens distortion (FRAZER et al., 1999). As in SLIM a polar projection and a default calibration were used.

Sky-Region Brightness describes the light intensity of a diffuse sky and it is usually analyzed using a Standard Overcast Sky model (SOC) or a Uniform Overcast Sky model (UOC). The UOC represents conditions avoiding reflections on the lens and blooming effect that are presented by a uniform cloudiness or in the hours before sunrise or after sunset, when no direct solar radiation is present. For the purpose of this comparative analysis a SOC model, that assumes sky brightness at zenith three times as at the horizon (FRAZ-ER et al., 1999), is used because it is considered more efficient under varying sky conditions (STEVEN and UN-SWORTH, 1979).

The number of sky regions, resulting from the intersection of zenith and azimuth regular division in the sky hemisphere, was set in each software as default (see Table 1). From the theoretical point of view, if the number of sky regions increases, the quality of the gap light transmission results should be improved (FRAZER et al., 1999).

Data analysis

The 160 photographs collected in the field were recorded with a resolution of $2,048 \times 1,536$ pixels and a compression of ¹/₄ (FRAZER et al., 2001). The images processed by the three softwares are compared using the descriptive statistics – mean, min, max and standard deviation (SD) – and the non-parametric test of Wilcoxon signed rank test. The Wilcoxon signed rank test was applied because the data does not have normal distribution (Test Shapiro-Wilk: SLIM and GLA W = 0.969, P = 0.001; WinScanopy W = 0.920, P < 0.0001) and we have paired samples (two values for the same observation obtained with different software image processing). Paired samples imply that each individual observation of one sample has a unique corresponding member in the other sample. The descriptive statistics and the Wilcoxon signed rank test were performed by XLStat 2007.

Finally, the method of bivariate line-fitting called standardized major axis (SMA) was used to summarize the relationship between the basal density (x-axis) and the differences between LAI values estimated with couple of softwares (y-axis). SMA has the advantage to use a single dimension (line) in order to describe twodimensional data (WARTON et al., 2006). The intercepts and slops of regression lines of relationship between couple of softwares were compared considering the forest type and the forest age. The graphical representation of the SMA and the estimation of slope, intercept and R² were performed using the "sma" function from "smatr" package in R software.

Results and discussion

The site and stand characteristics measured in the 32 sample plots subdivided in the four forest types (silver fir, Norway spruce, European larch and beech forests) are synthesized in Table 2. The slopes of the plots are in a range of 0° and 23° , while the altitude is between around 550 m and 1,600 m. In order to consider all possible structural situations, the samples included both young stands with high number of stems (more than 900 stems/ha) and low average diameter (25.8 cm) and mature stands with few stems per hectares and high average diameter (41.5 cm). The average volume for all

Table 2. Forest site and stand characteristics of 32 sample plots

Plot	Coord (WC	linates 3884)	Altitude [m]	Forest type	Density [stems ha ⁻¹]	Basal area [m ² ha ⁻¹]	Average diameter [cm]	Average height [m]	Volume [m ³ ha ⁻¹]	Age
	East	North				[]	L' J	ĽĴ		
1	677607	5089786	1,290	Silver fir	714	43.86	21.60	20.9	426	Young stand
2	677472	5089809	1,307	Silver fir	959	31.48	15.30	11.9	338	Young stand
Plot	Coord (WC	dinates 3S84)	Altitude [m]	Forest type	Density [stems ha ⁻¹]	Basal area	Average diameter	Average height	Volume [m ³ ha ⁻¹]	Age
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	East	North				[m² na-¹]	[cm]	[m]		
3	678344	5091521	1,191	Silver fir	714	55.96	26.90	30.0	531	Young
4	691132	5110523	1,336	Silver fir	150	46.77	56.70	38.2	428	Mature
5	690180	5110897	1,432	Silver fir	338	33.03	46.90	29.1	665	Mature
6	691182	5110886	1,240	Silver fir	714	48.32	27.80	20.0	595	Young stand
7	681665	5088305	1,590	Silver fir	752	42.62	29.50	23.1	473	Young stand
8	681665	5088305	1,596	Silver fir	921	52.92	24.90	22.9	461	Young stand
9	672881	5090442	1,109	Norway spruce	1,015	40.45	18.40	8.8	296	Young stand
10	681094	5093158	1,509	Norway spruce	414	19.32	30.20	22.7	262	Young stand
11	681367	5087689	1,450	Norway spruce	620	26.59	25.60	24.8	350	Young stand
12	683752	5097134	984	Norway spruce	395	17.50	33.40	33.3	351	Young stand
13	675247	5112688	1,018	Norway spruce	884	28.24	24.80	14.4	379	Young stand
14	693632	5112669	1,320	Norway spruce	338	48.44	40.10	30.9	472	Mature stand
15	693634	5112672	1,320	Norway spruce	508	35.20	33.10	27.3	452	Young stand
16	694648	5113605	1,400	Norway spruce	282	26.91	43.90	31.0	440	Mature stand
17	673006	5090454	1,123	Beech	959	41.83	20.50	16.3	172	Young stand
18	681367	5087689	1,448	Beech	432	32.73	21.20	22.4	127	Young stand
19	684996	5097349	969	Beech	489	35.12	24.20	19.1	187	Young stand
20	684998	5097352	969	Beech	320	30.71	22.90	23.5	121	Young stand
21	668526	5101564	1,039	Beech	301	18.40	23.30	16.6	162	Young stand
22	669978	5094424	1,230	Beech	395	44.84	28.60	22.6	180	Young stand
23	668972	5095335	1,006	Beech	377	66.73	22.20	15.7	92	Young stand
24	682231	5086941	1,512	Beech	1,450	62.44	14.30	13.2	357	Young stand
25	678659	5105722	1,390	European larch	188	57.03	37.30	24.9	165	Mature stand
26	678976	5106214	1,418	European larch	451	54.82	32.10	27.8	466	Young stand

Table 2. Forest site and stand characteristics of 32 sample plots – continued

Plot	Coord (WC	dinates 3S84)	Altitude [m]	Forest type	Density [stems ha ⁻¹]	Basal area [m ² ha ⁻¹]	Average diameter [cm]	Average height [m]	Volume [m ³ ha ⁻¹]	Age
	East	North								
27	678976	5106214	1,418	European larch	395	56.02	25.80	31.2	209	Young stand
28	678973	5105587	1,549	European larch	301	54.22	22.50	13.5	108	Young stand
29	679003	5105956	1,502	European larch	339	50.20	36.10	28.4	251	Young stand
30	679082	5105837	1,518	European larch	264	25.83	34.40	23.5	178	Young stand
31	679224	5116193	1,102	European larch	188	11.73	45.80	33.1	215	Mature stand
32	666766	5100859	564	European larch	226	41.69	39.60	24.5	189	Mature stand

Table 2. Forest site and stand characteristics of 32 sample plots - continued

sample plots is 316 m³ ha⁻¹, while the average volume per forest type ranged from 175 m³ ha⁻¹ in the beech forests to 490 m³ ha⁻¹ in the silver fir forests. Referring to the forest age, the mature stands have an average volume of 366 m³ ha⁻¹ (average basal area: 37.9 m² ha⁻¹), while the young stands have an average volume of 301 m³ ha⁻¹ (average basal area: 40.7 m² ha⁻¹).

The data obtained with WinScanopy show the lowest LAI mean value (1.91) ($SD_{WinScanopy} = 0.38$), while GLA and SLIM show similar LAI mean values (2.6 and 2.4 respectively) but different standard deviation (SD_{GLA} = 0.38, SD_{SLIM} = 0.45). Wilcoxon signed rank test shows that there are statistically significant differences in LAI (P < 0.0001) among WinScanopy and the other two softwares, while the differences between GLA and SLIM are non-significant. It was hypothesized that these differences are linked to the forest stand density and in order to test this hypothesis the standardized major axis (SMA) was used. The results show that the basal area is not the key variable to explain these differences (Table 3 and Fig. 1). Besides it seems that the three softwares, even starting from the same set of images and applying the same elaboration, calculate different values of LAI that in some cases are also relevant from the statistical point of view. Another study confirms that the outputs obtained by using GLA and WinScanopy for the hemispherical image analysis are not equal in the case of canopy characteristics and below-canopy light conditions (JARČUŠKA et al., 2010). However, these authors asserted that the outputs are mainly influenced by the threshold values set for pixel classification.

Table 3. Coefficients of standardized major axis (SMA) obtained by comparing the results of the softwares

	SLIM_GLA	SLIM_	GLA_
		WinScanopy	WinScanopy
Intercept	-0.843	-0.511	-0.440
Slope	0.022	0.025	0.022
\mathbb{R}^2	0.001	0.032	0.029
Р	0.864	0.322	0.351

Moreover, LAI values estimated by each software were compared to the LAI value calculated as average of three softwares. In 18 cases (56.2%) the single LAI value closest to the mean LAI value is that obtained by GLA, in 12 cases (37.5%) is the value obtained by SLIM and only in 2 cases (6.3%) is the value obtained by WinScanopy. Considering the differences, it is assumed that values of SD < 0.30 are negligible, while the critical threshold is considered a SD \ge 0.5. The plots in the latter situation are 12.5% of the total (plot 3, 9, 11 and 12). In these plots characterized by high differences among LAI values, the choice of one software or another should influence the relationship between estimated LAI and the other forest features (i.e. regeneration, evapotranspiration, canopy water interception). It is interesting to highlight that the four plots are young stands and three of them are Norway spruce forests. Instead, the above mentioned plots differ for the other stand characteristics: the basal area ranges between 18 $m^2 ha^{-1}$ to 56 $m^2 ha^{-1}$ and the stand density is between 395 stems ha^{-1} to 1,015 stems ha^{-1} .



Fig 1. Scatter plots of the basal area (m²) versus the LAI values differences obtained by comparing the results of the softwares.

The descriptive statistics of LAI values per forest type obtained using the three softwares are reported in Table 4. The data obtained by SLIM and GLA show comparable LAI mean values for two forest types (Norway spruce and beech forests), while there are high differences for European larch and silver fir forests. The LAI mean values obtained by SLIM are highest for three forest types, while the highest LAI mean value for the European larch forests are estimated by GLA. WinScanopy estimates LAI mean values lowest for all forest types when compared to the others two softwares. The maximum LAI value is in beech forests for SLIM and WinScanopy, while it is in Norway spruce forests for GLA. Otherwise the minimum LAI value is in European larch forests for SLIM and GLA, while it is in Norway spruce forests for WinScanopy. These data of LAI per forest type are slightly lower if compared with the average LAI values calculated using the data of the two databases of SCURLOCK et al. (2001) and MORI-SETTE et al. (2006). Considering only the indirect optical methods (hemispherical photographs and LAI-2000)

the average LAI values per forest type are the following (PASTORELLA and PALETTO, 2013b): $LAI_{beech \text{ forests}} = 3.33$, $LAI_{European larch forests} = 2.98$, $LAI_{Norway spruce} = 3.49$.

Wilcoxon signed rank test per forest type shows that there are statistically significant differences (P = 0.008) in the following cases: in the silver fir forests between SLIM and GLA, and between SLIM and Win-Scanopy; in the Norway spruce forests between SLIM and WinScanopy; in the beech forests between SLIM and WinScanopy, and between GLA and WinScanopy. Instead, in the European larch forests statistically significant differences were not found.

The graphical representation of the standardized major axis (SMA) and the regression coefficients per forest type highlight a positive relationship between the basal area and the LAI values differences obtained by GLA versus WinScanopy in the silver fir and European larch forests (Table 5 and Fig. 2). Similar results in the same forest types are obtained by SLIM versus WinScanopy but with a lower R² values.

Forest SLIM				GLA				WinScanopy				
type	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD
Silver fir	2.57	2.29	2.85	0.16	2.30	2.09	2.44	0.12	1.89	1.54	2.27	0.25
Norway spruce	2.56	2.19	2.88	0.23	2.53	2.05	3.21	0.43	1.91	1.51	2.27	0.28
Beech	2.53	2.17	3.04	0.31	2.49	2.14	2.74	0.23	1.99	1.54	2.55	0.33
European larch	1.92	1.15	2.43	0.41	2.15	1.89	2.57	0.24	1.84	1.62	2.17	0.19

Table 4. Mean, min, max and SD of LAI values per forest type obtained by three softwares

	SLIM_GLA	SLIM_	GLA_
		WinScanopy	WinScanopy
Silver fir fore	ests		
Intercept	-0.077	-0.352	-0.677
Slope	0.008	0.023	0.024
\mathbb{R}^2	0.018	0.521	0.414
Р	0.752	0.043	0.085
Norway spru	ce forests		
Intercept	1.126	1.134	-0.635
Slope	-0.036	-0.016	0.041
\mathbb{R}^2	0.067	0.0418	0.021
Р	0.537	0.627	0.730
Beech forests	5		
Intercept	0.581	0.984	-0.031
Slope	-0.013	-0.011	0.013
\mathbb{R}^2	0.185	0.002	0.160
Р	0.288	0.922	0.327
European lar	ch forests		
Intercept	-0.977	-1.032	-0.542
Slope	0.017	0.025	0.019
\mathbb{R}^2	0.408	0.689	0.273
Р	0.088	0.011	0.184

Table 5. Coefficients of standardized major axis (SMA) obtained by comparing the results of the softwares per forest type

The descriptive statistics of LAI values per forest age obtained using the three softwares are reported in Table 6. These results show that in the young stands the differences among softwares are greater in comparison with the mature stands. Wilcoxon signed rank test emphasizes statistically significant differences in the young stands between GLA and WinScanopy (P < 0.0001) and between SLIM and WinScanopy (P < 0.0001), while no differences were found in the mature stands. The differences in the young stands are probably due to the fact that these stands are characterized by low density and volume stock that poorly affect LAI estimation. The graphical representation of the standardized major axis (SMA) and the regression coefficients per forest age are shown in Table 7 and Fig. 3.

Synthesizing, the main research finding is that WinScanopy shows LAI values different from the other two softwares suggesting that the choice of the software may strongly influence the estimation. In particular, the highest differences were registered in young and dense forests such as silver fir forests. Results confirm that the need for harmonization and objectivization of techniques at both image capturing and analysis is of fundamental importance as highlighted by JARČUŠKA (2008). Indeed to obtain comparable LAI values is very important for the optimal choice of the silvicultural practices. In particular, in young forests, basal area and LAI have an influence on the choices of the intensity of thinning in order to increase the economic outcome or the car-





Fig 2. Scatter plots of the basal area (m²) versus the LAI values differences obtained by comparing the results of the softwares per forest type.

Table 6. Mean, min, max and SD of LAI values per forest age obtained by three softwares

Forest		SLIM				GLA				WinScanopy			
age	Mean	Min	Max	SD	Mean	Min	Max	SD	Mean	Min	Max	SD	
Young stands	2.48	2.01	3.04	0.28	2.43	1.98	3.21	0.31	1.93	1.51	2.55	0.28	
Mature stands	2.16	1.15	2.85	0.59	2.17	1.89	2.44	0.21	1.84	1.62	2.13	0.19	

bon storage in forest standing biomass (SLODICAK et al., 2005).

Table 7. Coefficients of standardized major axis (SMA) obtained by comparing the results of the softwares per forest age

	SLIM_GLA	SLIM_ WinScanopy	GLA_ WinScanopy
Young forest	stands		
Intercept	-0.219	0.818	-0.470
Slope	0.019	-0.019	0.024
\mathbb{R}^2	0.000	0.014	0.011
Р	0.961	0.571	0.627
Mature forest	stands		
Intercept	-1.052	-1.183	-0.250
Slope	0.035	0.029	0.016
\mathbb{R}^2	0.215	0.096	0.205
Р	0.295	0.500	0.307

Conclusions

Leaf area is a relevant information to investigate biogeochemical fluxes and productivity, consequently different results in the estimation of the LAI value may influence the predictive models. The choice of the software and the parameters settings are two important aspects in consideration of the objective of the analysis. It should take into account technical considerations (i.e. minimum number of available sky regions, possibility of selecting a specific range of colors for thresholding) and the purposes of the work (i.e. research study or management practices).

In general terms, the image analysis may be affected by errors depending from thresholding in pixel classification, Sky Brightness model and number of sky regions. It seems that using automatic processes and standardized methods may improve the quality analysis (JARČUŠKA et al., 2010). Sky brightness may cause an under- or over-estimation of LAI, due to a direct effect



Fig 3. Scatter plots of the basal area (m²) versus the LAI values differences obtained by comparing the results of the softwares per forest age.

on color classification, while the number of sky region may influence direct openness and LAI estimation. Sky Brightness was set as a SOC model in the three softwares so this aspect does not influence our research. The number of sky regions was used as a default parameter but, as showed by VAN GARDINGEN et al. (1999), it seems that a number of segments higher than 100 does not influence LAI estimation. Moreover, during the image analysis it is recommended to choose the software that makes the best thresholding in terms of image quality. In addition, our results show limited differences between softwares using manually (GLA, WinScanopy) and automatically (SLIM) thresholding. In our opinion, the key aspect linked to the thresholding is to monitor step by step the image quality in order to obtain the most precisely estimated LAI. Moreover, findings from this research suggest that threshold setting is not always adequate to explain differences in LAI estimation.

Our results suggest that probably the differences in LAI estimation using different softwares may be due to differences in software approaches (i.e. knowledge, image processing techniques, threshold choice methods). These findings are confirmed by ESPAÑA et al. (2008) and JARČUŠKA et al. (2010). These authors indicated that the estimation of LAI might be very dependent on the

gap fraction model used and associated inversion techniques. The current international literature concerning indirect LAI estimation from hemispherical digital photography focuses primarily on the determination of an optimal threshold value (i.e. GLA, WinScanopy) (CHIA-NUCCI and CUTINI, 2012). In this research framework we investigated the performances of a new software named SLIM.

Our findings indicate that the differences in LAI estimation are not explained by the main forest stand characteristics (e.g. forest type and age). Differences in LAI estimation are related to forest structure (in terms of basal area) only in silver fir and European larch forests. Further researches should deeply analyze these relationships.

Differences in LAI estimation using different softwares are not a relevant problem when the LAI is not the target of the investigation and the data collected are used as support data. Vice versa, if LAI is the target of the analysis and the data are compared to the outcomes of other studies these differences are crucial. Recently, more complex algorithms and softwares (i.e. CAN_EYE software) were developed by adjusting a clumping index in order to provide not only the effective LAI, but also several estimates of the true LAI (DE- MAREZ et al., 2008). Consequently, the future steps of the analysis could be the comparison of the outcomes with the true LAI and the LAI obtained by destructive measurements.

We recommend to implement this kind of studies with up-to-date results about LAI estimation using different softwares. From these further studies it would be possible to foster international sharing of experiences that represents a key to reach higher successfulness in using and developing softwares which concretely support LAI estimation in forest research.

Acknowledgement

This work is a post-doc research programme funded by CARITRO (Cassa di Risparmio di Trento e Rovereto) Foundation. Authors wish to acknowledge CARITRO for their contribution to the realization of this research. We are also grateful to two anonymous referees whose comments considerably helped to improve the article.

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Received December 11, 2012 Accepted May 27, 2013

Selected properties of soil in Nature Reserve Alúvium Žitavy, Slovakia, Danube plain

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Abstract

POLLÁKOVÁ, N., MACÁK, M., HORVÁTOVÁ, M. 2013. Selected properties of soil in Nature Reserve Alúvium Žitavy, Slovakia, Danube plain. *Folia oecol.*, 40: 237–242.

Sufficient knowledge about wetlands, which belong to the most threatened ecosystems in the world, can contribute to their maintaining or recovery. In this work selected properties of three soil profiles in Nature Reserve Alúvium Žitavy were characterized. On studied area, the soil forming substrate was represented by floodplain sediments of the rivers Žitava and Nitra on which Calcaric Fluvisol was developed. With comparison to soil profiles 1 and 2 dug on the left and right bank of river Žitava, soil profile 3 dug near confluence of the rivers Nitra and Žitava contained distinctly higher percentage of clay and had higher porosity, pH values and higher carbonates content. On the contrary, the lowest values of hydrolytic acidity, sum of bases, cation exchange capacity were found in soil profile 3, mainly in Fvc/Gl and Glp horizons. It can be assumed that in this profile the majority of base cations, mainly Ca^{2+} , were bound with CO_3^{2-} anions to solid particles or were part of shell fragments. Obtained results showed, that soil properties in NR Alúvium Žitavy were distinctly influenced by different sediments deposited by the river Nitra versus Žitava and ground water level.

Keywords

carbonates, floodplain, hydromorphic soils, hydrophysical characteristics, pH, soil texture

Introduction

Wetlands are lands where saturation with water is the dominant factor determining the nature of soil development and the types of plant and animal communities living in the soil and on its surface. Wetlands vary widely because of regional and local differences in soils, topography, climate, hydrology, water chemistry, vegetation, and other factors, including human disturbance (KADLEC and KNIGHT, 1996).

Wetlands represent a border area between terrestrial and aquatic ecosystems. They are characteristic by three basic properties: 1) the soil is flooded or saturated with water, 2) presence of wetland plants (hydrophytes and hygrophytes), 3) presence of hydromorphic soils (ŠEFFER, 1996). Hydromorphic soils are characteristic by the loss or accumulation of various forms of Fe, Mn, S, or C. Iron, when reduced to Fe²⁺ form, became sufficiently soluble and can migrate away from reduced zones and precipitate as Fe³⁺ compounds in more aerobic soil zones. The zones of which where due to reduction conditions removed or depleted the iron coatings from mineral grains are termed redox depletions. They commonly exhibit the grey, low chroma colours of the bare, underlying minerals. Also iron itself turns grey to blue-green when it is reduced. The contrasting colours of redox depletions, or reduced iron zones and zones of reddish oxidized iron, result in unique mottled redoximorphic features. Other redoximorphic features are pointing on reduced Mn and the presence of hard black nodules. Under severely reduced conditions the entire soil matrix may exhibit low-chroma colours (BRADY and WEIL, 1999).

Ecological importance of wetlands includes the protection of biodiversity, natural filtering and water retention, slowing the flow of surface water and reducing the negative impact of flooding, preventing soil erosion, removing and storing greenhouse gases from the atmosphere. Despite important functions, many wetlands are disappearing and many unique organisms are losing with them. The result of widespread wetlands destruction is that they belong to the most threatened ecosystems in the world (NOSKOVIČ et al., 2009; ŠEFFER, 1996).

On the other hand, sufficient knowledge of wetland ecosystems can contribute to their maintaining or recovery. Therefore, since soils are important component of wetland environment, the aim of the work reported here was to characterize Calcaric Fluvisols in Nature Reserve Alúvium Žitavy. Obtained results can contribute to the better knowledge and understanding of soil properties of wetlands that have been preserved in Slovakia.

Material and methods

Alluvium of river Žitava was declared a Nature Reserve in 1993, on acreage of 32.53 hectares. The site is characterized by a diversity of habitats and water, marsh and wetland vegetation.

Studied area is located in the south part of Slovakia at the Danube plain, at the confluence of the rivers old Žitava and old Nitra (LUKNIŠ et al., 1972; LELKES et al., 2006). The climate in area is warm and dry, long term (1951–1980) average annual air temperature is 9.9 °C and long term (1901–1990) average annual rainfall is 558 mm (LAPIN, 2012).

The geological substrate is composed of Quaternary Holocene alluvial sediments. Sandy layers form their substantial part. The bottom is formed by gravel or gravel with medium content of sand. The Quaternary sands and gravels are overlaid by loess on most of the area. The permeability of porous aquifers is very good. The area has high level of ground water and is rich on flowing and standing water. Predominant relief consists of planes and floodplains. Major soil types in the wider area are Mollic Fluvisol and Haplic Chernozem (LUKNIŠ et al., 1972; LELKES et al., 2006).

Soil properties were characterized in three soil pits dug in the autumn 2009:

- Soil profile 1 (47°51'83" N, and 18°09'25" E) was located 30 meters from the left bank of the river Žitava and in 2.3 km distance from the estuary of river Žitava, on wet meadow behind the dyke.
- Soil profile 2 (47°51'58" N, and 18°08'30" E) was located 2 meters from the right bank of the river Žitava and in 1.7 km distance from the estuary of the river Žitava.

Soil profile 3 (47°50'81" N, and 18°07'60" E) was located 2 meters from the right bank of the river Žitava, in 30 m distance from the point, where river Žitava flows into the river Nitra.

Morphological properties were described in each soil profile and samples for analysis of soil physical and chemical characteristics were taken.

Basic physical and hydrophysical parameters (FIA-LA et al., 1999) were determined for each of 0.1 m layer till depth of 0.5 or 0.7 m; soil texture was determined for each horizon by pipette method (FIALA et al., 1999); total soil organic carbon content (C_T) by Tyurin method (ORLOV et al., 1981); soil reaction – potentiometrically in 1 mol dm⁻³ KCl (1:2.5); content of exchangeable bases and hydrolytic acidity by Kappen's method, carbonate contents – volumetrically (FIALA et al., 1999).

Average value of each soil parameter was calculated from three repeated analyses.

Results and discussion

The soil forming substrate was presented by floodplain sediments of the rivers Žitava and Nitra on which was developed Calcaric Fluvisol (WRB, 2006). Studied profiles differed in some morphological characters, mainly in horizons thickness, colour, texture, structure, shell fragments and gravel content, ground water level, occurrence of Fe^{3+} mottles reflecting seasonal water table fluctuations giving rise to cycles of reducing and oxidising conditions. The abundance of mottles increased with soil depth, above which the water table fluctuated.

Morphological description of soil profile 1

Calcaric Fluvisol

Ac 0.0–0.20 m, 7.5 YR brownish black (3/2), without mottles, moist, coherent, loamy, without gravel, granularly-angular structure, strongly penetrated by roots, slightly calcareous

Fvc 0.20–0.48 m, 7.5 YR brownish black (2/2), without mottles, moist, coherent, sandy-clay-loamy, without gravel, massive structure, medium penetrated by roots, slightly calcareous

Fvc/Gl > 0.48 m, 7.5 YR brownish black (2/2), Fe^{3+} mottles (30%), moist, coherent, clay-loamy, without gravel, massive structure, slightly penetrated by roots, moderately calcareous.

Morphological description of soil profile 2

Calcaric Fluvisol

Ac 0.0–0.20 m, 7.5 YR brownish black (3/2), without mottles, moist, crumbly, loamy, without gravel, granularly-angular structure, strongly penetrated by roots, slightly calcareous

Fvc 0.20–0.38 m, 7.5 YR brownish black (3/2), without mottles, moist, crumbly, loamy, without gravel,

angular structure, medium penetrated by roots, slightly calcareous

Fvc/Gl > 0.38 m, 7.5 YR black (2/1), without mottles, wet, coherent, loamy, with low content of fine gravel, angular structure, Mn nodules, medium penetrated by roots, moderately calcareous, contained shell fragments.

Ground water was found at depth of 0.9 m and the ground water level was stabilised at depth of 0.7 m.

Morphological description of soil profile 3

Calcaric Fluvisol

Ac 0.0–0.20 m, 7.5 YR brownish black (3/2), without mottles, moist, crumbly, clay-loamy, without gravel, granularly-angular structure, strongly penetrated by roots, moderately calcareous

Fvc/Gl 0.20–0.40 m, 7.5 YR dull brown (6/3), with Fe³⁺ mottles, wet, crumbly, clay-loamy, without gravel, massive structure, Mn nodules, medium penetrated by roots, strongly calcareous

Glp > 0.40 m, 7.5 YR grayish brown (4/2), with Fe³⁺ mottles, wet, coherent, clay-loamy, without gravel, massive structure, Mn nodules, slightly penetrated by roots, strongly calcareous, contained shell fragments.

Ground water was found at depth of 0.9 m and the ground water level was stabilised at depth of 0.55 m.

Physical properties of wetland soils cannot be for most cases easily generalized. Texture is a fundamental index of soil physical properties. Knowledge of this property allows prediction of many other soil characteristics. Soils in flood plains show different textural patterns as a result of differences in parent material and modes of deposition of the materials (OBI, 1989). Textural composition of studied soil profiles reflects textural composition of substrate, which rivers Žitava and Nitra deposited in alluvial plain. The textural composition of individual horizons within soil profiles 2 and 3 slightly differed, but overall, texture in whole soil profile 2 was loamy, and in soil profile 3 clay-loamy. The different textural classes for each soil horizon were determined only within soil profile 1 (Table 1). Compared to other soil profiles, the most clay fraction was found in soil profile 3, dug in the locality, where the river Žitava flows into the river Nitra. This can be due to heavier alluvial sediments deposited by river Nitra which texturally differed from that of river Žitava, owing to the redoximorphic processes began already at depth of 0.2 m. Generally, by action of redoximorphic processes the clay production occurs too. Compared to our results, KUKLA and KUKLOVÁ (2009) found in Fluvisol in NR Chynoriansky luh much higher content of clay fraction (32-63%), with maximum in central parts of studied profiles. They stated that influence of processes of illimerization and colmatation could be coupled (argilization caused by percolation of turbid flood water).

Particle density (ς_s) is relatively stabile soil parameter and usually increases with depth. It depends on density of soil minerals and organic matter (ZAUJEC et al., 2009). Considerable variation of ς_s values in soil profiles was caused by accumulation of alluvial deposits with different particle density, which influenced ς_s parameter of these soils (Table 2).

Regular increases of bulk density (ς_d) and decreases in porosity (P) with depth reflect increased compaction by the overlying sediment in soil profiles 1 and 2. On the contrary, for values of ς_d and P there is no systematic pattern of variation with depth in soil profile 3 (near confluence of rivers Nitra and Žitava). Moreover, the layer 0.2–0.3 m exceeded critical values ($\varsigma_d > 1.4 \text{ tm}^{-3}$; P < 47%) for clay loam (FULAJTÁR, 2006) and was compacted. In this layer macro-pores and air porosity were nearly completely reduced (Table 2).

Critical values of bulk density and porosity were exceeded in whole soil profile 2 beside the layer 0.0-0.1 m. FULAJTÁR (2006) noted, that compacted soil horizon exhibiting P values below 47% for clay loam and be-

Soil	Horizon	Depth	Texture			Textural fr	actions [%]		
profile			_	>0.25	0.25-0.05	0.05-0.01	0.01-0.001	< 0.01	< 0.001
		[m]		mm	mm	mm	mm	mm	mm
1	Ac	0.00-0.20	sh	22.1	25.9	11.8	17.0	40.2	23.2
Calcaric	Fvc	0.20-0.48	spi	33.1	17.1	10.2	14.4	39.7	25.3
FIUVISOI	Fvc/Gl	>0.48	si	27.2	14.7	12.9	14.9	45.2	30.3
2	Ac	0.00-0.20	sh	29.8	11.5	13.3	25.5	45.3	19.8
Calcaric	Fvc	0.20-0.38	sh	12.9	19.7	22.1	24.8	45.3	20.5
FIUVISOI	Fvc/Gl	>0.38	sh	13.6	24.0	30.9	17.6	31.5	13.9
3	Ac	0.00-0.20	si	21.1	9.8	17.6	22.2	51.5	29.3
Calcaric	Fvc/Gl	0.20-0.40	si	19.3	8.2	21.6	20.0	50.8	30.8
FIUVISOL	Glp	>0.40	si	17.9	17.4	18.0	14.1	46.7	32.6

Table 1. Particle-size composition of soils

sh, loam; spi, sandy clay loam; si, clay loam.

Soil	Depth	ρ _s	ρ_d	Р	Pk	Ps	Pn	V _A	Θp
profile	[m]	[t r	n ⁻³]			[% vol.]			
	0.0-0.1	2.53	1.15	54.5	42.6	3.0	8.9	9.7	25.9
	0.1–0.2	2.52	1.35	46.4	37.9	2.0	6.5	7.0	23.4
1 Calaaria	0.2-0.3	2.58	1.37	47.8	36.4	2.0	9.4	9.8	18.3
Fluvisol	0.3-0.4	2.62	1.42	45.8	30.8	6.9	8.1	8.5	13.1
	0.4–0.5	2.64	1.44	45.4	31.2	4.6	9.6	10.0	14.1
	0.5-0.6	2.70	1.43	47.0	35.4	1.7	9.9	9.9	25.3
	0.0-0.1	2.54	1.08	57.5	46.9	2.3	8.3	9.4	29.7
	0.1–0.2	2.41	1.35	44.0	40.8	0.7	2.5	2.9	24.6
2	0.2-0.3	2.49	1.41	43.4	39.4	0.9	3.1	3.3	16.0
Calcaric	0.3–0.4	2.54	1.44	43.3	37.7	0.8	4.8	5.1	13.9
Fluvisol	0.4–0.5	2.47	1.45	41.3	37.3	0.7	3.3	3.5	14.9
	0.5-0.6	2.58	1.47	42.0	37.6	_	4.4	4.1	14.2
	0.6-0.7	2.56	1.46	43.0	39.5	0.6	2.9	3.1	19.7
	0.0-0.1	2.67	0.93	65.2	51.9	3.1	10.2	11.4	32.9
3	0.1–0.2	2.41	1.20	50.2	48.6	0.6	1.0	1.3	27.6
Calcaric	0.2–0.3	2.46	1.51	38.6	38.2	_	0.4	0.3	18.1
Fluvisol	0.3–0.4	2.54	1.23	51.6	46.5	0.5	4.6	4.8	27.6
	0.4-0.5	2.51	1.19	52.6	49.1	0.6	2.9	3.2	29.2

Table 2. Soil physical and hydrophysical characteristics

 ρ_s , particle density; ρ_d , bulk density; P, porosity; Pk, capillary pores; Ps, semi-capillary pores; Pn, non-capillary pores; V_A , air filled porosity; Θp , available water capacity.

low 45% for loam tend to inhibit root penetration. High bulk density and low porosity may adversely affect soil biological properties and lead to decreasing of microbial biomass due to oxygen deficiency in the compacted soils (TAN et al., 2005). OBI (1989) noted that alluvial soils do not exhibit any definite pattern with regard to porosity. Where sandy deposits dominate, macro-pores would expectedly dominate. On the other hand, where the deposited material is of high clay and organic matter content, water-logging may be expected to cause pore instability with resultant tendency towards the formation of smaller pores. Nevertheless, according to BEDR-NA et al. (1989), values of total pore space (P) do not give any indication of pore size distribution. Optimal pore distributions are as follows: 1/3 macro-pores (Pn) where aeration and water drainage take place and 2/3 meso (Ps) and micro-pores (Pk) for water retention and capillary elevation. When considering optimal pores distribution, very low amount of macro-pores of total porosity was found in studied profiles (in soil profile 1: 14-21%, in soil profile 2: 6-14% and in soil profile 3: 1-16%). Air porosity was also very low, mainly in soil profiles 2 and 3 (Table 2). High content of capillary water and low aeration caused reduction conditions in lower horizons what resulted to the development of described redoximorphic feature.

Values of basic chemical parameters are written in Table 3. Total soil organic carbon (C_T) decreased with depth in all examined soil profiles, with values ranging from 28.4 to 40.1 g kg⁻¹ for A horizons and 9.1-17.6 g kg⁻¹ for subsoils. Exchangeable soil reaction (pH_{KCI}) ranged from slightly acidic to slightly alkaline. Increased content of carbonates in deeper parts of soil profiles corresponded to higher values of pH_{KCI}. Compared with soil profiles 1 and 2 dug on the left and right bank of river Žitava, soil profile 3 dug near the confluence of rivers Nitra and Žitava contained significantly higher amount of carbonates. This was possibly due to different chemical composition of alluvial sediments deposited by the river Nitra versus Žitava. According to ČURLÍK and ŠEFČÍK (2006) around 80% of the territory which crosses river Nitra contains 5.72% carbonates in humus horizon, whereas the river Žitava crosses territory with 5.72% carbonates only on 65% and the rest of territory contains only 0.16% carbonates. Analogously, concentration of carbonates in soil forming substrates was higher in soils of river Nitra basin compared to soil of the river Žitava basin. Moreover, SZOMBATHOVÁ et al. (2007) reported that Eutric Fluvisol in NR Žitavský wetland (48°09' N, and 18°19' E, 40 km from NR Alúvium Žitavy) did not contain carbonates, and they were presented only in CGo horizon of Mollic Fluvisol. On the contrary, TOBIAŠOVÁ (2010) found in Eutric Fluvisol in floodplain of the river Nitra 1-5% of carbonates.

Additional reason of higher amount of carbonates in studied soil profile 3 could be the content of shell fragments in lower parts of profile which contributes to the rise of carbonate concentration. Consequently, in soil profile 3 the lowest values of hydrolytic acidity (H) were found, since acidic protons were well neutralized by carbonates (Table 3).

In general, the cation exchange capacity (CEC) is related to the type of sediments of predominantly loam to clay-loam character. In the surface layers, the value is also affected by the content of humus. Sum of bases (S) ranged from 86 to 205 mmol kg⁻¹ and CEC from 88 to 212 mmol kg⁻¹. Comparing the sum of bases it is evident, that the lowest values were found in soil profile 3, mainly in Fvc/Gl and in Glp horizons. Presumably, in this profile the majority of base cations, mainly Ca²⁺ (NOSKOVIČ et al., 2010), were bound with CO_3^{2-} anions to solid particles or they were part of the shell fragments. Therefore, the sum of bases and consequently also the cation exchange capacity was low, in spite the clay content was the highest just in soil profile 3 (Tables 1, 3). On the contrary, the values of CEC and also S proportionally increased with increasing of clay content in soil profile 1, where the lowest concentration of carbonates was determined and shell fragments were not occurred (Tables 1, 3). In all soil profiles, the sorption complex was saturated with base cations, mostly ranging between 97-98 %. Studied soils exhibited high buffering capacity related to carbonates content, middle CEC capacity and the high degree of base saturation.

Results obtained in this study showed that soil properties in soil profile 3 (dug in the locality, where the river Žitava flows into the river Nitra) distinctly differed from soil properties in profiles 1 and 2 dug at a greater distance (2.3 and 1.7 km) from the estuary of

the river Žitava. Differences were mainly determined in soil texture, content of carbonates and pH values. Soil properties in NR Alúvium Žitavy were distinctly influenced by different sediments deposited by the river Nitra compared to Žitava and ground water level.

Acknowledgement

The paper was published thanks to G.P. No. 1/0513/12 and 1/0544/13 of the Scientific grant Agency of Ministry of Education, Science, Research and Sport of the Slovak Republic.

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Soil	Horizon	C _T	Н	S	CEC	BS	CO ₃ ^{2–}	pН
profile		[g kg ⁻¹]		[mmol kg ⁻¹]		[9	%]	KCl
1	Ac	40.1	27.4	132.6	160	83	0.8	5.73
Calcaric	Fvc	15.8	7.4	176.6	184	96	0.9	6.19
FIUVISOI	Fvc/Gl	9.1	3.5	204.5	208	98	3.4	6.37
2	Ac	28.4	7.1	204.9	212	97	1.4	6.58
Calcaric	Fvc	17.6	4.7	171.3	176	97	2.0	6.92
FIUVISOI	Fvc/Gl	12.5	3.8	156.2	160	98	3.6	6.97
3	Ac	33.2	3.1	164.9	168	98	4.4	6.92
Calcaric	Fvc/Gl	12.5	2.8	109.2	112	97	13.6	7.24
LIUVISOI	Glp	11.4	2.4	85.6	88	97	17.6	7.43

Table 3. Selected ecological properties of soils

 C_T , total soil organic carbon; H, hydrolytic acidity; BS, base saturation; S, sum of bases (Na⁺, K⁺, Ca²⁺, Mg²⁺); CEC, cationic exchange capacity.

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Received January 28, 2013 Accepted March 6, 2013

Ecophysiological research of European beech (*Fagus sylvatica* L.) in high-productive mixed forests of the Pol'ana Mts in Central Slovakia

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Abstract

PRIWITZER, T., ČABOUN, V. 2013. Ecophysiological research of European beech (*Fagus sylvatica* L.) in high-productive mixed forests of the Pol'ana Mts in Central Slovakia. *Folia oecol.*, 40: 243–250.

This paper presents the results of ecophysiological research of European beech (*Fagus sylvatica* L.) in highproductive mixed forests of the Slovak Pol'ana Mountain. This research was performed in the research and demonstration object Pol'ana - Hukavský grúň. The radiation, temperature and humidity regimes, as well as daily dynamic of photosynthetic activity and electric resistance of cambial tissue are presented within the whole beech crown profile. The impact of meteorological conditions on selected physiological processes was studied. The results confirmed close correlation between a diameter of trees $d_{1,3}$ and biofield, as well as between a biofield and cambial tissue's electric resistance. The considerable differences in CO₂ uptake within individual beech crown layers were determined.

Keywords

beech, cambial tissue, electric resistance, gas exchange

Introduction

The site conditions can be considered to be the determining complex of indices and factors from the point of view of quantity and quality of physiological processes in forest trees. Knowledge on time and spatial dynamic of individual characteristics within the whole forest ecosystem is necessary for quantification of impact of meteorological characteristics on photosynthesis and production processes, damage of foliage (frost, radiation, immissions), water and energy regime of tree species, etc.

This paper presents the results of ecophysiological research of European beech (*Fagus sylvatica* L.) – the tree species with the largest distribution in the Slovak forests. The main attention is paid to the impact study of site conditions on selected physiological processes (mainly photosynthetic activity and electric resistance of cambial tissue). This contribution is based on the results gained within the framework of ecological and ecophysiological research being realized in the research and demonstration object Pol'ana – Hukavský grúň.

Material and methods

Description of research plot

All measurements were done in the research and demonstration object (RDO) Pol'ana - Hukavský grúň which is a part of the Biosphere Reserve – Protected Landscape Area Pol'ana. The highest attention of research activity has been paid to the Permanent Research Plot – 0 (PRP 0) of which more detailed description, as well as the whole spectrum of problems being solved were published by ČABOUN et al. (1996). Basic geographic, pedological, meteorological and typological characteristics of the RDO and PRP 0 are presented in (Table 1).

Measurement of meteorological characteristics and ozone concentration

All meteorological characteristics within the PRP 0 were continuously measured and recorded to the data logger (DELTA T). The following meteorological characteristics were measured: air temperature (Ta) and rel-

Characteristics of RDO		Characteristics of PR	P – 0 (area 0.55 ha)	
Orographic unit	Pol'ana	Elevation	850–860 m	
District	Detva	Slope	5-15 %	
Forest Enterprise	Kriváň	Stand age	90-120	
Forest Administration	Poľana	Number of trees	342	
Elevation	820–915 m	Tree species	Beech	70.0 %
Exposure	North-east		Spruce	19.6 %
Slope	5-35 %		Sycamore maple	4.6 %
Geological substrate	Volcanic		Fir	3.5 %
Forest Management	Beech forests (411)		Ash	2.0 %
Type Group fertile	Fertile fir beech		Aspen	0.3 %
	Forests (511)	Canopy	90	
Number of PRP	8	Forest type	nitrophilous fir beech fores	t (5302)
		Soil type	Ando-eutric Cambisol	

Table 1. Selected characteristics of the RDO Poľana - Hukavský grúň and the PRP 0

ative air humidity (RH) (0.3 m, 6 m, 29, 34, 46 m above the soil surface), soil temperature (Ts) (depth 0, 5 and 10 cm), global radiation (Q) (34 m), photosynthetically active radiation (PhAR) (37 m, 32 m, 29 m, 19 m above soil surface), precipitation (34 m), wind direction (46.5 m), wind speed (46.5 m, 38.8 m, 37m). More detailed technical parameters of meteorological measurements were published by ČABOUN et al. (1996).

A daily dynamics of Ta, RH was determined for three height levels (34, 29, 6 m). The amount of incident PhAR upon the forest stands was calculated from the values of Q according to the relations presented by ROVŇÁKOVÁ (1986). The values obtained by this method were consequently quantified for individual height levels on the basis of direct measurements (cloudless days with typical daily course of PhAR).

In addition to meteorological characteristics, the concentrations of atmospheric ozone (O_3) were continuously measured above the stand. The analyzer ML 8810 (based on UV photometry) was used. The ozone concentrations were calculated according to Lambert–Bersch rule. More detailed methodology of atmospheric quality measurement in PRP 0 is presented by ČABOUN et al. (1996).

Determination of photosynthetic activity of beech foliage – ecophysiological measurements were realized on representative co-dominant beech (diameter $d_{1,3} = 49$ cm and height 35 m.). Its crown was divided into three separate crown layers (upper 32 m, central 29 m, lower 19 m from soil surface) on the basis of the previous measurements of leaf area, anatomical leaf structure, density of stomata, chlorophyll content (PRIWITZER et al., 1996), as well as the incident PhAR.

For determination of photosynthetic activity was used the portable photosynthetic system Li-6200 (Licor, Nebraska, USA). The detailed description of instrument and method of work with it was published by PRIWITZER (1993, 1998). All measurements were performed directly in the crown space using the tower. The measurements were usually performed from June to September during favourable weather (days without precipitation) on physiologically mature leaves (leaves in phenophase of adult leaf) and ambient CO_2 concentration (330–350 ppm). Daily dynamic of photosynthetic activity was determined by two methods as follows:

- Measurement of CO₂ exchange in hourly intervals for upper part of crown (sun type of leaves). More detailed methodology was published by PRIWITZER et al. (1996).
- Calculation using light response curves (relation of photosynthesis and PhAR intensity) determined on the basis of direct measurement, as well as conversed for individual crown layers according to the methodology presented by MAREK et al. (1989), MAREK et al. (1992) and PIROCHTOVÁ and MAREK (1991).

Measurement of electric resistance of cambial tissue – within the framework of observing the relative tree vitality was used the TREE VITALITY METER – TVM 01. It is a portable electronic instrument for determination of health condition of standing trees, and occasionally damage of wood mass, operating on principle of electronic measurement of cambial layer resistance and wood tissue respectively. Air temperature is being scanned by thermal sound. The 24-hour dynamic, as well as seasonal resistance dynamic were finding out. More detailed methodology of measurement of forest trees' cambial resistance was published by ČABOUN (1994, 1997). Measurement of tree species biofield – a magnitude of tree species biofield was measured in sense of the methodology published by ČABOUN (1993).

Results

As an example of beech ecophysiology we present the measurement performed on warm summer day with allday occurrence of cumuliform cloudiness.

Radiation regime – the curve of global radiation showed typical daily course with the values around 900 Wm⁻² between 10 a.m. and 2 p.m. (daily max. 950 Wm⁻² around the midday). Daily course of ozone concentration has slightly upward character with maximal values (58–63 ppb) between 2 p.m. and 4 p.m. At that time, the maximal values of air temperature were measured (Fig.1).

The daily dynamic of PhAR above the stand has a similar character as for global radiation. Changes in intensity and amount of PhAR in crown space are showed in the Fig 1. The individual daily course differs significantly in certain parts of crown and during the day. While PhAR in the upper part of crown showed very similar daily dynamic like above the crowns (max. $-1,397 \ \mu E \ m^{-2} \ s^{-1}$ around the midday), in central part of crown (29 m) the max. daily value of PhAR (1,085 $\ \mu E \ m^{-2} \ s^{-1}$) was recorded between 7 a.m. and 8 a.m. Fairly balanced intensity of PhAR characterized the lower part of the crown during the whole day. An increase of PhAR was recorded only in very short periods of time, while maximal daily values (520 µE m⁻² s⁻¹) were found out between 8 a.m. and 9 a.m. As regards a decrease of PhAR amount, due to its penetration through the crown (Fig. 1), it can be observed for selected part of the day, that 95% of PhAR from the amount measured above the crown was found out in the upper part of crown at 8 a.m., 61% at midday, and 39% at 6 p.m. Within the central crown layer the values of PhAR reached 50, 42 and 8% of values measured above the crown resp. 11%, 5% and 2% in the lowest part of crown. Similarly there was found out that in the upper part of crown 70% of PhAR values were in the interval of 500–200 μ E m⁻²s⁻¹, while in the lower layer 70% of values in the interval of $0-50 \ \mu\text{E m}^{-2} \text{ s}^{-1}$ during the day.

Temperature and humidity regime – daily course of air temperature and air humidity above the stand, as well as in the individual layers of the stand is shown in the Figure 2. Air temperature (Ta) in all height layers has gradually increased since early morning and it reached maximal daily values (from 18.7 to 20.4 °C)



Fig. 1. Daily course of global radiation (Q) and ozone (O_3) concentration above the forest stand (34 m) and PhAR measured in 37, 32, 29, 19 m above the forest floor.

at around 3 p.m. The highest values of Ta during the day were measured in the height of 34 m above the surface of soil. It is a consequence of quicker warming of air layer in the area immediately above the crowns and followed radiation of crowns' active surface within the infrared range of spectrum.

Air humidity (RH) was fairly constant (80%) during the period between 6 a.m. and 10 a.m. in the whole vertical profile of the stand. After that time we recorded its decrease while minimal daily values (48–57%) it reached in all height layers at around 3 p.m. The lowest RH values were measured in 37 m, between 10 a.m. and 6 p.m.

Photosynthetic activity of individual parts of the crown – daily dynamic of CO_2 (A_N) uptake determined for individual parts of the crown is shown in the Figure 3. When we look at the course of measured values (Fig. 3 – right), we find out the considerable differences in CO_2 uptake in individual crown layers.

While assimilation apparatus reached maximal daily values A_N (8.09 µmol CO_2 m⁻² s⁻¹) at around 11 a.m. in the upper part of the crown (sun type of leaves), in lower part of crowns (shade type of leaves) it was between 8 a.m. and 9 a.m. In the central crown layer (occurrence of both types of leaves), the maximal daily values A_N (4.37 µmol CO_2 m⁻² s⁻¹) were found out at around the midday. A considerable depression of CO_2

uptake was recorded mainly in the central (at around 2 p.m.) and lower (between 11 a.m. and 1 p.m.) part of the crown. After a decrease of CO_2 uptake we recorded more considerable increase of A_N value (crown centre at around 3 p.m., lower part at around 2 p.m.) in the both crown layers. Significant decrease of CO_2 uptake in all layers of the crown occurred after 4 p.m. When comparing the maximal values A_N we can find out that central part of the crown reaches 49% and in lower crown part only 28% of A_N of the upper crown part.

When assessing the daily dynamic of CO_2 uptake (sun leaves) obtained on the basis of direct measurement we recorded certain differences in comparison with simulated daily dynamic. While in direct measurement the daily maximum was between 6 a.m. and 8 a.m., and after that there occurred the consequent whole day decrease of values A_N , in case of simulated determination of A_N the daily maximum was between 10 a.m. and 11 a.m., and values of CO_2 uptake were fairly constant during the bigger part of the day (7 a.m.–3 p.m.).

Electric resistance of cambial tissue – daily course of electric resistance of cambial tissue in the investigated beech is given in the Figure 4. From the values measured in various cardinal points we can see that values of electric resistance of cambial tissue have similar daily dynamic, however, the values of resistance measured in northern direction are higher than in three other direc-





and measured in upper (32 m) part of beech crown.

tions. The increased resistance of tissues in the north side has been significantly expressed also in average values. The difference between average values of electric resistance of cambial tissue calculated from three and four measured values can be seen in the Figure 4. The highest dependence between electric resistance of cambial tissue and air temperature was discovered.

Biofield of beech – the daily dynamic of biofield's value has not been expressed. The following values of biofield: 56 cm, 47 cm, 38 cm, 29 cm, 20 cm and 11 cm from the stem of the tree were recorded. On the basis of measured values we can determine a regular 9-centimetre interval, it means a wave course of biophysical component of observed beech's biofield.

Discussion

The daily dynamic of physiological processes can, according to SCHULZE and HALL (1982), provide the basic information on responses and adaptation of plants to the nature conditions of the environment. At the same time it is a reflection of effect of outside factors on individual physiological processes, as well as it can provide a great number of information about behaviour of tree species in their natural environment. KOZLOWSKI et al. (1991) present that photosynthetic rate frequently varies between the tree species and their provenances, between the sun and shade type of leaves, during the day, as well as during the growing season. These variations are the result of interactions between the characteristics of plants such as leaf age, its structure and position, development of canopy, behaviour of stomata, amount and activity of Rubisco, as well as the factors of the environment such as light intensity, temperature, water supply, atmospheric CO, concentration, air pollutants and soil conditions. LARCHER (2003) presents that the gas exchange rate, investigated directly under the conditions of the forest stand, is a result of mutual effect of number of internal factors and factors of the environment of which the specific effect can be recognized in a very difficult way. From the whole group of factors, one of them usually limits photosynthetic rate, while others support it further. We can see from the daily dy-



Fig. 4. Daily course of electric resistance of cambial tissue measured from north, east west and south direction and average values from 3 direction (dotted) and 4 direction (hatched) (E,W,S, N).

namic of selected physiological processes a close connection between individual processes and meteorological characteristics.

The daily dynamic of photosynthetic activity of beech leaves stated by simulation is decisively being determined by light conditions in the individual parts of tree crown. The reason is the fact that intensity of PhAR was considered in advance to be the main factor influencing the CO_2 uptake. The influence of other factors has not been considered since the measurements of dependence of PhAR – A_N were done at leaf

temperature 20 °C and relative atmospheric humidity 60%. In addition, we can see from the daily dynamic of meteorological characteristics that Ta and RH have not reached the values which influence more considerably the CO₂ uptake in beech leaves (SCHULZE, 1970). As for the Ta, SCHULZE (1970) presented that max. values of A_N in mature beech leaves were measured at air temperature between 18–20 °C.

A favourable RH (80%), sufficient intensity of PhAR, as well as low ozone concentration can be considered a reason for the maximal daily values of A_N in

upper part of crown in the morning. The consequent whole-day decrease of CO_2 uptake, as well as the occurrence of considerable depression of photosynthesis between 11 a.m. and 3 p.m. could be caused by a high intensity of PhAR, reduction of RH, increase of leaf temperature (ČABOUN et al., 1996), and by an increase of ozone concentration during this part of the day. For instance MASAROVIČOVÁ and ŠTEFANČÍK (1990), XU and SHEN (1996) present the high intensity of PhAR, low RH and high Ta as causes of midday depression of photosynthesis.

The most well-known dependence of values of cambial tree tissue's electric resistance, frequently denoted as relative vitality, is a dependence on diameter of measured tree. The dependence of values of cambial tissue's electric resistance on their diameter, was found out in observing the daily or seasonal dynamic of electric resistance within all cases. On the basis of our current and previous research results (ČABOUN et al., 1993; ČABOUN, 1994, 1997) we can state that the less diameter tree species has, the higher annual variability of values of cambial tissue' electric resistance is. Thus, tree species with lower diameter react more sensitively on the environment's influences. Similarly, daily dynamic in trees with bigger diameter is not so marked than in trees with low diameter. Considerably it has been expressed in all cases and had an opposite course than atmospheric temperature. We have found out a considerable correlation in all our measurements between temperature and electric resistance of cambial tissue. The closest correlation has been found out between electric resistance of cambial tree tissue and average maximal temperature calculated from maximal temperatures of three days before measuring the resistance. From the above mentioned follows that the resistance of cambial tissue is influenced more by three-day, mainly maximal temperature, resp. by the weather (where temperature plays a dominant role especially in beech) than moment temperature during measurement. On the basis of previous results we can state that since the yearly or seasonal dynamic of electric resistance is considerably higher than daily dynamic, the date of measurement is essentially more important than day's time.

We have found out, within our long-term measurements in a great sample of tree species, very close correlation between a diameter of trees $d_{1,3}$ and biofield (ČABOUN, 1993; ČABOUN et al., 1996). We have also found out a close correlation between biofield and electric resistance since it correlates very closely with diameter of measured tree species as well. In spite of certain variability of a magnitude of tree species biofield, considerable trend of biofield change during the year has not been expressed. From the measurements we can see that correlation between the magnitude of biofield and temperature of atmosphere does not exist as well. The differences in magnitude of biofield during the year and also during the day, however, are not so high that we could speak about the seasonal or daily biofield dynamic but only about average values of biofield individual levels. However, besides diameter and kind of tree species also site conditions and intraecosystem relations have an influence on a magnitude of tree species biofield. On the basis of our previous observations we can state that biofield of trees is not the same for each measuring man. When we proceed with the definition of allelopathy (ČABOUN, 1990); each organism can react differently on biochemical and biophysical effect of other organism.

Acknowledgements

This work was supported by the Slovak Research and Development Agency under the contracts No. APVV-0608-10 and No. APVV-0111-10.

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Received December 6, 2012 Accepted April 8, 2013

Somatic embryogenesis: method for vegetative reproduction of conifers

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Abstract

SALAJ, T., FRÁTEROVÁ, L, CÁRACH, M., SALAJ, J. 2013. Somatic embryogenesis: method for vegetative reproduction of conifers. *Folia oecol.*, 40: 251–255.

For *Pinus nigra* Arn. somatic embryogenesis has been initiated from immature zygotic embryos enclosed in megagametophytes. The initiated embryogenic tissues contain bipolar structures – somatic embryos consisted of meristematic embryonal part and long vacuolised suspensor cells. The embryogenic tissues/ cultures are usually maintained on solid or liquid nutrient media. For long-term storage, recently the method of cryopreservation has been used to replace the time consuming regular transfers to nutrient media. The initiated cell lines represent individual genotypes and the structure of somatic embryos as well as their maturation is cell line dependent. The maturation of early somatic embryos occurs on media containing abscisic acid and osmotica. The process of somatic embryogenesis is completed by plantlet (somatic seedlings) regeneration.

Keywords

cryopreservation, in vitro, micropropagation, pine, somatic embryogenesis

Introduction

Somatic embryogenesis refers to the process in which somatic or non-sexual cells are induced to form bipolar embryos through a series of developmental steps similar in those occurring during *in vivo* embryogenesis (ST-ASOLLA et al., 2002). The initiated bipolar structures are capable of development producing cotyledonary stage somatic embryos that in appropriate conditions germinate and their development is completed by whole plants (somatic seedlings) regeneration. Owing to the fact through the developmental process of somatic embryogenesis large number of plants can be obtained in relatively short period of time, the method became an attractive tool for clonal propagation (KLIMASZEWSKA and CYR, 2002).

For conifers somatic embryogenesis has been initiated for different species belonging to genera *Picea*, *Pinus*, *Abies*, *Pseudotsuga* and plantlet (somatic seedlings) regeneration has been achieved, but some problems still remain. For Pinus species the relatively low initiation frequencies represent serious problem and recently efforts have been made to optimize the initiation process. For *Pinus radiata* HARGREAVES et al. (2009) obtained on average 55% initiation rates although the initiation was depending on families and collection time of explants. The initiation process can be enhanced through seed family screening, zygotic embryo staging as well as media adjustment (MONTALBÁN et al., 2012). The development of early bipolar somatic embryos present in embryogenic tissues can be stimulated by using abscisic acid (ABA) combined with non-penetrating osmoticum as polyethylene glycol (Svobodová et al., 1999; VOOKOVÁ and KORMUŤÁK, 2009) or carbohydrates maltose and sucrose (SALAJOVÁ et al., 1999; LELU-WAL-TER et al., 2008). The advantage of embryogenic tissues is their ability to regenerate after cryopreservation storage in liquid nitrogen at -196 °C.

In our laboratory, somatic embryogenesis for *Pi*nus nigra Arn. has been repeatedly initiated as well as plantlets/somatic seedlings regeneration has been achieved. The aim of presented paper is to evaluate somatic embryogenesis for the mentioned species.

Material and methods

Plant material

In our experiments megagametophytes containing immature embryos have been used as explants. The green cones of *Pinus nigra* Arn. have been collected at the beginning of June (usually between 1 and 15). The cones were stored at 4 °C for several days, after washed in tap water and the immature seeds were dissected. Surface sterilization of seeds was done by 10% H_2O_2 for 10 min. and then four washings in sterile distilled water followed. Finally, the megagametophytes were excised and placed on the culture medium.

Culture media

For the initiation as well as maturation of somatic embryos in most of cases DCR medium (GUPTA and DUR-ZAN, 1985) has been used. Other media as LV (LITVAY et al., 1981) or LP (QUOIRIN and LEPOIVRE, 1977) were also tested. The media were supplemented with enzymatic caseinhydrolysate (500 mg l-1), glutamine (50 mg 1⁻¹) as well as myo-inositol (200 mg 1⁻¹) and solidified with 0.3% gelrite (Ducheva). Plant growth regulators as 2,4-dichlorophenoxyacetic acid (2,4-D, 2 mg l⁻¹) and 6-benzyladenine (BA, 0.5 mg l⁻¹) have been incorporated into the nutrient media as well. Sucrose (2%) was used as carbohydrate source. Maturation occurred on DCR medium containing abscisic acid (25 mg l⁻¹) and 6-9% maltose or high concentration of Phytagel (1%). After appearing of cotyledonary somatic embryos the tissues were transferred to media without ABA. The germination medium contained activated charcoal (1%). The cultivation of explants occurred in dark at 23 °C (except the culture of regenerated somatic seedlings that were transferred to light (110 μ M s⁻¹/day).

Cryopreservation of embryogenic tissues

For cryopreservation of embryogenic tissues the slowfreezing method has been used. In the experiments altogether 46 cell lines were included. On the 8th day of growth cycle 3.0 g of tissues was re-suspended in 9 ml of DCR medium containing 180 g l⁻¹ sucrose. After 1 hour incubation in this medium gradually 15% DMSO was added to reach the final concentration 7.5%. Subsequently 1.8 ml of suspension was pipetted into cryovials and placed into the Mr. Frosty container. The samples were incubated in deep freezer (-80 °C) until the temperature in controlled cryovial reached -40 °C. Finally, the cryovials were plunged into liquid nitrogen and kept there for 1 hour to 1 year.

Thawing of tissues occurred at 40 °C in water bath. Following, the tissues were cultured on DCR medium as mentioned above in dark at 23 °C. Pretreated but not cryopreserved tissues were considered as control 1 (C1). Visual observations have been done in 3–4 days intervals. Growth analysis of tissues occurred three months after cryopreservation.

Microscopic observations

The structure of somatic embryos was investigated by light microscopy (Axioplan 2, Zeiss) using squash preparations and 2% acetocarmine staining.

Results and discussion

Initiation of embryogenic tissues

The production of embryogenic tissues has been observed approximately 3 to 5 weeks after placing the explants to the culture medium. The tissues were protruded from micropylar end of the megagametophyte (Fig. 1a), and reaching the size about 5 mm in diameter, they were separated from the explants and cultured individually as cell lines. The embryogenic tissues are of white color, translucent and relatively rapidly growing (Fig. 1b). Microscopic observations revealed the presence of bipolar structures - somatic embryos as the most important components of the tissues (Fig. 1c). The initiation frequencies reached values from 1.53% to 24.11% and changed from year to year. Plant growth regulation treatment as well as basal media formulations affected the initiation process. The highest initiation frequencies were obtained on medium DCR. The most important factor as we experienced is the developmental stage of zygotic embryos used as starting explants. For Pinus nigra the very early developmental stages - immature precotyledonary embryos gave the best initiation frequencies. As the maturation of zygotic embryos progressed the initiation frequencies dropped and finally the explants failed to produce embryogenic tissues. Although the bipolar structures are present in embryogenic tissues their structural organization is changing in dependence on the cell line. Relatively low initiation frequencies are characteristic for Pinus species and many attempts were done in order to achieve improvement. For Pinus nigra the developmental stage of original zygotic embryos is decisive and this phenomenon was also confirmed for Pinus radiata (HARGREAVES et al., 2009; MONTALBÁN et al., 2012), Pinus pinaster (MIGUEL et al., 2004), Pinus sylvestris (KEINONEN-METTÄLÄ et al., 1996).



Fig. 1a. Initiation of embryogenic tissues (ET) on megagametophyte explants (MG).



Fig. 1b. Proliferating embryogenic tissue 8 days after transfer to fresh medium.



Fig. 1c. Bipolar somatic embryos composed of meristematic embryonal cells (E) and long vacuolised suspensor (S).

Maturation of somatic embryos

The early bipolar structures are capable of development and plantlets (somatic seedlings) production. Transfer of tissues from proliferation medium containing 2, 4-D and BA to maturation medium with ABA and maltose resulted in the development of early somatic embryos. Approximately around the fifth week of culture on maturation medium numerous precotyledonary somatic embryos appeared on the surface of embryogenic tissues. The suspensor was still present and connected the developing embryos to the tissue. Cotyledonary somatic embryos appeared around the eighth week of maturation (Fig. 1d). Their quantity was strongly cell line dependent. For germination somatic embryos at least with four cotyledons were selected. The germination occurred on ABA free medium and resulted in somatic seedling formation (Fig. 1e).The obtained plantlets were transferred to soil and survived five to six months.

During somatic embryo development (maturation) structural changes were visible in the developing somatic embryos. The most conspicuous features were differentiation of root meristem and later the procambium formation. The maturation of conifer somatic embryos is genotype dependent (KEINONEN- METTÄLÄ et al., 1996; SALAJOVÁ et. al., 1999) and is influenced by composition of the maturation medium (CARNEROS et al., 2009; MONTALBÁN et al., 2010).



Fig. 1d. Cotyledonary somatic embryos (arrows) developed on the maturation medium.



Fig. 1e. Plantlets (somatic seedlings) regenerated from somatic embryos.

Cryopreservation of embryogenic tissues

Cryopreservation of embryogenic tissues enables their storage for long-term period. Out of 46 cell lines cryopreserved 35 survived the storage in liquid nitrogen. The regeneration of tissues after thawing (Fig. 1f) was dependent on cell line although the duration of storage in liquid nitrogen had no significant effect on the growth and behaviour of tissues. Our examinations also showed no correlation exists between the maturation ability and cryotolerance of cell lines. The use of cryopreservation method for long-term storage of conifer embryogenic tissues has been demonstrated for several species (ARONEN et al., 1999; VONDRÁKOVÁ et al., 2010).



Fig. 1f. Re-growth of embryogenic tissue (ET) after cryopreservation.

The obtained results for *Pinus nigra* give evidence that it is possible to initiate embryogenic cell lines from immature zygotic embryos, but to obtain higher initiation frequencies the method/approach needs refinement. Although the maturation of somatic embryos is cell line dependent, cotyledonary somatic embryos were produced and somatic seedling regeneration occurred as well. Cryopreservation of embryogenic tissues using slow-freezing was also successful for the majority of tested cell lines.

Acknowledgement

The study was supported by the Slovak Grant Agency VEGA, proj. No. 2/0144/11.

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Received December 6, 2012 Accepted April 15, 2013

Selection and breeding of stress-tolerant woody ornamentals for urban plantings

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Abstract

SCHMIDT, G. 2013. Selection and breeding of stress-tolerant woody ornamentals for urban plantings. *Folia oecol.*, 40: 256–260.

Because of this geographic position, climate and soils, Hungary lends itself for selection of woody plants which tolerate environmental stresses. Selection and breeding of the woody ornamentals for extreme urban conditions started in the early 1950s at the former University of Horticulture and Food Industry (at present: the Faculty of Horticulture of Corvinus University), Budapest. The first results were 8 *Sorbus*, 3 *Tilia* and 2 other cultivars, and selected clones from *Fraxinus, Cornus, Juniperus* and others. In the recent 20 years, many new hardy cultivars and named clones are brought up, the most important of which are as follows: *Ailanthus altissima* (Mill.) Swingle cv. Purple Dragon, *Acer campestre* L. cv. Zentai Upright`, *Celtis occidentalis* L. cv. Straight Stem, *Crataegus pinnatifida* Bunge cv. Tahi, *Hedera helix* L. 9 cultivars, *Platanus × hispanica* Münchh. cv. Budapest, *Prunus padus* L. cv. Aurora, *Prunus × davidopersica* cv. Rubin (*P*. L. cv. Piroschka), *Prunus tenella* Batsch. cv. Pink Carpet, *Pyrus nivalis* Jascq. cv. Kartália, *Salix matsudana* KOIDZ. cv. Golden Spiral, *Syringa josikaea* J. Jacq. ex Rchb. cv. Smaragd, *Tilia tomentosa* MOENCH. cv. Zenta Silver, *Tilia × euchlora* K. Koch. cv. Saint Stephan.

Keywords

Ailanthus, new cultivars, Prunus, stress-tolerance, Tilia, urban trees

Introduction

Because of this geographic position, Hungary lends itself for selection of woody plants which tolerate environmental stresses: the summer is warm with temperatures reaching a maximum of 30-35 °C, and the winter is cold and irregular with temperatures falling sometimes -26-30 °C. These extremities are multiplied by the poor sandy and salinity soils of the Great Hungarian Plains, the dry limestones and dolomites of low hills and by the dry warm and polluted atmosphere of cities and towns. Selection of woody ornamentals for such conditions started in the early 1950s by the Department of Horticulture and Dendrology of the former University of Horticulture and Food Industry (at present: the Faculty of Horticulture of Corvinus University, Budapest). The first results were 8 *Sorbus*, 3 *Tilia* and 2 other cultivars, and selected clones from *Fraxinus, Cornus*, *Juniperus* and others (SIPOS, 1964; READ and SCHMIDT, 1987).

In the recent 20 years, many new hardy cultivars and named clones are brought up (LUKÁCS et al., 2009; ORLÓCI et al., 2009; SCHMIDT and SÜTÖRINÉ, 2011).

The aim of the present paper is to give a short description of (and experiences with) the new selections of this period.

Material and methods

The breeding work was carried out in two basic ways: 1. on-site selection of adult specimens in urban or roadside plantings; 2. selection of young seedlings from mass-propagation in the nursery. In the case of on-site selection of adults specimens in urban or roadside plantings, expeditions were organized to the larger cities and towns for the search of such adult specimens which in the given urban environment showed higher tolerance towards urban injuries than the other ones (their neighbours standing in the same alley or group) and also showed some increased ornamental or technological value, like better crown shape, shiny and healthy or colourful leaves, good structure of branch system, etc.

In the case of selection of young seedlings from mass-propagation in the nursery, this sort of breeding started with mass-propagation by seed of such species or individuals which were known as genetically stresstolerant in Hungary (and having sometimes also additional values like red leaves, or disease – resistance etc.) Later from the seedling lot (minimum 400 liners, but usually much more) those were picked out, which showed better qualities than the other ones.

In both cases, at the first step minimum 10-12 specimens were selected (picked out) and propagated vegetatively (by budding or by cuttings) in an amount of minimum 50 cuttings or grafts per clone. The second step was the nursery trial of the clones (speed of growth, healthiness of leaves, straightness of stem of trees or bushiness of shrubs) and, of course, repeated bonitations were made on their ornamental value. The third step was to plant and try them out in urban conditions, usually in Budapest. Finally, those clones which proved to be the best both in the nursery and in urban plantings were propagated again in the nursery. They were given cultivar names and submitted to official cultivar - recognition to the respective Institution and Community for testing and approval (see later at chapter New cultivars for urban planting.)

Results

Description of the recent cultivars and selections

New cultivars in Hungary are inspected and tried for several years by the National Institute for Agricultural Quality Control. If they meet the necessary criteria, including the DUS-requirements, an official recognition and certificate is awarded by the Hungarian Cultivar Qualification Council (HCQC).

In the recent 20 years, many new hardy cultivars and named clones are brought up and recognized by HCQC, the most important of which are as follows:

Ailanthus altissima (Mill.) Swingle cv. Purple Dragon. The heaven tree, *Ailanthus altissima* (Mill.) Swingle, tolerates drought and bad soils extremely well in Hungary and grows like weed in polluted urban environment. The cultivar Purple Dragon is a female form found in Budapest. It has a straight leader, fast growth and a regular rounded crown becoming flattened with aging. The dark purple winged fruits are born in abundant clusters and retain their intensive colour from July through August and early September. Foliage is shiny green with red petioles and leaflet nerves, the shoots are purplish brown. (Breeder: G. Schmidt, 1996.)

Celtis occidentalis L. cv. Straight Stem. The Hackberry Tree Celtis occidentalis L. is perhaps the hardiest urban tree in Hungary, which equally tolerates the poor urban soils, polluted atmosphere and also the negative effects of human vandalism like heavy injuries of the trunk, cutting the branches and the roots, etc. The only (but great) disadvantage of the traditionally used type is the irregular growth (curved trunk) and the overhanging branches which create problems both in the nursery and in the street plantings. The new cultivar Straight Stem has the high tolerance of the traditional species without its disadvantage in habit and growth: The stem of this cultivar is growing straight (and fast) in the nursery so (in contrast to the "traditional" type) it does not need staking (Fig. 1); later (on the final place) along urban streets. The crown becomes upright oval. The branches are not overganging at all, so they do not disturb the traffic. (Breeder: G. Schmidt 2006.)



Fig. 1. Celtis occidentalis L. cv. Straight Stem.

Hedera helix L. Hungarian cultivars. The English Ivy *Hedera helix* L. is native to practically all woodlands in Hungary (BÉNYEI-HIMMER, 1994a). It is a multifunctional plant in landscaping. The juvenile form

is suitable for a groundcover (both in shadow and on the sunshine), for covering walls of the buildings and fences and climbs easily on pergolas or on trunk of trees as well. The adult form makes a fine and hardy rounded bush, or, if pruned, can be planted as a low evergreen semi-low hedge.

The cultivars of the Faculty of Horticultural Sciences originate from different parts of Hungary and fall into two groups: 1. the spreading climbing (juvenile) cultivars, and 2. the bushy (adult) ivies. They were bred by Mrs Bényei-Himmer M. at the Department of Botany (BÉNYEI-HIMMER, 1994b).

Spreading - climbing (juvenile) cultivars are:

H. helix L. cv. Börzsöny. A fast spreading cultivar, with thick growth. Leaves are elongated triangular in form (f. *sagittifolia*), the leaf blade leader them, dark green with well marked nerves (Fig. 2). Makes an excellent groundcover. (Breeder: Bényei-Himmer M. 2000).



Fig. 2. Hedera helix L. cv. Börzsöny.

H. helix L. cv. Zebegény. Middle-strong growth, very good tendency for branching. Leaves are markedly five-lobed (f. *pedata*) with vivid green colour and silvery leaf-nerves. Suitable as a ground-cover (especially in small gardens) or for balcony-boxes. (Breeder: Bényei-Himmer M. 2000).

H. helix L. cv. Krokó. A slow to medium-strong growing spreading form, with slightly lobed and widely silvery-nerved leaves which give the plant a spectacular ("crocodile-like" appearance, Fig. 3). Excellent as a grand cover for small gardens or in balcony-boxes. (Breeder: Bényei-Himmer M., G. Botlik 2004).

H. helix L. cv. Negro. A medium-strong growing spreading form, whose leaves are very dark green (almost black). An interesting ground cover, makes a good contrast if planted in one group with silvery or with golden-coloured cultivars. (Breeder: Bényei-Himmer M. 2004).



Fig. 3. Hedera helix L. cv. Krokó.

Bushy (adult) cultivars are:

H. helix L. cv. Soroksár. A wide-rounded low or medium-sized bush, with vivid green shiny leaves. Black fruits from February till the end of March. (Breeder: Bényei-Himmer M. 2000).

H. helix L. cv. Blue Star. A medium-sized bush, with shape and size like that of the former cultivar, but the fruits are shiny blue, appearing in abundant loose cymes. (Breeder: Bényei-Himmer M. 2000).

H. helix L. cv. Marble. An upright bush of smaller (later medium) size. Dark green leaves with undulate margins and marked light-green veins are giving a marbled effect. It has abundance of yellowish flowers in September–October and black fruits (in compact cymes) during winter. (Breeder: Bényei-Himmer M. 2000).

H. helix L. cv. Csocsoszan. A rounded bush whose leaves are not lanceolate but wide obovate with crenate leaf-margins resembling a Japanese fan. Not so winter-hardy as the former cultivars. (Breeder: Bényei-Himmer M. 2004).

Prunus \times **davidiopersica** cv. Rubin (P. \times d. cv. **Piroschka**). A small tree with flattened crown. Ultimate height is 4–5 m, diameter 6–8 m. Leaves are dark ruby-red during the intensive shoot growth and are turning dull greenish red when the growth stops. This change of

colour (with new and new flushes of growth) is repeating 2–3 times in one vegetation. Large white flowers with a small pink eye bloom in late March–early April immediately after bud-brake. The cultivars are resistant to mildew and to *Taphrina deformans* (BERK.) TUL. Tolerant to drought and early frost. (Breeders: G. Schmidt and F. Incze. 1996).

Prunus padus L. cv. Aurora. The "Bird Cherry" Prunus padus L. is a medium-sized bushy tree widely distributed on the Northern Hemisphere including Hungary. The first red-leaved Prunus padus cultivar Coloratus was introduced to Hungary some 30 years ago. This cultivar did not distribute in Hungary because of its poor tolerance to continental climate and the limy soils (the cultivar was selected in Sweden, under the humid climate of Scandinavia). The Hungarian cultivar Aurora is developing leaves which are much darker red under our conditions, than those of cv. Coloratus and, in contrast to the mentioned Scandinavian cultivar do not burn neither become necrotic (yellow) in the hot summer. It brings abundance of dark pink flowers (Fig. 4) blooming in upright panicles during mid- or late April. (Breeder: G. Schmidt, 2005, further selected clones still in process of trials).



Fig. 4. Prunus padus L. cv. Aurora.

Pyrus nivalis Jacq. cv. Kartália. A 4–6 m high, slow growing small tree with wide columnar form. Side shoots are short, squat and grow horizontally when

young. Later the side-shoots turn upright and are only slightly thorny. Flowers are white, blooms 2 weeks later than *Pyrus communis* L. The fruit is a 3 cm wide, yellowish green pear. No pests. Suitable in parks, small streets. Drought tolerant. (Breeders: I. Tóth, and A. Terpó 1995).

Salix matsudana Koidz. cv. Golden Spiral. A fast growing corkscrew-willow, probably a spontaneous hybrid between *S. matsudana* Koidz. '*Tortuosa*' and *S. alba* L. '*Tristis*'. It was found as a chance seedling near Velencei lake. Shoots, twigs and branches are much contorted, light yellow in the summer turning rich golden orange in the winter (Fig. 5) (Breeder: G. Schmidt. 1993).



Fig. 5. Salix matsudana Koidz. cv. Golden Spiral.

Syringa josikaea J. Jacq. ex Rchb. cv. Smaragd. A 2–3 m high, strong growing, compact shrub with stiff, upright branches. Leaves are 6–10 cm wide, elliptic, dark emerald green, leathery through the whole summer. Dark lilac-coloured flowers, bloom in upright compact panicles, in mid- or late May (2 weeks after the common lilac (*S. vulgaris* L. cultivars). Cv. Smaragd is not susceptible to mites, and tolerates half-shade. Use: alone or in groups in parks and in home gardens. (Breeder: G. Schmidt. 1993). *Tilia tomentosa* Moench. cv. Zenta Silver. A strongly upright-growing tree, making a straight leader in the nursery and reaching an ultimate height of 15–20 m in parks. The crown is wide columnar when young, becoming compact conical with aging. Branches are upright. Shoots are greyish green, downy. New leaves are light dull green above, strongly silvery tomentose beneath. Highly scented flowers bloom in late June-early July. Grows fast in the nursery and tolerates urban climate well. Suitable for streets and parks. (Breeders: B. Nagy and G. Schmidt. 1996).

Tilia × *euchlora* K. Koch. cv. Saint Stephan. A strongly growing tree, making a straight stem and continuous leader in the nursery and reaching an ultimate height of 15–20 m. Crown is narrow-ovate, with pointed top (Fig. 6). Branches are upright. Shoots are brownish-green, glabrous. Leaves are leathery, shiny green above, dull green and glabrous beneath. Slightly fragrant flowers bloom in early July and almost no fruits later. Tolerates urban climate well. (Breeders: E. Jámbor-Benczúr, Z. Ifjú., I. Tóth and G. Schmidt 2000).



Fig. 6. Tilia × euchlora K. Koch. cv. Saint Stephan.

Conclusion

In the recent 20 years 11 new urban trees and shrubs have been bred at the Department of Floriculture and Dendrology and 8 *Hedera helix* L. cultivars at the Department of Botany of the Faculty of Horticulture, Corvinus University, Budapest. All of them are available in the leading Hungarian tree nurseries, registered at the Central Agricultural Office and also are kept records by the civil organization "Commission for Hungarian Ornamental Cultivars". The home page of this organization (http://www.magyarfajtak.hu) contains a more detailed description of the above-listed cultivars too, illustrated by digital photos.

Acknowledgement

The Author would like to express his thanks to the organizers of the excellent Conference in the Mlyňany Arboretum (Sept. 18–19., 2012) and also to Stephan Bakay for completing the Slovakian abstract of this paper.

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Received December 6, 2012 Accepted April 8, 2013

Influence of vegetation on microclimate in the urban environment

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Abstract

STRELKOVÁ, M., HEČKOVÁ, Z., RÓZOVÁ, Z., TIRPÁKOVÁ, A., MARKECHOVÁ, D. 2013. Influence of vegetation on microclimate in the urban environment. *Folia oecol.*, 40: 261–265.

There are many factors in the urban environment influencing its microclimate conditions. Vegetation is one of the main components participating in this process. Our study compares microclimatic factors (the air temperature and the air humidity) of two sites with different ratio between built-up area and greenery. The measurements have been realized in the chosen areas of the built-up area of Nitra town in the spring months of 2012. The aim of the research was to compare the air temperature and the air humidity depending on the percentage of the vegetation cover in the urban environment of the built-up area of Nitra town.

Keywords

air humidity, air temperature, microclimate, vegetation

Introduction

Greenery in the urban environment is irreplaceable. The cities are being characterized by the presence of different surfaces changing the microclimatic characteristics of the environment. The presence of different surfaces is particularly reflected in the influence of the air temperature and the relative air humidity conditions of the human environment.

Greenery is a major component of the nature being involved in the treatment of this condition in the cities. It is characterized by several functions that make us a great deal in such disturbed environment as the urban settlements are. The microclimatic function of the greenery is the basic one. Microclimatic function consists of cooling of the urban environment by greenery during the warm months and avoiding of the large temperature fluctuations during the day and night. All the trees and bushes regulate the air humidity of the atmosphere. Their space capacity and biomass assimilation adapt the climate, air temperature, solar radiation and air flow (SUPUKA et al., 1991; Rózová and MIKULOVÁ, 2009). The microclimatic function is being understood as the ability of greenery to influence the air humidity, shade provision, reduction of temperature fluctuations, etc., by its transpiration function. For example, the adult birch in growing season can evaporate up to 7,000 l of water, the urban parks reduce the air temperature by an average of 1 °C if compared to the temperature in the streets. The green areas increase the air humidity (the average value is 5 to 7 per cent) (HUDEKOVÁ et al., 2007).

The aim of this work was to evaluate the development of the microclimatic characteristics of the areas with different ratio of built-up area and greenery in the spring of the year 2012. The relative air humidity in % and the air temperature in °C, the main indicators of the microclimatic conditions, have been the measured characteristics.

Material and methods

The research was carried out in the selected parts of Nitra town. There were selected two model areas: Site

1/Chrenová III. – parking place on Akademická Street, area with 0–50% of greenery and Site 2/Chrenová I. – place behind the Student Dormitory Nitra, area with 51–100% of greenery.

The ratio of the built-up areas and the green areas on the same size surface has been the main criteria of the selection process. The squares of 50×50 m have been determined within the area. Chosen squares had to include parts with different types of cover (hard surface, grassland and vegetation section). There were created two categories, 0–50% and 51–100% of greenery of the whole area. The research was carried out during the spring months (March, April and May) in 2012. Anemometer with multifunctional climate probe, VELOCICALC ((9565)) – TSI, has been used for the measurements. The relative air humidity in % and the air temperature in °C were measured simultaneously.

Data logger recorded data at weekly intervals (7 days), beginning on the second Monday of month at 7 a.m., 2 p.m. and 9 p.m. with the periodic repetition. The measurements have been realized in both localities in the middle of the selected areas with the vegetation, grass and hard surface and at the contact interfaces of these parts: grass – vegetation, vegetation – hard surface, hard surface – grass and at a distance of 2 meters from the building. The measurements have been performed at 2 m above the ground level.

Twenty entries were recorded at the specific time in each area for the purpose of the statistic evaluation. Collected data have been processed to the tables in Excel 2010 program and evaluated by two-factor analysis of variance by Statistical Program.

Results and discussion

The measurements were realized in spring during the months of March, April and May in 2012. Change in the development of the air temperature was observed in the selected months in the monitored areas. We assumed that the air temperature would be higher in the areas with the higher ratio of the built-up area to the greenery. Such assumption was confirmed in March (Fig. 1). The air temperature during the monitored week was higher at Site 2 than at Site 1 with the higher ratio of the greenery coverage. If sub-sites were considered, the biggest differences in temperature were recorded at sub-site with the hard surface and the smallest differences were at the contact interface of the grass-vegetation.

In April (Fig. 2) the air temperatures became the equal at both sites. A significant difference is visible at the sub-sites with the grass coverage. The minimal differences were observed at the sub-sites with the hard surface and the contact areas with grass and vegetation.

The measurements in May (Fig. 3) showed differences in the air temperature values at the selected sites. The greatest difference in temperature was observed at the sub-site grass – vegetation. The higher temperatures in May were measured at Site 1, in March at Site 2. Our assumption that the air temperature is higher at site with



Fig. 1. Comparison of mean air temperatures measured during one week at sub-sites of sites 1 and 2 in March 2012. Site*Sub-site; LS Means. Current effect: F (6.280) = 0.01259, p = 0.99999. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.



Fig. 2. Comparison of mean air temperatures measured during one week at sub-sites of sites 1 and 2 in April 2012. Site*Sub-site; LS Means. Current effect: F (6.266) = 0.01815, p = 0.99997. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.

the higher ratio of the built-up area to the countryside has not been confirmed. This situation might be caused by the consequence of the fact that the measurements were taken at the beginning of the growing season of the trees. The vegetation in our latitude is without leaves in March. The leaves start to grow during April and the full foliage of trees occurs in May. The growing season of trees in Slovakia lasts from April 1 to September 30.

The relative air humidity and the air temperature were measured at the same time and at the same places. The research was performed in spring of 2012. The higher values of the air humidity were measured in all



Fig. 3. Comparison of mean air temperatures measured during one week at sub-sites of Sites 1 and 2 in May 2012. Site*Sub-site; LS Means. Current effect: F (6.280) = 0.00496, p = 1.0000. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.

months at Site 1, where the greenery dominates over the built-up area.

was the biggest difference in values of the air humidity between the monitored sites. The difference in the following month, in April, was smaller one. In May, the values of the humidity became equal at sites 1 and 2.

Figures 4–6 show the changes in the air humidity at the sub-site with the hard surface. In March, there



Fig. 4. Comparison of values of mean relative air humidity measured during one week at sub-sites of Sites 1 and 2 in March 2012. Site*Sub-site; LS Means. Current effect: F (6.280) = 0.03431, p = 0.99983. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.



Fig. 5. Comparison of values of mean relative air humidity measured during one week at sub-sites of Sites 1 and 2 in April 2012. Site*Sub-site; LS Means. Current effect: F (6.266) = 0.00456, p = 1.0000. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.


Fig. 6. Comparison of values of mean relative air humidity measured during one week at sub-sites of Sites 1 and 2 in May 2012. Site*Sub-site; LS Means. Current effect: F (6.280) = 0.00243, p = 1.0000. Effect hypothesis decomposition. Vertical bars denote 0.95 confidence intervals.

This situation is caused by the equalizing of the temperature daily routine. The air temperature is higher and its fluctuations are reduced during the day. We did not take into consideration the precipitation when evaluating data and results. In the future, the precipitation regime of Nitra town needs to be considered in comparison with the microclimatic characteristics.

Acknowledgements

This study is the result of the project implementation: Environmental Aspects of the Urbanized Environment, ITMS: 26220220110, supported by the Research & Development Operational Programme funded by the ERDF, scientific projects: Slovak Ministry of Education, project VEGA No. 1/0042/12 and Constantine the Philosopher University in Nitra, project UGA No. VII/35/2012.

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Received December 6, 2012 Accepted March 8, 2013

Development, changes and assessment of tree alleys in town streets

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Abstract

SUPUKA, J. 2013. Development, changes and assessment of tree alleys in town streets. Folia oecol., 40: 266-271

Street tree alleys were started to plant as a part of great reconstruction of European and world cities after industrial revolution from the beginning of 19th century. Important participation at first street alleys plantation have had decoration associations organised at town's of Slovakia territory on 2nd half of 19th century. Street tree alleys and river embankments have wide spectrum of functions useful for man and improving of environmental conditions. In a new terminology we are talking on services those should be classified as follow: supporting, provisioning, regulating and cultural services. Street trees have high value at designing of the pedestrian zones and new squares. In Nitra town conditions we have assessed 26 most important street tree alleys. For this purpose was elaborated a new methodical approach, where besides basic biometrical dates the following tree characteristics were assessed: potential of environmental adaptability, potential of biology-ecological value, potential of culture value, potential of disturbing and undesirable influences. Each group of characteristics was valuated in three following levels of significance (1) low, (2) medium, (3) high. There were assessed 22 woody plant species and cultivars mapped at Nitra town streets.

Key words

street trees, changes, Nitra town, service assessment

Introduction

The street free alleys represent important component in vegetation structure of town settlements (SUPUKA et al., 2008). Tree alleys were in the past used as a component part of designed landscape, planted especially along with roads and as internal composition element of historical gardens and parks or prolongation element from those parks into open country.

Street alleys as town design element were mainly established after industrial revolution during urban reconstruction of larger cities for instance Paris, where spacious street boulevards and alleys were constructed in time after beating up Bastila in 1779. Since that time establishment of new public parks has been dated (KALUSOK, 2004; KUPKA, 2010). Moreover, such reconstructions were realised at other European cities also including Bratislava which was succeeded by creation of new esplanades, reconstruction of former market squares to the public spaces. Since period of the 2nd half of 18th century, establishment the first public park – Aupark has been dated in central European countries in Bratislava (in 1775, nowadays as Sad Janka Kráľa in Petržalka). There are registered plantations of street trees and alleys such as locust-tree, linden, maple, and plane. At tree plantings was importantly participated city's decorative association (TOMAŠKO, 1967). Activities related to Nitra town reconstruction,

mainly after revolution during 1948–1949 years, were aimed to the construction of new buildings of state and public governance, theatre and other objects of culture, recreation, amusement and military services (barracks). The new streets were paved, installed public lighting and planted trees at streets and squares under active participation of Nitra decorative association, which was established in 1888s by 123 founding members (FUSEK and ZEMENE, 1998). Activity of decorative associations passed over progressive development in Slovakia and at present time, the care of cities green areas including street trees passed under leadership of the city authority

profession departments. Street tree alleys, at river embankments and bank side roads have wide spectrum of functions, those in a new terminology are classified as services for man and improving of environmental conditions. Basically they might be divided into four groups as are supporting, provisioning, regulating and cultural services (REYERS et al., 2009; YONG, 2010). Particular classification and quantification of functional importance and effectiveness of green spaces including street trees have been published by SUPUKA et al. (1991). Accent is given on environmental quality improving (climate, physical, chemical, micro-organism air quality, and water regime), ecological aspects of urban spaces (biodiversity, ecological stability, ecological and green nets), social and cultural ones (trees as urban design elements, aesthetic, culture, education, recreation effect). Moreover most actual research articles at international level have presented functional and service tree's values in urban environment. Green spaces and trees in USA cities have absorbed annually 711 thousand tons of allochtonous components on average, especially glasshouse gases such as ozone, carbon, nitrogen and sulphur oxides (Nowak et al., 2006). Climate improving like shade, refrigeration, air humidity up to 10% effectiveness was published in other article (ARMSON et al., 2012; Томаšко, 1996). The negative influences of woody plants in the relation to human body where a lot of woody species have caused allergy sickness were published as well (ZLÍNSKA, 1996). Assessment of the landscape gardening values and healthy conditions of poplar-trees in Nitra streets and green spaces has been published by VEREŠOVÁ (1999), tree alleys of 9 streets of Nitra town were evaluated from the phytopatology point of view (TKÁČOVÁ, 2003). The cities are characterised as spaces with expressive environmental changes, which limits selection of trees for street alleys. For this reason a lot of authors and research projects have aimed in issue solution and definition of criterions on advisable tree selection for urban environment (QUIGLEY, 2004; SAEBO et al., 2005; SUPUKA, 2005; VREŠTIAK, 1994 and others).

The aim of the paper is to assess framework development in establishment and changes of streets in Nitra town, to evaluate current state of selected street spaces from point of species composition, ecological importance, environmental adaptability and cultural-aesthetic values. The specific street's tree assessment method was defined and applied in Nitra town conditions.

Material and methods

Historical development of street tree alleys establishment in relation to urban structure reconstruction at Nitra town was assessed by using of published historical documents (FUSEK and ZEMENE, 1998) and as a result of own field study. Current state of selected 26 most interested street alleys were assessed complexly by using of published particular criterions (BENČAŤ, 1982; MACHO-VEC et al., 2000; JUHÁSOVÁ et al., 1991; SUPUKA, 2005; VEREŠOVÁ, 1999; VREŠTIAK, 1994; ZLÍNSKA, 1996). Throughout syntheses particular marks and characteristics we have defined following categories and assessing criteria for street trees alleys in Nitra town:

a) Characteristic dates

- aa) Locality, street, square
- ab) Name of woody plant and cultivar
- ac) Average tree alley high
- ad) Average tree alley age
- ae) Average landscape gardening value
- b) Potential of environmental adaptability to
 - ba) Urban soils
 - bb) Urban climate
 - bc) Soil salinity
 - bd) Complex of immission impact
- c) Potential of biology-ecological values
 - ca) Potential for ecological networks and ways
 - cb) Topical and trophycal potential for biodiversity support
 - cc) Potential of phytoncidy activity and microbiology regulation
 - cd) Potential of pest and disease impact
 - ce) Potential of tree pruning tolerance and regeneration ability
- d) Potential of culture values
 - da) Potential of aesthetic effect and perception
 - db) Potential of species rareness and gene-pool value
- e) Potential of negative influences
 - ea) Potential of phyto-allergy
 - eb) Potential of invasive demonstration
 - ec) Potential of litter fall contamination.

Each potential within particular categories might be valuated in three degrees: (1) low, (2) medium, (3) high.

Results and discussion

The graphic vedutes from 1st half of 19 century shows Nitra as romantic town surrounded by gardens with domain of sacral and governance buildings and churches. To the town leaded road bridged over Nitrička (second branch of the Nitra river to the south of castle hill), lined by both side tree alleys. In that time street trees were rather ambition that reality, the trees were most often as river bank side tree line, in domain of poplars, willows, ashes and alders. Effective planting of street tree alleys and park areas was started after revolution years 1848– 1849s, which was closely associated with reconstruction of urban structure of settlements. The contribution is coursed only at those new buildings and reconstruction which relates to new tree planting. In 1870–1873s there were made terraced modifications of the square in front of Piaristic church and placed 12 apostles' sculptures and throughout circumference were 100 pieces of horse chestnuts tree planted, which are found there at present time. In period of 1878–1885, the 14 chapels of cross-route were built-up at Mariansky hill associated with linden and horse chestnuts tree plantings. Construction of region theatre on contemporary Svätopluk square in 1885 was accompanied by tree planting of *Acer platanoides* 'Globosum' at square circumstance. In 1905 was reconstructed the New Elisabeth road from town barracks to gasworks, 8 m in width, paved surface and tree alleys planted on both sides. Road from Nitra's capitol till Taufel garden at Chmelova valley, constructed in 1888 was lined by tree alleys plantings.

After the Nitra river regulation on the turn out of 19th and 20th century there was established city park Sihot' and embankment of new canal was planted up by tree alleys (linden, horse chestnut, and poplar). Tree alley was planted along with road from down town to rail way station, at Palanok street under castle were planted coniferous trees. High merit at tree alley planting has had Nitra's decorative association established in 1888 at Chmelova valley town zone that was renamed to Zobor's decorative association in 1889 and on 24 October 1897 to Town's decorative association (first chair MUDr. K. Tarnóci). At tree alley plantations, parks establishment and a forestation of Mariansky and Šibeničny hill there was very active forester Marcel Boroš. In 1917 association has expired because of the 1st world war. The historical documents and photos of Nitra town from period of 1920ies show tree alleys from the Kralik restaurant as far as theatre. The new dwelling zones of Nitra town have been built up after 2nd world war and parallely there were planted street alleys and new park was established. In sixties of the 20th century there was appointed town's enterprise of gardening and technical service, which has had solicitude at street trees and green areas. Since 1990 all activities related to city greenery have been under profession auspices of city authority Department of main architect, section of urbanism and architecture. Reconstruction, new plantings and maintenance of street trees, park trees and green areas are served by profession private enterprises.

Establishment of street tree alleys is directly associated to urban development of the town. Constitution of the 1st Czechoslovak republic creates extensive opportunity for social-economy development of new state human society including Nitra town. There were established new dwelling zones such as Číneš, under Calvary, Post colony, under Zobor. In the new built up streets were planted new streets alleys, especially linden, maple, ash, locust-tree, in natural tree shape or as cultivars mostly in globes or column ones. After the 2nd world war additional dwelling town zones were built up, e.g. Chrenová, Párovce, Čermáň, Klokočina, Diely, Zobor (further intensification). Those zones were established on area of original villages or on new areas at former agriculture land and vineyards. Along with transport lines, small squares and public spaces were planted new tree alleys. Besides traditional trees, a new woody species and cultivars were planted, such as Acer platanoides 'Globosa', Acer dasycarpum, Carpinus betulus 'Columnare', Fraxinus excelsior 'Globosa', Prunus cerasifera 'Atropurpurea', P.c. 'Nigra', Ribinia pseudoacacia 'Umbraculifera', R. p. 'Bessoniana', and others. To the newest planted woody species and cultivars in the streets belongs e.g. Acer platanoides 'Columnare', Koelreuteria panniculata, Gingko biloba, Liquidambar styraciflua, Prunus fruticosa 'Globosa', Sorbus aria 'Lutescens' and others. Those new tree forms and cultivars were successively planted to alleys during reconstruction of Svätoplukovo square and Štefánikova street at present time as pedestrian zone. Those two public spaces have representative and culture aesthetic dominant functions therefore they are designed in high architectonical level with applying of modern trees in alleys. New woody plant species and attractive cultivars were also planted as a part of reconstruction of old tree alleys or within process of humanisation of the down town and housing estates.

Very important and high effective was change of tree alleys in Naperville street, where original older trees of *Populus* × *canadensis* were replaced by *Celtis occidentalis* street trees, mainly for reason of human body allergy elimination. Smaller poplar's alley (*P*. × *canadensis*) is planted at the Nitra river embankment in neighbouring of birch Grove Park and also throughout circumference of football and hockey stadium as protection green barriers. Particular survey of species composition of contemporary tree alleys at selected street spaces and squares of Nitra town is presented in Table 1, including the tree assessment according to described methods.

At the Nitra territory there have been mapped 26 most important street tree alleys where 22 species and cultivars were identified and assessed. To the most traditional street trees belongs linden, which represents the oldest alley at Nábrežie mládeže embankment. To the latest tree elements belong e.g. Gingko biloba L. 'Fastigiata' and Liquidambar styraciflua L. Originally, perspective of Prunus fruticosa 'Globosa' species shows its weak point because attack by Erwinia amylovora fungus has been identified at older trees in Sládkovičova street. At assessment method of the street tree alleys we paid attention to the marks, which have been used very rare till now in the research and practical mapping. Most often valuation marks up to the present time were pest and diseases occurrence at the vegetation organs, tree vitality characteristics, tree stability and social values of trees as well. Those assessment characteristics and methodical approaches according to individual authors are described in complex publication of group authors (HRUBÍK et al., 2011).

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No	Locality, street, square	Name of woody plant	a) (a	naracter	ISLICS		o) Envir adapta	onmenta ability	_		c) Biol valu	ogy-ecol	ogical		d) Cult value	es	e) Ne inf	gauve	
			ac	ad	ae	ba	рр	þc	pq	ca	cb	cc	cq	ce	da	db	ea	eb (0
1	Štefánikova Street OD MLYNY	Robinia pseudoacacia L. Bessoniana´	3	7	5	3	3	3	3	2	1	2	1	2	2	1	1	1	1
5	7. Pešieho pluku Street	Tilia platyphyllos L.	12	70	4	2	5	5	5	3	3	3	2	3	3	2	1	1	2
3	Damborského Street	Crataegus monogyna L. 'Paul's Scarlet'	4	×	3	5	з	ю	5	-	2	5	5	-	2	-	-	-	_
4	Bernolákova Street	Tilia platyphylos L., Tilia cordata Mill.	10	80	ω	7	7	7	7	ω	б	б	5	m	e	7	-	-	12
5	Štúrova Street ZSS + SOU	Tilia cordata Mill.	5	50	4	5	5	5	5	3		3	2	3	3	2	_	-	0
9	Štúrova Street Dom služieb	Fraxinus excelsior. L. 'Globosa'	9	20	5	5	5	5	5	5	5	5	5	3	3	5	5	5	_
7	Square of Cyril a Metod	Aesculus hippocastanum L.	12	90	4	2	2	2	2	3	3	3	3	2	3	2	3	1	3
8	Farská Street	Acer platanoides L. 'Globosa',	5	40	33	2	2	5	2	2	2	2	2	2	3	2	-	-	1
		Tilia plataphyllos L.	6	50	б	7	7	7	7	3	3	3	5	3	3	2	-	-	2
6	Kupecká Street	Gingko biloba L. Fastigiata´	5	9	5	ω	e	3	ю	5	5	5	-	5	3	3	_	-	_
10	Square of Svätopluk	Corylus avelana L. 'Globosa'	7	20	4	5	3	3	3	3	e	5	-	5	5	5	3	5	0
=	Štefánikova Street pedestrian zone	Prunus fruticosa Pall. 'Globosa'	ŝ	×	4	5	5	5	5	5	2	3	5	5	3	2	_	_	_
		Sorbus aria (L.) Crantz. 'Lutescens'	5	10	5	3	3	3	3	2	3	7	-	2	3	2	2	1	1
12	Kúpeľná Street	Tilia platyphyllos L.	12	80	33	2	2	2	2	3	3	3	2	3	3	2	1	1	5
13	Sládkovičova Street	Prunus fruticosa Pall. 'Globosa'	4	15	5	7	7	7	7	7	5	ю	5	5	3	5	_	-	_
14	B. Němcovej Street	Fraxinus excelsior L. 'Globosa'	Ξ	80	ŝ	5	7	0	7	7	2	7	2	3	ю	2	2	5	
15	Kmeťkova Street	Tilia platyphyllos L. Tilia poulate Mill	12	80	4	5	5	5	5	ю	ю	ю	5	ŝ	ю	5	-	-	5
		<i>Tilia cordata</i> Mill.																	
16	Nábrežie mládeže and Wilsonovo nábrežie Embankment	Tilia platyphyllos L. Tilia cordata Mill.	18	90	5	2	2	2	2	3	3	3	2	3	3	2	1	1	5
17	Slančíkovej Street, Chrenová	Prunus cerasifera Ehrh. ´Nigra´	4	7	5	3	ю	3	3	2	2	5	1	2	3	2	1	1	_
18	Fraňa Mojtu Street	Prunus serrulata Franch. 'Hisakura'	5	15	5	2	3	2	2	2	2	3	2	2	3	2	1	1	5
19	Štefánikova Street, OD-TESCO	Koelreuteria panniculata Laxm.	4	9	4	3	3	3	3	1	1	1	1	1	3	2	1	1	5
20	Štefánikova Street, VÚB	Catalpa bignonioides Walt. 'Nana'	5	20	4	2	2	7	2	2	2	2	2	2	3	3	1	-	_
21	Štúrova Street, OD-MLYNY	Acer platanoides L. 'Columnare'	4	×	5	7	7	0	7	7	5	7	2	5	ю	3	_	_	_
22	Čsl. Armády Street	Liquidambar styraciftua L.	7	12	5	ю	e	e	ю	7	5	5	1	5	ю	3	_	_	
23	Nepervillská Street	Celtis occidentalis L.	9	12	5	3	3	ю	ю	3	3	5	3	3	3	2	-	-	_
33	Mostná Street	Carpinus betulus L. 'Columnare'	8	50	3	3	2	3	2	3	3	2	1	2	2	2	3	1	-
		Pyrus calleryana Decne. 'Chanticleer'	3	7	5	3	3	3	3	2	2	2	1	2	3	3	1	1	1
25	Župné Square	Acer platanoides L. 'Globosa'	5	40	ю	7	2	7	7	7	2	7	2	2	5	2	1	-	_
		Populus alba L. Boleana	10	40	3	2	3	2	3	2	2	2	2	3	2	2	1	1	_
26	Janka Kráľa Street	Acer saccharinum L. (yearly prunned)	S	50	4	3	ю	ю	3	5	5	5	5	5	23	6	1	1	_

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Achieved results of our research activity have showed positive trends in tree alley reconstruction on the on hand. On the other hand many tree alleys in Nitra town are destroyed, trees are too old, and unseemly that would be reason for more effective decision to remove them from street soon. In some streets we may see alleys with different tree species, which looks very diversified and out of composition rules, e.g. Štefánikova street, Župné square. Every year tree branches cutting at Acer saccharinum L. in J. Kráľ street creates compacted and unified street alley. Species of Populus genus, with regards to fast growing character and big tree crown should be useful to the future also as elements for protection barriers, as shade tree and also for windbreaks around large sport centres, but selected male gender trees only (VARGA, 1994).

In conclusion, our research results show positive and negative feedback. In our opinion Nitra town needs to elaborate complex master plan for reconstruction and quality improving of main and the most attractive streets regarding tree alleys.

Acknowledgement

Paper was elaborated thanks to financial supporting by the grant project KEGA No. 020SPU-4/2011 and No. 019SPU-4/2011 on Ministry of Education, Science, Research and Sport of the Slovak Republic.

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Received December 6, 2012 Accepted September 30, 2013

Effect of delayed tending on development of beech (*Fagus sylvatica* L.) pole stage stand

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Abstract

ŠTEFANČÍK, I. 2013. Effect of delayed tending on development of beech (*Fagus sylvatica* L.) pole stage stand. *Folia oecol.*, 40: 272–281.

The paper deals with assessment of the long-term experiment (45 years of investigation) in beech (*Fagus sylvatica* L.) stand with delayed tending started at stand age of 60 years. The research was performed on four partial plots by different methods of their management: (i) plot with heavy thinning from below (C degree according to the German forest research institutes from 1902), (ii) plot with the free crown thinning (thinning interval of 5 years), (iii) plot with the free crown thinning (thinning interval of 10 years) and (iv) control plot (with no thinning). From qualitative point of view, the best results according to the number of target (crop) trees were found on plots tended by the free crown thinning (thinning interval of 5 years), and the worst on plots with heavy thinning from below and/or plot with no tending (control plot). Consequently, the results showed lower number of target (crop) trees in comparison with our assumption and/or the model developed for beech stands in the past. On the other hand, from quantitative point of view, the best results were achieved on plot tended by heavy thinning from below, followed by the plot with the free crown thinning (thinning interval of 5 years).

Keywords

beech, crop trees, quantitative production, stand structure, tending

Introduction

Tending of each forest stand has a crucial importance for its development. As a rule, it takes more than half of rotation age of forest stand. For the management of beech stands originated from natural regeneration, is very important not only the method of their tending, but also the stand age, when to start with tending.

The papers dealt with the history of beech stands tending in Slovakia (ŠTEFANČÍK, 1985) and other literature focused on problems of thinning in beech stand (ŠTEFANČÍK, 1984) concluded, that tending of beech stands had not so special tradition in comparison to France, Denmark or Germany. It should be stated, that tending was realized only according to foreign knowledge and poor experiences of internal forest practitioners. The systematic research started at the end of the 50-ies, of the last century.

The aim of the research at this time was mainly to find the first scientific results on the mentioned topic under the natural condition of Slovakia. Within the framework of thinning problems, the attention was especially paid to know, which kind of selective thinning method should be considered the most suitable for beech stands under our natural condition. Nevertheless, actual experiences by application of the methods developed abroad, and also the condition of beech premature stands in Slovakia, have been taken into account (ŠTEFANČÍK, 1984).

Research was by the first time focused on the beech thickets, not systematically tended until then (RÉH, 1968, 1969) and/or small pole stage stands or pole

stage stands (ŠEBIK, 1969; ŠTEFANČÍK, 1974). Within the framework of the research, all principal silvicultureproduction questions of thinning started to be solved step by step. In the initial stage of the research it was especially the problem related to thinning type (thinning from below, crown thinning), method of selection (positive, negative) and structure of pure beech stands, as well as thinning intensity, i.e. intensity, frequency and thinning interval, later on.

Effect of two degrees of thinning from below (B and C according to German forest research institutes from 1902) and two crown thinning methods (qualitative according to Schädelin) and the free crown thinning according to ŠTEFANČÍK (1984) started to be verified by the research.

Since the 70-ies of last century, the results of longterm investigation have been published (ŠTEFANČÍK, 1974, 1984; ŠEBÍK and POLÁK 1990; ŠTEFANČÍK et al., 1991, 1996; ŠTEFANČÍK, 2007; ŠTEFANČÍK and BOLVAN-SKÝ, 2011). The outcomes showed better results by application of crown thinning in comparison with thinning from below. Especially, the free crown thinning (ŠTEFANČÍK, 1984, 2007) appeared to be suitable for tending of pure beech stands in Slovakia. Since 1958, the above-mentioned method has been applied in the thinning research of beech stands. Nowadays, after long-term verification it was successfully put into the practise.

The aim of this paper was to ascertain the changes of selected parameters of quantitative and qualitative production in beech stands, tended by different thinning methods for a long time (45 years of investigation).

Material and methods

The research was carried out on the series of permanent research plot (PRP) Cigánka, established in the stand located in compartment 50, forest district Muráň, forest enterprise Revúca. The given beech stand originated from a natural regeneration. The stand age on the PRPs at their establishment (in autumn 1966) was 60 years.

The mentioned series of PRPs consists of four partial plots (C, H, H2, 0) with the area of each plot of 0.25 hectare. The basic mensurational characteristics are presented in Table 1.

On the plot (marked as C) a heavy thinning from below (C degree according to German forest research institutes from 1902) was realized. On the second and the third plot (marked as H and H2), the method of the free crown thinning (according to ŠTEFANČÍK, 1984) was applied. The mentioned method is focused on individual tending of the trees of selective quality (promising and target trees). These trees are selected on the base of the three criteria (quality, dimension and spacing). Thinning interval on plot H is 5 years, and on plot H2 10 years.

Table 1. The basic characteristics of the giver	series
of permanent research plots (PRPs)	Cigánka

Characteristic	PRP Cigánka
Establishment of PRP	Autumn 1966
Age of stand [years]	60 (in 1967)
Site index	30
Geomorphologic unit	Stolické vrchy
Exposition	Northwest
Altitude [m]	560
Inclination [degree]	20
Parent rock	Gneiss (biotitic)
Soil unit	Haplic Cambisol (Dystric)
Forest altitudinal zone	4 th beech
Ecological rank	A (Acid)
Management complex of forest types	405 – acid beech woods
Forest type group	<i>Fagetum pauper</i> (Fp) higher tier
Forest type	4301 woodrush beech woods (higher tier)
Average annual temperature [°C]	5.5
Sum of average annual precipitation [mm year ⁻¹]	918

The plot marked as 0 is control (with no thinning).

No planned silvicultural interventions were carried out up to establishment of PRPs. The first measurement was performed in 1967. Since establishment of PRPs, 10 biometrical measurements were realized on each partial plot with the interval of 5 years, including the intervention (only on treated plots) with thinning interval of 5 years (plot C and H) and/or 10 years (plot H2). On all plots, the standard biometrical measurement and evaluation of stem and crown of trees were carried out.

Within the framework of the measurement, the quantitative parameters (breast height diameter, both height of tree and base of tree crown, crown width) were assessed according to silvicultural and commercial classification. They were focused on evaluation of each tree, and separately on the trees of selective quality (promising and target trees).

Silvicultural classification consists of:

- a) Biosociological position of trees according to growth (tree) classes (ŠTEFANČÍK, 1984):
 - 1. dominant tree
 - 2. co-dominant tree
 - 3. intermediate tree
 - 4. suppressed tree decreased
 - 5. suppressed tree dying out
- b) Degree of stem quality:
 - 1. well-shaped and straight, best stem quality, without burrs

- average shaped average stem quality, crooked only in upper third of the stem, low number of burrs
- 3. bad shaped worse quality of the stem, high number of burrs, very crooked
- c) Degree of crown quality:

According to the type (form of ramification): 1. crown with continuous axis of stem up to the top of tree; 2. bouquet (cluster); 3. broom; 4. forked. According to size: 1. oversized; 2. normal size; 3. small size, asymmetrical developed, but able to regenerate; 4. too small size, not able to regenerate. According to crown density (sufficiency of foliage): 1. good density with complete foliation, also inside of the crown; 2. sufficient density, with foliation in outside of the crown only; 3. sparse, foliation quite well; 4. very sparse, insufficient foliation.

Within the framework of commercial classification, only lower part of the stem up to crown base was assessed, separately for lower and upper half of the stem, respectively. Quality classes: 1. very high (A), 2. average (B), low quality, but industrial wood (C), 4. fuelwood (D).

The calculation of the results was performed by standard methods for tending evaluation and production-silviculture relations, utilized the software package of QC Expert and growth simulator Sibyla (FABRIKA, 2005). To find out the statistical significance of the differences, the single-factor analysis of variance (ANOVA) was used.

Results and discussion

Diameter structure

The diameter development of the investigated PRPs is characterized by the diameter frequency distribution (Figs 1 and 2), as well as by the values of mean diameter (d_{u}) presented in Table 2.

In the initial stage of the research, the course of curves of diameter frequency distribution was found similar to all partial plots (Fig. 1). It is a type of lefthand asymmetric distribution, typical for young stands, as well as for the middle age ones, which were untouched (neglected by tending) until then. This is also the case of PRP Cigánka, where tending started at the growth stage of pole stage stand (60 year old), which is considered to be too delayed for beech stands. On the base of numerous experimental experiments, it is recommended to start with tending already in the thickets (Réh, 1968, 1969; JURČA and CHROUST, 1973; KORPEĽ et al., 1991) and/or no later than in small pole stage stand (ŠTEFANČÍK, 1974). The highest values of the mean diameter (d) were found on plot C, and the lowest on control plot (0).

After 45 years under different thinning regime, the differences among the plots increased (statistical significant differences at the level $\alpha = 0.05$ were found only between plot C and each other plots). The order of plots remained unchanged, when the highest values of d_{α} were found on plot C (heavy thinning from below),



Fig. 1. Diameter frequency distribution on plots in the initial stage of the research in 1967.



Fig. 2. Diameter frequency distribution on investigated plots in 2012.

and the lowest on control plot (with no treatment), Table 2. Simultaneously, the diameter frequency distribution was more or less changed to double-peak course (Fig. 2). The above-mentioned development response the thinning methods realised. On the plots tended by the free crown thinning (H, H2), the interventions

Plot	Stand	Age	N	G	V _{7b}	Mea	an
						Diameter d _{1,3} [cm]	Height [m]
		[year]	[pcs ha ⁻¹]	$[m^2 ha^{-1}]$	$[m^3 ha^{-1}]$	$[d_g]$	$[h_g]$
0	Total	60	2,940	34.784	337.444	12.3	19.1
	Main	65	2,276	35.768	357.892	14.1	19.8
		70	2,004	35.956	392.920	15.1	21.4
		75	1,736	36.980	437.292	16.5	23.3
		80	1,592	38.036	477.004	17.4	24.7
		85	1,472	38.604	516.368	18.3	26.2
		90	1,224	39.208	537.228	20.2	26.7
		95	1,144	41.404	570.180	21.5	26.7
		100	1,068	41.556	585.912	22.3	26.6
		105	1,012	43.956	631.524	23.5	26.9
Н	Total	60	2,632	36.436	365.172	13.3	20.1
	Main	65	1,140	25.888	303.516	17.0	23.2
		70	816	22.480	286.980	18.7	25.0
		75	756	23.120	300.944	19.7	25.1
		80	624	21.172	287.976	20.8	26.2
		85	620	25.128	351.032	22.7	26.6
		90	584	27.388	389.152	24.4	26.8
		95	584	31.064	455.964	26.0	27.4
		100	564	31.724	485.060	26.8	27.5
		105	548	34.452	540.876	28.3	27.8

Table 2. Development of stand characteristics

Plot	Stand	Age	N	G	V _{7b}	Mea	ın
						Diameter d _{1,3} [cm]	Height [m]
		[year]	[pcs ha ⁻¹]	$[m^2 ha^{-1}]$	[m ³ ha ⁻¹]	$[d_g]$	$[h_g]$
H2	Total	60	2,568	35.500	354.504	13.3	20.1
	Main	65	1,552	28.968	307.740	15.4	21.0
		70	1,032	23.808	284.196	17.1	23.6
		75	992	25.948	319.808	18.3	24.0
		80	800	22.012	279.256	18.7	24.5
		85	784	24.992	334.212	20.2	25.7
		90	740	26.128	355.132	21.2	25.4
		95	732	28.916	398.292	22.4	25.5
		100	712	29.128	418.452	22.8	25.6
		105	704	31.792	468.432	24.0	26.1
С	Total	60	2,308	40.060	444.152	14.9	21.6
	Main	65	520	26.696	387.996	25.6	29.8
		70	440	27.096	416.032	28.0	31.5
		75	324	26.024	420.956	32.0	33.2
		80	312	28.468	482.532	34.1	34.7
		85	312	31.896	563.500	36.1	35.9
		90	280	32.104	595.804	38.2	37.2
		95	280	35.168	668.400	40.0	38.0
		100	272	36.052	690.564	41.1	38.2
		105	272	38.728	778.980	42.6	38.8

Table 2. Development of stand characteristics - continued

N, number of trees; G, basal area; V_{7b} , volume of the timber to the top of 7 cm o.b.

 $C \rightarrow plot$ with thinning from below. $H \rightarrow plot$ with thinning from above, thinning interval 5 years. $H2 \rightarrow plot$ with thinning from above, thinning interval 10 years. $0 \rightarrow control plot$ (with no treatment).

were performed in the whole vertical profile, which resulted in a better diameter differentiation. It was also confirmed by the values of indices of diameter differentiation (TM_d) according to (FULDNER, 1995), which were found the highest, just in the plots treated by the free crown thinning (for H = 0.578 and H2 = 0.516). The values above 0.500 represent the strong type of differentiation. For comparison, in the control plot it was 0.398 (medium type of differentiation) and the lowest values of indices were obtained in plot C (0.173 – little differentiation), where total suppressed level of the stand was removed by the treatment.

Height structure

The height (stand) structure of the investigated plots was expressed by the relative number in the growth (tree) classes (Fig. 3). The proportion of trees in the crown level of the stand $(1^{st} + 2^{nd} \text{ growth class})$ and the suppressed level of the stand $(3^{rd} \text{ to } 5^{th} \text{ growth class})$ is very important from the silvicultural point of view. The

structure depends especially on site, tree species, stand age and tending measures (ŠEBík and POLÁK, 1990).

In the initial stage of the research, the height structure was practically the same. The proportion of the suppressed level of the stand ranged from 28.4% on control plot to 29.9% on plot H2. The differences (shifts) in the height structure (proportion between the crown level of the stand and the suppressed one), after 45 years of investigation were found only at about 10% (plot 0 and H). Contrary to the mentioned plots, plot H2 remained unchanged. These results are in accordance with the outcomes published by ŠEBÍK and POLÁK (1990), who stated the shift of the trees to the higher growth (tree) classes, when heavy crown thinning was applied. The mentioned authors also concluded that decreased number of co-dominant trees in the stand with shade-bearing species is typical, together with increased amount of the suppressed ones. The most proportioned are being the fourth, or the 4th and the 5th growth class, which was also confirmed by our research on PRP Cigánka. The similar results were published by



Fig. 3. Relative number according to the growth classes on plots after 45 years in 2012.

Assmann (1968) for 102 years old beech stand tended by mild crown thinning, where proportion of the crown level of the stand and the suppressed level of the stand was found of 53.8% and 46.2%, respectively.

The highest changes were registered on plot C (heavy thinning from below), where in a consequence of removed suppressed level of the stand remained only intermediate individuals (the 3^{rd} growth class) with low proportion of 17.6%.

Very interesting should be considered the fact, that control plot left to self-development showed practically the same height structure in comparison with the plots tended by the free crown thinning (Fig. 4). This was also confirmed by the statement published in the past (ŠTEFANČÍK, 2007), that according to its conception, the mentioned thinning method is very similar to principles of close to nature silviculture. It was also proved by the values of indices characterized the vertical structure (APi) according to PRETZSCH (1992). On the plots with the free crown thinning, the values were found of 0.791 and 0.758. For example, in the stand with a selection structure, the mentioned index is able to achieve the value of 0.900 (PRETZSCH, 1992). For comparison, we suggest, that on the control plot (0) in PRP Cigánka, the index was found of 0.447. Consequently, the indices of the height differentiation (TM_h) according to FULDNER (1995) were found the highest for plots tended by the free crown thinning (H = 0,514 and H2 = 0,439), contrary to control plot (0.302) and plot C (0.037).

As for the comparison of the values of the mean height (h_g), after 45 years, the highest differences (statistical significant at the level $\alpha = 0.05$) were found between plot C and each other plots. The rest three plots showed similar values, but differences among them were statistically insignificant.

Fig. 4. Stand structure on PRP Cigánka plot 0 (left) and plot H (right).

Development of quantitative production

The development of stand characteristics during the investigated period is presented in Tables 2 and 3. At establishment of the plots, the highest initial number of trees (N) was found on control plot (0) and the lowest on plot C. After 45 years, the order was not changed, whereby on control plot remained 34.4% out of the initial number of trees, but on the plot C only 11.8%.

As for the other stand characteristics (basal area – G, and volume of the timber to the top of 7 cm o.b. – V_{7b}), the highest values were found on plot tended by heavy thinning from below and the lower on plots with the free crown thinning (H, H2). These results are in accordance with the experiences of numerous thinning experiments established in the past, concluded by ASSMANN (1968), ŠEBÍK and POLÁK (1990), ŠTEFANČÍK (1990).

The analysis of the total decrease (thinning, selfthinning, abiotic injurious factors) according to G and V_{7b} for the period of 45 years showed the highest percentage on plots tended by the free crown thinning (H, H2) and the lowest on control plot (Table 3).

As for the total production (according to G and V_{7b}), the highest values were found on plot with heavy thinning from below and the free crown thinning (thinning interval of 5 years). The same results were also obtained, by expression of growth index of the total production in investigated period. It suggests suitable effects of even though delayed tending measures in beech stands. Additionally, beech species is well-known of its very good responses to liberation (releasing) up to the oldest period (ASSMANN, 1968; ŠEBIK and POLÁK, 1990). It was fully confirmed by the results from PRP Cigánka.

It should be concluded, that from quantitative point of view, the best results were obtained on plots tended by heavy thinning from below and the free crown thinning with thinning interval of 5 years, contrary to control plot (0), characterized by the worst outcomes. It was also confirmed by the values of the current annual increment on basal area and/or volume increment in 5 years periods (Figs 5 and 6). The total mean annual volume increment during the investigated period was found 14.3 m³ha⁻¹ on plot C, followed by plot H – 11.1 m³ha⁻¹, plot H2 – 9.0 m³ha⁻¹ and control plot – 8.7 m³ ha⁻¹.

Development of target (crop) trees

Information related to the target (crop) trees (TT) development, representing qualitative production in commercial forests is presented in Table 4. It can be seen, that from quantitative parameters point of view, in the initial stage of the research, the highest values were found on plot C and/or the lowest on plot H2. Number of TT ranged from 176 to 208 individuals per hectare.

During the tending period of 45 years, the situation was changed unambiguously in favour of plots treated by the free crown thinning (H and H2). On the mentioned plots, double number of TT was cultivated in comparison to plot tended by heavy thinning from below (plot C). The same results were obtained, if we take into account the production parameters (basal area, volume of the timber to the top of 7 cm o.b.). The proportion of TT out of the main stand is considered to be a very important parameter. The plots managed by the free crown thinning showed also the best results according to the mentioned quantitative parameters in comparison with plots tended by heavy thinning from below, or control plot. The model of the future mature beech stand developed by ŠTEFANČÍK (1984) assumed at stand age of 110-130 years, in acid site, the number of TT presented 173 to 200 trees per hectare and 376 $m^3 ha^{-1}$ of volume of the timber to the top of 7 cm o.b. Its proportion had to be of 75% out of the main stand. Mean diameter d_{1,3} was assumed to achieve 40 cm. It can be seen, that the results from the PRP Cigánka obtained at stand age of 105 years are very close to the mentioned model, except for number of TT, which is much lower. It is a consequence of delayed tending, which started at stand age of 60 years. It is a generally

Plot	Age			Total deci	ease of tre	es				Total produc	tion	
	range	Ν		(3	V	7b	Ν		G	,	V _{7b}
		[pcs ha ⁻¹]	% of	$[m^2 ha^{-1}]$	% of TP	$[m^3 ha^{-1}]$	% of TP	[pcs ha ⁻¹]	$[m^2 ha^{-1}]$	Index of total stand	$[m^3 ha^{-1}]$	Index of
	[years]					ТР				total stand		total stand
0	60–105	1,928	65.6	12.856	22.6	98.616	13.5	2,940	56.812	1.633	730.140	2.164
Н	60–105	2,084	79.2	29.688	46.3	321.528	37.2	2,632	64.140	1.760	862.404	2.362
H2	60–105	1,864	42.6	27.192	46.1	291.424	38.4	2,568	58.984	1.662	759.856	2.143
С	60–105	2,036	88.2	27.884	41.9	307.660	28.3	2,308	66.612	1.663	1,086.640	2.447

Table 3. Development of quantitative production of the stand for 45 years

N, number of trees; G, basal area; V_{7b} , volume of the timber to the top of 7 cm o.b.; TP, total production.

 $C \rightarrow \text{plot}$ with thinning from below. $H \rightarrow \text{plot}$ with thinning from above, thinning interval 5 years. $H2 \rightarrow \text{plot}$ with thinning from above, thinning interval 10 years. $0 \rightarrow \text{control plot}$ (with no treatment).

Fig. 5. Current annual basal area increment in the 5 years period of investigation.

Fig. 6. Current annual volume increment in the 5 years period of investigation.

Plot	Age	Ν	(ĩ	V	7b	Me	an
				% out of		% out of	diameter	height
	[years]	[pcs ha ⁻¹]	$[m^2 ha^{-1}]$	main	$[m^3 ha^{-1}]$	main	d _{1,3} [cm]	[m]
				stand		stand	$[d_g]$	$[h_g]$
0	60	200	6.688	19.2	80.992	24.0	20.6	25.4
	105	108	11.420	26.0	191.048	30.3	36.7	32.4
Н	60	188	6.428	25.2	79.308	29.1	20.9	25.6
	105	124	18.332	53.2	320.988	59.3	43.4	33.5
H2	60	176	6.512	24.0	81.312	29.4	21.7	26.0
	105	132	16.724	52.6	282.404	60.3	40.2	32.7
С	60	208	10.372	38.3	138.636	40.1	25.2	27.5
	105	68	13.303	34.3	277.620	35.6	49.9	40.0

Table 4. Development of target (crop) trees

N, number of trees; G, basal area; V_{7b} , volume of the timber to the top of 7 cm o.b.

 $C \rightarrow \text{plot}$ with thinning from below. $H \rightarrow \text{plot}$ with thinning from above, thinning interval 5 years. $H2 \rightarrow \text{plot}$ with thinning from above, thinning interval 10 years. $0 \rightarrow \text{control plot}$ (with no treatment).

known fact, that the best stand age in order to determine and cultivate the TT is considered at the period of 30–40 years (ŠTEFANČÍK, 1974, 1984). As it can be seen, the obtained results from PRP Cigánka showed that it is possible to achieve assumed quantitative production in case of delayed, but systematic tending. On the other hand, it is not possible to cultivate desired qualitative production represented by number of trees with the best quality (target trees), especially on plot managed by heavy thinning from below, or plot without tending.

Conclusions

Based on the 45 years of investigation of beech stand development managed by delayed tending, where different methods of tending were applied, it can be concluded:

- The differences of diverse tending regime were increased between plots after 45 years of investigation in comparison with the initial stage of the experiment. The differences were found significant at the level $\alpha = 0.05$ between plot C and each other plot. From diameter structure point of view, the order of plots remained unchanged. The highest mean diameter (d_g) was found on plot managed by heavy thinning from below, from the initial stage up to now. The lowest one showed the control plot.
- The differences (shifts) in the height structure (proportion of the crown level of the stand and the suppressed level of the stand) on plots during the investigated period of 45 years were found at about 10% on plot 0 and H. For plot H2 it remained unchanged. The highest changes were registered on plot C (heavy thinning from below), where due to remov-

ing of the suppressed level of the stand, only intermediate individuals (the 3^{rd} growth class) remained in the stand with lower proportion of 17.6%.

- The control plot, left to the self-development showed practically the same height structure like the plots tended by the free crown thinning (H and H2).
- From quantitative point of view, the best results were found on plots tended by heavy thinning from below and the free crown thinning with thinning interval of 5 years. Consequently, the worst results were obtained from control plot.
- As for the total production (expressed by basal area and volume of the timber to the top of 7 cm o.b.), the highest values were found on plot tended by heavy thinning from below and plot with the free crown thinning (thinning interval of 5 years). The same results were also obtained according to the index of the total production. It suggests suitable effect of tending, although delayed, in older beech stands.
- The number of target (crop) trees in the initial stage of stand ranged from 176 to 208 individuals per hectare. At the stand age of 105 years, after tending for 45 years, the highest number of crop trees was showed by the plot tended by the free crown thinning (124 and 132 pieces per hectare), and the lower by the plots managed by heavy thinning from below and control plot (68 and 108 pieces per hectare, respectively).
- The results, found by long-term investigation (period of 45 years) confirmed, that by systematic and intensive tending, although delayed, it is possible to achieve desired quantitative production, but not qualitative production, represented by the number of the best quality (target) trees, especially on control plot and plot tended by the free crown thinning.

Acknowledgements

This work was supported by the Slovak Research and Development Agency under the contract No. APVV-0262-11 and Technology Agency of the Czech Republic TA02021250 Silvicultural-ecological and economic optimum of forest stand tending.

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Received February 6, 2013 Accepted April 8, 2013

Results of an ecological-production research on forest ecosystems of woody plants introduced to Slovakia

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Abstract

TOKÁR, F. 2013. Results of an ecological-production research on forest ecosystems of woody plants introduced to Slovakia. *Folia oecol.*, 40: 282–289.

The work gives a chronological list of the results obtained in an ecological and production research on 49 coniferous and 10 broadleaved exotic woody plants in 298 parks and woody subjects across Slovakia. The results can be used in orchard and forestry practice. Since 1971, the research has been oriented to assessment of forest ecosystems and phytotechnique for forest stands consisting of selected exotic woody plants Pinus nigra Arnold, Castanea sativa Mill., Quercus rubra L. and Juglans nigra L. In the area of the Little Carpathians the best results in growth and production were achieved in *Pinus nigra* Arnold at the age of 100 years under proportion rate up to 30% in the group of forest types (slt) Querceto-Fagetum (464 m³ ha⁻¹), Fagetum pauper $(463 \text{ m}^3 \text{ ha}^{-1})$ and Fageto Quercetum $(432 \text{ m}^3 \text{ ha}^{-1})$. In the pure stands the highest stock was observed in the group of forest types Querceto-Fagetum (310 m³ ha⁻¹). In Castanetarium Horné Lefantovce the best results out of 86 Castanea sativa progenies were obtained in 15 progenies (Jelenec 2, Horné Lefantovce A, Tlstý Vrch 1, 2, 2', 3, 4, 9, Duchonka 2, 3, 5, 6, 10, 12, Bratislava 4) and the worst results were obtained in seed progenies Stredné Plachtince 5, Krná 3, Modrý Kameň 7. Following evaluation of phytotechnique impact on production of different stand types of Castanea sativa Mill. at age of 38 years, the highest stock was observed in mixed stands *Tilia cordata* Mill. (416 m³ ha⁻¹, 190 t ha⁻¹, total production 635 m³ ha⁻¹, 333 t ha⁻¹). In mixed stands of Juglans nigra L. (20%) and Quercus rubra L. (80%) in the locality Ivanka pri Nitre, the highest stock was observed at the age of 48 years (438 m³ ha⁻¹, 263 t ha⁻¹) and total production 662 m³ ha⁻¹ and 410 t ha⁻¹. In the locality Sikenica in pure stands of Juglans nigra L. the highest stock at the age of 64 years was found in the stand with the strong crown thinning (464 m³ ha⁻¹, 195 t ha⁻¹, total production 573 m³ ha⁻¹ and 246 t ha⁻¹). In addiction to these production characteristics also leaf area indices were assessed (LAI).

Keywords

ecology, exotic woody plant, forest ecosystems, production

Analysis of the issue and the research focus

In the past in Slovakia attention was given predominantly to the growth and production of autochthonous woody plant species (HALAJ, 1963; HALAJ and ŘEHÁK, 1979; ŠEBÍK and POLÁK, 1990; ŠMELKO, 2000). In allochthonous woody plant species issues of growth, production and distribution were evaluated (HOLUBČÍK, 1968; BENČAŤ, 1982). The aim of our work is to introduce chronological survey of obtained results from ecologicproduction research.

The Department of system and ecology of woody plants of the former Institute of Dendrobiology SAS in

the Arboretum Mlyňany SAS focused their research on forest ecosystems of woody plants introduced to Slovakia on the following points:

- Taxonomy of exotic species in selected dendrological subjects in Slovakia (assortment, mensurational data, fertility, natural regeneration)
- Valorisation of structure and production (volume, mass) and quality of various stands of selected exotic woody plants in Slovakia (*Castanea sativa* Mill., *Quercus rubra* L., *Juglans nigra* L., *Pinus nigra* Arnold)
- Assessment of effects of phytotechnique (thinning) on production, dendrochronology, quality, leaf area

index (LAI) and energy potential in a variety of stand types of woody plants introduced into Slova-kia

- Monitoring of physiological-biochemical aspects of biomass production in various stand types of exotic woody plants (fluorescence, contents of selected elements in soil and leaves)
- Assessment of resistance of stands of selected exotic woody plants against biotic and abiotic harmful agents
- Evaluation of herb vegetation in various stand types of exotic woody plants and changes to this component due to long term introduction (Castanetarium in Jelenec, Castanetarium Horné Lefantovce)
- Evaluation of natural regeneration of stands of exotic species in Slovakia
- Quantitative assessment of selected chemical elements accumulated in aboveground biomass and in soil in stands consisting of exotic woody plants in Slovakia.

Material and methods

The ecological description of the exotic species distribution in parks and dendrological objects in Slovakia has been adapted from BENČAŤ, 1982.

With using our own measured data and the data from Forest management plans (FMP), we have accomplished ecological-production analysis for 613 stands of black pine (*Pinus nigra* Arnold) in the region Malé Karpaty Mts. We considered the group of forest types (gft), stand age and structure and black pine proportion (1–30%, 31–60%, 61–90% and 100%). The results were processed with using the Korf growth function, on a computer TESLA 200 in the Computing centre of the Technical University in Zvolen.

The phytotechnique of various stand types (pure and mixed stands with different rates of domestic and alien woody plants) of Castanea sativa Mill., Quercus rubra L. and Juglans nigra L. works with thinning from above applied in graded intensity (moderate, heavy), with positive selection, at different repetition intervals (5-10 year), focusing on tending promising trees on the permanent research plots series (PRP) Žirany (7 partial PRPs with homogeneous and mixed stands of Castanea sativa Mill.), Ivanka pri Nitre (6 partial PRPs with homogeneous and mixed stands of Quercus rubra L. and Juglans nigra L.), Sikenica (3 partial PRP with homogeneous stands of Juglans nigra L.) and Castanetarium Lefantovce (86 seed progenies of Castanea sativa Mill. from 12 localities in Slovakia). The ecological description of the PRP series Žirany, Lefantovce, Ivanka pri Nitre and Sikenica can be found in TOKÁR, 1987, 1998; the Castanetarium Lefantovce is characterised in Tokár, 2003; Tokár and Kukla, 2006.

Biometrical measurements of the stand height, diameter $d_{1,3}$ and standing volume (volume production) were performed by methods commonly used in forestry practice (HALAJ, 1963; ŠMELKO, 2000). For the calculation of the volume of large black pine timber, we used, due to the lack of our own tables, the mass tables for forest pine, red oak, black nut; for edible chestnut the mass tables for oak converted per one hectare.

The aboveground wood biomass was obtained by the destructive method (method of sample trees). The total number of sample trees for each woody species in the stand was determined by stratified selection (ŠMELKO and WOLF, 1977). The mass of stem, branches, annual shoots and leaves was obtained by weighing on a scale Kamor in dry mass at 105 °C.

Photosynthetically active leaf surface area was estimated with the aid of a photo-planimeter EIJ-KELKAMP.

The time dependence of the values of standing volume and mass as well as the values of overall volume and mass production (growing stock + thinning + mortality + other losses) and the LAI values was fitted with a mathematical function – specific for each tree species and stand type (an exponential or a 2^{nd} degree polynomial). On each PRP series, the production results expressed through growth index and through index increment per cent were compared with the control PRP (without intervention) and tested statistically with the t-test (ŠMELKO and WOLF, 1977).

The principle of phytotechnique of stands of exotic woody plants is tending the promising trees (Tokár 1987, 1998). The contents of elements (Mg, Ca, K, Na, Zn, Pb, Fe, Cu, Mn) in the aboveground biomass and in the soil were assessed with the aid of an absorption spectre-photometer IL VIDEO 12 (Tokár and Konôpková, 1995).

Results

The ecology and production of exotic woody plants is an issue studied by the researchers in the "Arboretum Mlyňany" - Institute of Dendrobiology SAS since 1966. Their activities began with an evaluation of growth and production performance of selected 59 exotic taxa (49 conifers, 10 broadleaves) in 298 parks and dendrological subjects in Slovakia. The results of this survey, useful for orchard management and, principally, for forest practices are in the works TOKÁR (1976, 1979). These results not only justify and confirm the success of introduction of these woody plants into our climatic conditions, mainly in terms of growth and production, but they also represent a new knowledge concerning fructification, natural regeneration and other important features (also concerning orchards - such as habitus). The results should be reputed as a valuable source of scientific knowledge about the gene pool of the cultural dendroflora in Slovakia waiting for use in dispersion, protection and saving of these taxa.

Beginning with 1976, the ecological-production research was oriented on valorisation of forest stands of exotic woody plants in the region Malé Karpaty Mts (in frame of the programme Man and Biosphere) and on phytotechnique of young forest stands of selected exotic woody plants (*Castanea sativa* Mill., *Quercus rubra* L. and *Juglans nigra* L.) on four PRP series (Žirany, Lefantovce, Ivanka pri Nitre and Sikenica).

In the region Malé Karpaty Mts (TOKAR, 1985, 1991b), exotic woody plants are grown on 2,270 ha of the actual forest area and 1,579 ha of the reduced forest area. The major part concerns *Pinus nigra* Arnold (2,212 ha actual forest area and 1,533 ha reduced forest area). Many minor proportions concern *Pinus strobus* L. (1.78 ha actual forest area), *Pseudotsuga menziesii* (Mirbel) Franco (26.07 ha), *Aesculus hippocastanum* L. (5.36 ha), *Castanea sativa* Mill. (17.53 ha), *Quercus rubra* L. (5.30 ha) and *Juglans nigra* L. (1.91 ha).

As for the ecology, in the Malé Karpaty Mts, the exotic woody plants are most abundant in the groups of forest types (GFT): Fageto-Quercetum (892 ha actual forest area), Corneto-Quercetum (490 ha), Fagetum pauper (302 ha) and Querceto-Fagetum (169 ha). From the viewpoint of age, the first age class of 1–10 years (597 ha) and the sixth class encompassing 51–60 years (585 ha) are dominant.

Black pine is mostly grown in the gft-s Fageto-Quercetum (867 ha), Corneto-Quercetum (490 ha), Fagetum pauper (295 ha), Corneto-Fagetum (156 ha), and Querceto-Fagetum dealpinum (100 ha).

The results of the ecological-production analysis of the black pine in the Malé Karpaty Mts demonstrate that the most favourable conditions for growth and volume (mass) production in this region are in the gft-s Querceto-Fagetum, Fageto-Quercetum and Fagetum pauper. The biggest overall standing volume is in the mixed stands – with the black pine proportion less than 30% (younger than 100 years, in gft FQ 432 m³ ha⁻¹, in QF 464 m³ ha⁻¹ and in Fp 443 m³ ha⁻¹). In the pure stands, the highest standing volume was found in the gft-s Querceto-Fagetum (310 m³ ha⁻¹) and Fageto-Quercetum (295 m³ha⁻¹). The mixed stands had by from 10% (Corneto-Quercetum) to 64% (Fagetum pauper) more volume stock than the pure stands.

The results documenting the influence of phytotechnique on volume and mass production in the homogeneous stands of *Castanea sativa* Mill. on the PRP series Žirany (Table 1, Figs 1–2) show that a stronger positive influence during the whole stand growth (years 1972–2001) was obtained in heavy thinning from above applied after each 10 years. The total mean increment in the 46-year-old trees was from 19.4 to 23.9 m³ ha⁻¹ year⁻¹ and from 10.5 to 13.3 t ha⁻¹ year⁻¹ (TOKÁR 1998, 2002).

The phytotechnique (moderate thinning from above with positive selection and 5-year interval of repetition) in the pure (Fig. 3) and mixed stands of *Castanea sativa* Mill. on the PRP series Lefantovce (Table 2) resulted in better volume and mass production in the mixed (*Castanea sativa* Mill. + *Tilia cordata* Mill. (Fig. 4), *Castanea sativa* Mill. + *Pinus sylvestris* L.) stands than in the pure *Castanea sativa* Mill. stands (in volume by 5.31%-34.3%, in mass by 10.53%-31.60%). The cause underlying the better production in the mixed stands compared to the pure ones should be assigned to the favourable allopathic relations and soil conditions developed in these stands. The total mean increments in the 38-year-old trees were from 14.42 to $19.17 \text{ m}^3 \text{ha}^{-1}$ year⁻¹ and from 7.23 to $9.79 \text{ th}^{-1} \text{ year}^{-1}$ (TOKÁR, 2002; TOKÁR and KREKULOVÁ, 2003, 2004).

Table 1. Volume and mass production in the pure stands of *Castanea sativa* Mill. on the PRP series Žirany in 2001 (stand age 46 years)

	Thinning		Growin	g stock	Total pro	oduction
PRP	degree	Interval	$m^3 ha^{-1}$	t ha ⁻¹	$m^3 ha^{-1}$	t ha ⁻¹
Ι	Moderate	5	536	271	939	493
II	Moderate	10	566	260	894	486
III	Heavy	10	749	372	1,102	612
IV	Moderate	10	621	292	888	487
V	Heavy	10	755	384	1,094	597
VI	Control		676	325	876	483
VII	Heavy	10	634	331	965	541

Fig. 1. Stem of a high-quality edible chestnut (*Castanea* sativa Mill.) tree on the PRP Žirany (photo F. Tokár).

Fig. 2. Homogeneous stand of edible chestnut (*Castanea* sativa Mill.) on the PRP Žirany (photo F. Tokár).

Table 2. Volume and mass production in various stand types
of Castanea sativa Mill. on the PRP series Lefantov-
ce in 2001 (stand age 38 years)

	g .	Grov	wing ock	To prod	otal uction
Partial PRP	Species	m ³ ha ⁻¹	t	m^3	t ha ⁻¹
			ha^{-1}	ha ⁻¹	
I (Control)	<i>Castanea sativa</i> Mill.	356	174	548	292
Π	<i>Castanea sativa</i> Mill.	336	152	556	282
III (Control)	Castanea sativa Mill	195	88	411	211
	Tilia cordata Mill	221	94	325	149
	Together	416	182	736	360
IV	Castanea sativa	261	129	426	232
	Mill.				
	Tilia cordata	150	61	209	101
	Mill.				
	Together	411	190	635	333

V	Castanea sativa	214	105	358	204
(Control)	Mill.				
	Pinus sylvestris	220	124	316	173
	L.				
	Together	434	229	674	377
VI	Castanea sativa	200	95	322	186
	Mill.				
	Pinus sylvestris	171	73	263	130
	L.				
	Together	371	168	585	316

Fig. 3. Homogeneous stand of edible chestnut (*Castanea sativa* Mill) on the PRP Lefantovce (photo F. Tokár).

Valuable ecological-production results were attained in the Castanetarium Horné Lefantovce (14.38 ha) by improving growth and production of 86 seed progenies of edible chestnut from 12 localities in Slovakia (BENČAŤ and TOKÁR, 1978). In a tree age of 35 years, very good results were obtained in 15 seed progenies (TOKÁR, 2003). The production and resistance potential has been evaluated in TOKÁR et al., 2004.

The results of assessment of soils and phytocoenoses in the Castanetarium Horné Lefantovce and in the Castanetarium Jelenec showed that the edible chestnut was an important factor causing changes in the phyto-

Fig. 4. Thinning PRP of edible chestnut and small-leaf linden on the PRP Lefantovce (photo F. Tokár).

coenoses. The phytocoenoses in these localities belong into the 3rd forest vegetation tier, the group of forest types Fagetum pauper inferiora (TOKÁR and KUKLA 2005, 2006).

In the pure (Fig. 5) and mixed stands of *Quercus rubra* L. and *Juglans nigra* L. on the PRP series Ivanka pri Nitre (Table 3), the overall production was most effectively controlled by moderate thinning from above

with positive selection and repetition interval of 5 years in the mixed stands of *Juglans nigra* L. and *Quercus rubra* L. or *Tilia cordata* Mill. (Fig. 7). The overall mean increments in the trees aged 48 years were from 12.76 to 16.29 m³ha⁻¹year⁻¹ and from 8.16 to 11.54 t ha⁻¹year⁻¹ (TOKÁR 1991a, 1998, 2005).

In the pure stands of *Juglans nigra* L. on the PRP series Sikenica (Table 4, Fig. 6), stronger posi-

Partial PRP	Species	Proportion	Age Growing stock Total production		Growing stock		oduction
		[%]	[years]	m ³ ha ⁻¹	t ha ⁻¹	$m^3 ha^{-1}$	t ha ⁻¹
Ι	Quercus rubra L.	20	49	32	24	61	48
	Juglans nigra L	80	48	402	343	552	421
	Together	100		434	367	613	469
II	Quercus rubra L.	100	49	438	263	662	410
III	Quercus rubra L.	80	49	304	216	460	331
	Juglans nigra L.	20	48	175	125	242	162
	Together	100		479	341	702	493
IV	Juglans nigra L.	100	47	430	320	630	416
V	Juglans nigra L.	20	46	369	258	407	281
	Tilia cordata Mill.	80	42	132	56	180	86
	Together	100		501	314	581	367
VI (Control)	Quercus rubra L.	80	49	426	293	505	355
	Juglans nigra L.	20	48	261	194	287	206
	Together	100		687	487	792	561

Table 3. Volume and mass production of various stand types of *Quercus rubra* L. and *Juglans nigra* L on the PRP series Sikenica in 2003 (stand age 48 years)

Fig. 5. Homogeneous stand of red oak (*Quercus rubra* L.) on the PRP Ivanka pri Nitre (photo F. Tokár).

Fig. 7. Mixed stand of blacknut with small-leaf linden on the PRP Ivanka pri Nitre (photo F. Tokár).

tive impacts on the overall volume and mass production in years 1979–2003 were found for heavy thinning from above with positive selection and 5-year interval of repetition. The overall mean increments in the 64-year-old trees were from 7.22 to 8.95 m³ ha⁻¹ year⁻¹ and from 3.31 to 3.84 t ha⁻¹ year⁻¹ (TOKÁR 1992, 1998; TOKÁR and KREKULOVÁ 2005).

Table 4. Volume and mass production in the pure stands ofJuglans nigra L. on the PRP series Sikenica in 2003(stand age 64 years)

PRP	Thinning	Growin	g stock	Total production	
	degree	$m^3 ha^{-1}$	t ha ⁻¹	$m^3 ha^{-1}$	t ha ⁻¹
III	Moderate	381	173	468	215
IV	Heavy	464	195	573	246
V	Control	454	208	462	212

Fig. 6. Homogeneous stand of blacknut (*Juglans nigra* L.) with natural regeneration on the PRP Sikenica (photo F. Tokár).

On all PRP series, we used thinning methods focused on tending promising trees, selected from the trees with suitable quantitative and qualitative parameters (TOKÁR, 1987, 1998).

The content of elements in aboveground biomass and in soil in the forest stands composed of exotic woody plants varied with the plant species and the biomass compartment (e.g. Ca bark, stem; K leaves, Na stem xylem) (TOKÁR and KONÔPKOVÁ, 1995).

In the forests of Slovakia (primarily in southern areas of gft Carpineto-Quercetum), black locust (*Robinia pseudoacacia* L.) – one of the first woody plants introduced to Europe, has a specific status. Today the black locust forest stands represent about 34,000 ha, which is 1.87% of the total forest land area in Slovakia. The black locust production in forest stands in SW Slovakia was evaluated by BENČAŤ (1988).

The destructive method used (sample trees) for assessment of aboveground biomass production in model stands of exotic woody plants was also suitable for deriving eco-physiological characteristics of these stands and woody plants (leaf area index – LAI, biomass production per leaf area unit, and similar) (TOKÁR 1987, 1998; KONÔPKOVÁ, 2003; KMEŤ and ŠALGOVIČOVÁ, 2003; ŠALGOVIČOVÁ and KMEŤ, 2004).

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Received December 12, 2012 Accepted May 27, 2013

Short communication

Preservation and restoration of living plant collections on the example of the Buda Arboretum of Corvinus University, Budapest

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Abstract

SCHMIDT, G., SÜTÖRI-DIÓSZEGI, M. 2013. Preservation and restoration of living plant collections on the example of the Buda Arboretum of Corvinus University, Budapest. *Folia oecol.*, 40: 290–294.

The Buda Arboretum was initiated in the winter of 1893/94. Now it covers 7.5 hectares and is surrounded by the constantly growing city of Budapest. At present, the Arboretum is under very strong urban effect. Within the framework of a EU-project "Preservation and Restoration of Living Plant Collections and Historical Gardens" the Buda Arboretum was profoundly reconstructed and developed between 2010–2012. There were reconstructed selected objects serving to special purposes, e.g.: 1. Special biotypes (garden pond and the surrounding wetland, rock-gardens, pergolas for the climbing plants; a retaining wall giving shelter for the Mediterranean collections; greenhouse as a biotope for tropical and subtropical plants), 2. The historic geometrical garden part (called Parade Square), 3. Ecological solutions for water supply, 4. Suppression of invasive species and development of *Laurocerasus, Malus, Potentilla, Prunus, Syringa* collections, wetland-perennials, collection of Hungarian bred woody ornamentals introduction and trial of new Mediterranean species, etc. After reconstruction, the plant material includes over 1,900 woody species and cultivars, more than 240 kinds of bulb-flowers, 500 different perennials, 250 annuals and round about 300 tropical and subtropical (greenhouse) taxa.

Keywords

arboretum, Buda Arboretum of Corvinus University of Budapest, draught- and pollution-tolerance, global warming-up, heat-tolerance, woody ornamentals

Introduction and review of literature

The Buda Arboretum is one of the richest plant collections in Hungary. It was initiated in the winter of 1893/94 on 3 hectares, on the premises of the Horticultural School (the predecessor of the present Faculty of Horticultural Sciences) (RADE, 1943). The other parts of the territory were utilized by orchards, vineyards, and glasshouses for ornamental plants and vegetables, according to the profile of the School. Later, the fruit- and vine-plantations and the glasshouses were moved to the outskirts of the city and the whole site was reverted to an arboretum. The different steps of the process were described in works of SCHMIDT (1994), ZALAINÉ (2003), PROBOCSKAI (1994), HÁMORI and SCHMIDT (2003).

Now it covers 7.5 hectares and is surrounded by the constantly growing city of Budapest. The site is situated on the southern foothill of the 235 m high Hill of Gellért. The original vegetation was probably a mixed carstwood forest (Ceraso mahaleb-Quercetum and Orno-Quercetum), with some elements of mixed floodplain hardwood forest (Fraxino pannonicae-Ulmetum) (FACSAR, 2008). At present, the Arboretum is under very strong urban effect: the summer is hot, the winter is mild, the air is polluted. The Buda Arboretum has been protected by law as a natural reserve (living gene collection of woody plants) since 1974 and also as a historical garden since 2005 (CSEPELY-KNORR and SÁROSPATAKI, 2009).

The collections serve three main purposes: 1) education of students and public (a "living textbook"); 2) display of Hungarian-bred woody ornamental cultivars, and 3) testing, examination and trying out of plants of subtropical and Mediterranean origin in order to show the possible benefits of urban microclimate and also as potential plant materials for the case of global warming (SCHMIDT, 2008). In 2010, a considerable EU-fund was earned (Project No KMOP-3.2.1/B-09-2009-0003) for the reconstruction and the development of the Arboretum. The first publications reporting on the funding and the preliminary results were published in Hungarian language by SCHMIDT and SÜTÖRI-DIÓSZEGI, M., 2011; HONFI et al., 2012a; HONFI et al., 2012b; SCHMIDT and SÜTÖRI-DIÓSZEGI, 2010. The head of project management was prof. Károly Hrotkó, the head of the reconstruction and planting was prof. Gábor Schmidt, the coordinators were dr. Peter Honfi and dr. Magdolna Sütöri-Diószegi.

Materials and methods

The reconstruction-project started on 1 June 2010 and ended on 31 March 2012.

The main parts (sub-projects) of the project were as follows:

- 1. Reconstruction of special biotypes
- 2. Reconstruction of a historical geometric part of the garden
- 3. Ecological solutions for heating and for water supply
- 4. Suppression of invasive species in the hardy plant collections.

Each of the mentioned elements needed different approach and methods. For the sake of simplicity, these methods will be described in the next chapter only.

Results and discussion

The results (and also the lessons) of the reconstruction project are as follows (see also the Figs 1–6).

1. Reconstruction of special biotypes

Because of the limited space, the present paper will concentrate mainly on the most important woody plant collections. The biotypes of herbaceous collection and those for minor woody collections will be shortly mentioned only, and illustrated by some photos. Such are: 1.1. Wetland biotypes (the garden pond and the surrounding artificial wetland, Fig. 1.); 1.2. Dry carstland-

and humid alpine biotypes (rock-gardens with collections from plants of dry native hills and also true alpine plants) were reconstructed on 1,400 m²; 1.3. Pergolas for the climbing plants were reconstructed on 240 m² (Fig. 2.); and 1.4. South-facing retaining wall as a biotope for the open-ground Mediterranean woody plant collections. A retaining wall giving shelter from the north, is extremely dry and warm and offers excellent conditions for true Mediterranean plants like cypresses (Cupressuses), Yuccas, pomegranates (Punica granatum L.), hardy cactuses (Cactaceae Ivss) and others. The Albizia julibrissin (Willd.) Durazz. tree brings a profusion of soft pink mimosa-like flowers from July through September. Also here grow specimens of the bead-tree (Melia azedarach L.) and the holly oak (Quercus ilex L.). Before reconstruction the wall was partially ruined and dangerous for life. After reconstruction it became safe and the area for Mediterranean collection increased by 600 m² (list of plants see later).

Fig. 1. Garden pond.

Fig. 2. Pergola for the climbing plants.

2. Reconstruction of the historical geometric garden – part called Parade Square.

The Buda Arboretum is maintained as a natural plant protection and also is registered and protected as a historical garden. The most characteristic part of it is the 3,000 m² large geometrical garden section called Parade Square. In the past, the square was fully planted with herbaceous flower-beds as well as with roses – hence the name. Now that park-maintenance became too costly, the former baroque style is just symbolised by two symmetrically arranged groups of arborvitae (*Thuja* ssp.), the regular outlines of the lawn and by some adjacent bedding plants. The statue in the upper centre (in front of Building F) commemorates the famous fruit-breeder Máté Bereczki (Fig. 3).

Fig. 3. Parade Square

3. Environmental-friendly solutions for heating and for water supply

3.1 Energy-saving solutions for heating and cooling

The Arboretum contains a relatively small glasshouse (110 m^2) for the tropical and subtropical ornamental plants. The glasshouse is 20 years old and, before reconstruction, it was far outdated and in a very bad condition. The heating during the winter (with gas) needed a lot of energy and money, and the cooling in the summer was carried out with outdated heaters (pipes) and methods. Simply said, the air-conditioning was insufficient for the plants and yet, very expensive (Fig. 4).

3.2 Reutilization of run-off water from the roofs of the buildings

Several solutions were used for reutilization of run-off rainwater, the best of which are shown on Fig. 5.

Fig. 4. Greenhouse for tropical and subtropical plant collection.

Fig. 5. Collection of run-off rainwater from the roofs of the buildings.

4. Suppression of invasive species and development of hardy perennial and woody plant collections

4.1 Suppression of invasive species

This work included the regional removing of herbaceous weed and also the moving woody species, first of all: *Ailanthus altissima* (Mill.) Swingle, *Acer negundo* L., *Clamatis vitalba* L., *Fraxinus pennsylvanica* Marsh., *Parthenocissus quinquefolia* (L.) Planch., *Cotoneaster multiflorus* Bunge, *Diospyros lotus* L.

4.2 Development of hardy plant collections

4.2.1 Woody plant collection

Hibiscus collection: 26 taxa (Fig. 6 a); *Malus* collection: 42 taxa (Fig. 6 b); *Potentilla* collection: 38 taxa; *Prunus*

Fig. 6a. Hibiscus syriacus 'Minerva'.

laurocerasus L. collection: 34, other *Prunus* collections include the following sub-genera: *Amygdalus, Cerasus, Padus, Prunus*: 74 taxa; *Syringa* collection: 46 taxa.

Fig 6b. Malus 'Professor Sprenger'.

- Collection of Hungarian bred woody ornamentals: 74 taxa: Betula pendula Roth cv. Karaca, Buxus microphylla Sieb. et Zucc. cv. Betlér, Campsis × tagliabuana (Vis.) Rehd. cv. Galen Select, Campsis radicans (L.) Seem. ex Bureau cv. Barack, Chamaecyparis lawsoniana (A. Murray) Parl. cv. Tekeres, Cotoneaster salicifolius Franch. cv. Rózsaszín Füzér × Cupressocyparis notabilis (A. F. Mitchell.) Farjon cv. Márta, Hedera helix L. cvs. Arács, Balkon, Blue Star, Börzsöny, Csocsoszan, Duna, Krokó, Marble, Negro, Perint and Zebegény, Juniperus × media Van Melle cv. Mint Julep Tarka, Juniperus chinensis L. cvs. Eldorado, Favorit, Gold Rush, Juniperus communis L. cv. Fancsika, Juniperus conferta Parl. cv. Sláger, Juniperus sabina L. cvs. Báránd, Szőke Tisza and Tarka, Juniperus virginiana L. cvs. Golden Rain and Little Mityu, Picea pungens Engelm. cv. Edith, Pinus sylvestris L. cv. Sé, Prunus cerasifera Ehrh. cv. Colos, Prunus laurocerasus L. cvs. Ani, Antonius, Cipora, Cleopátra, Gabi, Hagar, Leander, Parviflora and Zita, Prunus padus L. cvs. 1/a sz. klón, 6 sz. klón, Aurora, Piros Oszlop (13. sz. klón) and Rózsaszín Május, Prunus persica cv. Orlóci Kiméra, Pyrus pyraster cv. Bihar, Rosa hybrids: cvs. Arany János, Bethlen Gábor, Házsongárd, Hild József, Máramaros, Nagyhagymás, Nyitra, Regéc, Szent Imre and Szent Margit, Salix matsudana Koidz. cv. Tarkabarka, Sorbus bakonyensis Jáv., S. borbasii Jáv. cv. Herkulesfürdő, S. borosiana Kárp. cv. Alba Regia, S. cv. Hainburg, *Taxus baccata* cv. Zöld, *Thuja occidentalis* L. cvs. Miki, Romantika, Szőllősi Klón (Malonyana Aurea), *Thuja orientalis* cvs. Dundi, Hunor, Jászkiséri, Lakatos and *Thuja orientalis* L. cv. Telihold, *Tilia platyphyllos* Scop. cv. Favorit, *Tilia platyphyllos* Scop. cv. Pannonia.

- Introduction and trial of new Mediterranean species: 61 taxa. In 2011–12, the following new tender species and cultivars were planted and tired: Acca sellowiana (O. Berg) Burret, Albizia julibrissin Durazz. cv. Summer Chocolate, Berberis darwini Hook.; Caesalpinia gilliesii (Wallich ex Hook.) Wallich ex D. Dietr.; Callistemon citrinus (Curtis) Skeels; Ceanothus delilianus Spach. cv. Gloire de Versailles; cv. Henri Defossé; Ceanothus pallidus Lindl. cvs. Marie Simon; Perle Rose, Cistus corbariensis Pourr.; C. pulverulentus Pourr. cv. Sunset; C. purpureus Lamn.; Cistus purpureus Lamn. cv. Alan Frad; Cordyline australis (Forst. f.) Hook. f.; Cotoneaster lacteus W.W.Sm.; Elaeagnus × ebbingei Boom ex Doorenb. cvs. Clône Erigé, Compacta, Eleador, Gilt Edge, Limelight; Escallonia cvs. Apple Blossom, Crimson Spire, Donard Seedling; Eucalyptus gunnii Hook f., Hebe arts; Itea virginica L. cv. Little Henry, Jasminum officinale L., Lagestroemia hybrids: cvs. Appalache, Hopi, Nivea, Pecos, Petite Pink, Rosea Nova, Rouge, Togo, Tonto, Lavandula angustifolia cvs. Hidcote, Munstead, Rosea; Lavandula × intermedia Loisel. cvs. Abrial, Chamallow, Edelweiss, Grosso, Imperial Gem; Leycesteria formosa Wall., Mahonia japonica Thunb.; Nandina domestica Thunb. cvs. Firepower, Richmond and Wood's Dwarf; Osmanthus heterophyllus (G. Don) P. S. Green cvs. Goshiki, Purpureus, Phormium tenax J. R. Forst. & G. Forst.; Photinia × fraseri Dress cvs. Camilvy, Nana, Pink Marble; Prunus lusitanica L.; Punica granatum L. cvs. Chico; Maxima Rubra, Teucrium fruticans L.; Viburnum tinus L. cvs. Eve Price and Gwenlian.

4.2.2 Perennial plant collections

Dryland perennials: from 60 to 75 taxa, wetland-perennials: from 20 to 70 taxa, alpine perennials: from 40 to 110 taxa.

Acknowledgement

The project was supported by KMOP-3.2.1/B-09-2009-0003 and TÁMOP-4-2.1.B-09/1/ KMR- 2010-0005 EU-project. The Authors would like to express their special thanks to the organizers of the excellent Conference in Mlyňany Arboretum (Sept. 18– 19., 2012) and also to Stephan Bakay for completing the Slovakian abstract of this paper.

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Received December 6, 2012 Accepted March 22, 2013

Survey paper

The Primeval Beech Forests of the Carpathians and Ancient Beech Forests of Germany: joint natural heritage of Europe

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Abstract

VOLOŠČUK, I., PICHLER, V., PICHLEROVÁ, M. 2013. The Primeval Beech Forests of the Carpathians and Ancient Beech Forests of Germany: joint natural heritage of Europe. *Folia oecol.*, 40: 295–303.

The European beech Fagus sylvatica L. ssp. sylvatica L. is exclusively found in Europe. The beech survived the last ice age in small refuges in the south and south-east Europe and went on the colonisation of large parts of the continent. The post ice colonization of the landscape by the beech took place parallel to the settlement of land by humans and the formation of a more complex society. For centuries much of the Carpathian mountain forests remained untouched. Virgin forests constitute a natural heritage of global significance. In 2007 the primeval beech forests of the Carpathians (Slovakia, Ukraine) were added to UNESCO's World Heritage List. On 25 June 2011, the UNESCO World Heritage Committee added five of Germany's beech forest regions to the World Heritage List. This extended the transboundary world natural heritage site "Primeval Beech Forest of the Carpathians", located in the Slovak Republic and Ukraine, to include a number of German forest regions, and renamed it "Primeval Beech Forests of the Carpathians and Ancient Beech Forests of Germany". The paper is aimed at the presentation of the outstanding universal value of the ecological processes in the Joint World Heritage Sites, and present principles of their Integrated Management Plan. Ultimate goal is to achieve that management and socio-economic sustainable development practices are in harmony with primary objectives of WHS protection, biodiversity conservation, ecosystem and landscape stability, rational use of natural resources, ecotourism development and with potential of the landscape in largest possible extend.

Keywords

ancient beech forest, Carpathy, Germany, primeval beech forest, World Heritage

Introduction

Europe's beech forests are deciduous forests which are dominated by the European Beech (*Fagus sylvatica* L.). The beech is endemic to Europe and beech forests are limited to Europe (GÖMÖRY et al., 2011). Such forests therefore share the fate of all deciduous forests of the northern hemisphere's nemoral zone. They have been exposed to an enormous development pressure (settlement, utilisation) for centuries so that natural forests have become scarce (BRITZ et al., 2009). Beech is one of the most important elements of forests in the Temperate Broad-leaf Forest Biome and represents an outstanding example of the re-colonisation and development of terrestrial ecosystems and communities after the last ice age, a process which is still ongoing (KNAPP, 2011). Forest communities built up and dominated by the beech are widespread across major parts of Central Europe. Potentially forming the predominant zonal vegetation in Western and Central Europe in terms of area, they are found at the montane level of the South European mountain ranges. They show the widest amplitude of soil trophic levels and altitude distribution, of all deciduous forests in Europe potentially occupying the largest area (BOHN and NEUHÄUSL, 2003).

The European beech forests stand out due to an exceptional variety of types. According BOHN and NEU-HÄUSL (2003), a total of 86 different biocoenotic units of the beech and mixed beech forests are found in the beech forest area, subdivided according to trophic and altitude levels as well as geographical and local forms. Of these units, 14 cover more than 50% of the potential natural range, with as many as eight units being also widespread in Germany with significant proportions of the overall area. A total of 28 biocoenotic units, which roughly equals one-third of all European units, are widespread in Germany, which emphasises Germany's particular responsibility for the preservation of the beech forests worldwide (BRITZ et al., 2009). The European beech forests show a decline in vascular plant species numbers from glacial refuges in Southern Europe to the north and northwest, in which directions they were advancing. Their centres of diversity lie in the Eastern Carpathians, the Dinaric Alps, and the Pyreneans (DIERSCHKE and BOHN, 2004). The particular evolutionary connection clearly reflects in the entire Central European Flora.

The different beech forest types are home to 20% of the terrestrial fauna in Central Europe – 7,000 to 10,000 animal species (Отто, 1994) that have mostly adapted their rhythm of life to the seasonal cycle. Alongside with the plants, fungi, and microorganisms, they are the determining factors in the beech forest system.

The history of the beech forests is closely linked with the history of European civilisation (BENNETT, 1994; BRITZ et al., 2009). The post-glacial colonisation of the landscape by the beech tree ran in paralel with the establishment of communities by mankind and the formation of more highly organised forms of society. That is why the beech is deeply rooted in European culture (PICHLER et al., 2007a).

Material and methods

The beech ecosystem research which has been the basis for elaborating on the World Heritage Nomination Project was carried out in two regions: in the Carpathian Mts and in the German Lowlands.

The complete ecological research of the mountain Primeval Beech Forests of the Carpathians started in the first half of the 20th century due to the famous Czech botanist and forest ecologist Professor Alois ZLATNÍK (ZLATNÍK, 1934, 1935, 1936; ZLATNÍK and HILITZER, 1932; ZLATNÍK et al., 1938). Valuable knowledge concerning ongoing ecological processes in the Carpathian primeval beech forest ecosystems has been obtained after Second World War during the past years (LEIBUND-GUT, 1978; JAWORSKI et al., 1994a, 1994b; KORPEĽ, 1989, 1995; KRICSFALUSY et al., 2001; COMMARMOT et al., 2000; BUBLINEC and PICHLER, 2001; SANIGA, 2011; SA-NIGA and SCHÜTZ, 2002; STOYKO et al., 1982; STOYKO and TASENKEVITCH, 1993; STOYKO, 2002; BRÄNDLI and DOW-HANYTSCH, 2003; VOLOŠČUK, 1992, 1994, 1995, 1999, 2003; HAMOR and COMMARMOT, 2005; COMMARMOT et al., 2000; PICHLER et al., 2007b) and utilized for practical forest and conservation management (VOLOŠČUK 1994, 1995; PICHLER et al., 2007a). The phytocoenological releves (stationary plots) were decribed according to ZLATNÍK (1976) and geobiocoenoses were classified according to ZLATNÍK (1959). In Primeval Beech Forests of the Carpathians prevail the group of forest types Fagetum pauper, Fagetum typicum, Fagetum tiliosum, Abieto-Fagetum and Fageto-Aceretum.

The ecological research in Ancient Beech Forests of Germany (lowlands) was carried out during the past 40–50 years (ASSMANN et al., 2008; DÖRFELT, 2008; PLACHTER et al., 2008; KNAPP, 2011; BRITZ et al., 2009).

Characteristics of the localities

The World Natural Heritage "Primeval Beech Forests of the Carpathians" is situated in the biogeographic region "Carpathian beech forests" (BRÄNDLI and DOWH-ANYTSCH, 2003) with a centre of diversity in the Eastern Carpathians. It is a part of the Inner Carpathians, which form a continuous mountain range over 1,300 km in length, 100 to 350 km in width, and up to 2,600 m in height. In the periphery and the montane-altomontane zone, large portions of this richly wooded mountain range are characterised by specious beech and mixed beech forests. The potential natural range of the beech forests therefore comprises an area of approx. 92,000 km² throughout the Carpathian centre zone, which corresponds to roughly one-tenth of the pan-European beech forest area. These areas, located in mountainous and sub-alpine altitudes (400-1,940 meters a.s.l.), are primarily representative of mountain beech forest. The geographic coordinates of Primeval Beech Forests of the Carpathians are: N 47°–49°, E 22°–24° (Table 1).

The last extensive primeval beech forests can now only be found in the Carpathians. This is the only place where there can still be experienced the uninterrupted dynamics of the coming and decline of beech forests since the last Ice Age. The great biodiversity of the

Component parts	Country	Core area ha	Buffer zone ha	Elevation a.s.l. m
Chornohora	Ukraine	2,476.8	12,925.0	640–1,550
Kuzyi-Trybushany	Ukraine	1,369,6	3,163.4	420-1,087
Maramorosh	Ukraine	2,243.6	6,230.4	600-1,470
Svydovets	Ukraine	3,030.5	5,639.5	720-1,500
Uholka – Shirokiy Luh	Ukraine	11,860.0	3,301.0	400-1,350
Stuzhitsia – Uzhok	Ukraine	2,532.0	3,615.0	600-1,221
Stužica – Buk. vrchy	Slovakia	2,960.0	11,300.0	512-1,210
Havešová	Slovakia	171.3	63.9	442- 741
Rožok	Slovakia	67.1	41.4	440- 789
Vihorlat	Slovakia	2,576.0	2,413.0	517-1,076
Total area ha		29,278.9	48,692.7	

Table 1. Location and area of the component parts of the Primeval Beech forests of the Carpathians

beech forests has managed to endure here. The World Heritage Site "Primeval Beech Forests of the Carpathians" represents the beech forest of the mountain range in ten component parts. Four areas are located in the Slovak Republic, six are located in the Ukraine. The smallest area is 67 hectares in size, the largest approx. 12,000 hectares. They are located in the Eastern Carpathians, one of the most unspoilt habitats in Europe. All the component parts are remnants of primeval forests which are embadded in beech forests that are extensively managed.

Germany is at the centre of distribution of the beech forests. If nature had its way they would cover approx. two thirds of the land area of Germany extending from the Alps over high and low mountains ranges and down to the lowlands at the sea coastlines. Now only approx. seven per cent of this surface is covered with beech forests due to deforestation and forest conversion. Larger contiguous forest areas are rare. The remaining forests are used in the forestry industry and beeches of approx. 120 years of age are harvested. The senescent and decay phases of a lifecycle that is naturally of more than 300 years duration are absent and thus also the living spaces that emerge in these phases as tree hollows and dead wood with their typical biocoenosis. Primeval beech forests have long since disappeared barring a few miniscule remnants and with them also species that are dependent upon them.

The Decision of the 35th Session of the World Heritage Committee, Paris 25 June 2011, approved the extension of the Primeval Beech Forests of the Carpathians (Slovakia and Ukraine), to include the Ancient Beech Forests of Germany, and becomes the Primeval Beech Forests of the Carpathians and the Ancient Beech Forests of Germany (Slovakia, Ukraine and Germany), on the basis of criterion (ix): outstanding examples representing significant on-going ecological and biological processes in the evolution and development of ecosystems and communities of plants and animals. The German extension in 2011 is another major step towards protecting this unique ecosystem for the long term.

The German part includes selected forest regions of the National Parks Hainich in Thuringia, Kellerwald-Edersee in Hesse, Jasmund and Müritz in Mecklenburg-Western Pomerania, and the forest of Grumsin in the Schorfheide-Chorin Biosphere Reserve in Brandenburg. These are the most valuable remaining examples of large, undisturbed beech forests in Germany. These German sites with their beech forests in the lowlands and central uplands are a perfect component to the mountain beech forests located in the Carpathians. This component part of the World Natural Heritage represents the characteristics and the natural processes of European beech forests under various ecological conditions.

The development history of beech forests since the Ice Age, the enormous competitiveness of beech *Fagus sylvatica* and the diversity of geographical, geological and ecological beech forest variations are a unique global phenomenon. The Ancient Beech Forests of Germany are indispensable to documenting the postglacial colonisation by *Fagus sylvatica* from south to north, from east to west, and spanning the entire spectrum of altitudinal zones from the sea-shore, to the lowlands and the submontane belt, to the upper timber line in the mountains (KNAPP, 2011). German's component parts are the most outstanding examples worldwide of the respective beech forest types. Each component part has its own specific characteristics and local peculiarities that make it unique and irreplaceable.

Jasmund: size 492.5 ha, buffer zone 2,510.5 ha, N 54°32′53′′ E 13°38′43′′ (0–131 a.s.l.). Jasmund is

a representative of the beech forest of the lowlands type. Half of Jasmund's property border follows to coastline. Although this border is subject to very slow natural dynamic changes based on the denudation of the steep coast, it is clearly identifiable by distinctive habitat limits at any given point. Jasmund represents the beech forests of the lowlands on lime and boulder clay. Beech forests, chalk cliffs and sea form a fascinating backdrop. The harsh coastal climate and the interaction of topography and climate lead to a broad range of different beech forest communities which are interspersed with streams and moors. Rare orchids, the great horsetail and the coral root are typical here.

Serrahn: size 268.1 ha, buffer zone 2,568.0 ha, N 53°20′24′′, E 13°11′52′′ (67–124 m a.s.l.). The best structured lowland beech forests in Europe. Demarcation in Serrahn has produced a compact core area of beech-dominated forests. In the Serrahn part the forest of the Müritz National Park lowland beech forests grow on sands from the Ice Age. In the midst of an extended forest and lake landscape this old beech forests help us to imagine what the German beech forests once looked like. Lakes and mires enrich the forest landscape, create a rich diversity of habitats and form the basis for a great amount of biodiversity. The beech forest of Serrahn is consequently documenting moisture-related distribution limits in an outstanding manner.

Grumsin: size 590.1 ha, buffer zone 274.3 ha, N 52°59'11", E 13°53'44" (76-139 m a.s.l.). Grumsin represents the beech forests of the lowlands on glacial sands and clay. The demarcation of the Grumsin component part largely follows the core area border of the Schorfheide-Chorin Biosphere Reserve, which was designated in 1990. Minor marginal zones which predominantly consist of pine woods rather than nearnatural deciduous forests and were likewise abandoned to natural development in 1990 have been assigned to the buffer zone. Water and forests are closely linked in Grumsin. Lakes, forest marches and moores in deep valleys interchange with marked ridges and conjure up atmospheric forest images in the ancient beech forests. These different structures in the most confined spaces form the basis for an exceptionally rich range of animal and plant species. The area represents an exceedingly textured young moraine landscapes with altitudes of between 60 and 140 m above sea level and all the typical elements in a unique fashion.

Hainich: size 1,573.4 ha, buffer zone 4,085.4 ha, N 51°04′43′′, E 10°26′08′′ (290–490 m a.s.l.). Hainich National Park encompasses what is, at present, the largest unmanaged deciduous forest area in Germany. Hainich represents the best reference area for the specious eutraphent beech forests of the European collinesubmontane zones with their ground vegetation rich in geophytes and the exceedingly attractive floral display in early spring, representing the seasonality of Central European deciduous forests in a unique manner. The most valuable beech forests that offer a very rich range of species grow on the central mountain ranges on limestone. It impresses through its extensive range of tree species and reveals lime beech forests of a magnitude, unspoilt nature and form that you will be unable to find in any other area. The demarcation in Hainich follows the distribution of the best-preserved beech forests with old growth stands. The buffer zone comprises the core area of the national park. The Hainich beech forest is unique proof of the currently ongoing ecological processes associated with the present climate change.

Kellerwald: size 1,467.1 ha, buffer zone 4,271.4 ha, N 51°08'43'', E 8°58'25'' (245–626 m a.s.l.). The acidophilous beech forests of the lower mountain ranges grow on slate and geywacke in the Kellerwald. No roads and no settlement cut through the exceptionally old, extensive forests of the Kellerwald in which unique primeval forest relics have survived. The beech reaches its natural forest boundary at the rocky and scree slopes and forms a bizarrely formed forest landscape. More of than 500 of the purest springs and streams form additional valuable habitats. In Kellerwald, the border was established taking into account the specific qualities of the component part, such as the high relief energy, the disjointed occurrence of small primeval-forest like steep slopes, and the spatial distribution of valuable beech forests. A coherent complex of valuable oldgrowth beech forests has been included. The demarcation of buffer zone follows the national park border. No buffer has been designated in a very small plot located on the northern border in order to integrate one of the primeval beech forest slopes into the property. Kellerwald contains the largest protected area of oligotraphent and mesotraphent beech forests, where undisturbed ecological and biological processes occur and is a perfect illustration of acidophilous beech forests.

Results and discussion

Specific peculiarities of the Carpathian forests include the richness in endemic species, the occurrence of Europe's largest population of predatory mammals with some 8,000 brown bears, 4,000 wolves and 3,000 lynxes as well as the most significant large-scale primary forest on the periphery of the European beech forests' distribution range. Representing its remaining primeval forests, the World Natural Heritage "Primeval Beech Forests of the Carpathians" is an essential part of these unique beech forests landscapes. These undisturbed, complex temperate forests exhibit the most complete and comprehensive ecological patterns and processes of pure stands of Fagus sylvatica across a variety of environmental conditions. The Carpathians Primeval Forests show a broad range of possible forest development stages from rejuvenation to decay (PICHLER et al., 2007a).

Ukraine and the Slovak Republic have taken on a pioneering role with the inscription of the Primeval Beech Forests of the Carpathians in the World Heritage List in 2007. The Carpathian Mountains are home to the last remaining large-scale primeval beech forests in Europe. Since the end of the last Ice Age, the forests here have been able to develop undisturbed. Mightly beech trees up to 50 meters high dominate the structurally rich forests (BRANDLI and DOWHANYTSCH, 2003; VOLOŠĆUK, 2003). The dynamics of the primeval beech forests, the natural comings and goings, are able to play out entirely free from anthropogenic influences here. Globally endangered species of fauna, fungi and flora have been able to preserve their natural gene pool.

The model of the main natural successional phases occuring in Central Europe (KORPEL 1995, PICHLER et al., 2007a): growing-up stage, optimal stage, decaying stage. In the growing-up stage, trees are found in all three layers - upper, middle and lower, and the crown closure is dense. As there is low mortality in trees of this age, there is little dead wood. At the end of phases, however, the competition between individuals is so great that strong dying off of juveniles occurs. In the following optimal stage, the maximum timber stock is reached, but the number of trees per area unit is low. With the lack of an understorey, the attainment of maximum height and a closed canopy, the forest in this phase is known as "hall-forest", being reminiscent of the interior of a cathedral or great hall, and also bears some resemblance to a commercial forest. During the transition to the decaying stage tree vitality decreases and the proportion of dead wood increases considerably. In this phase, the number and size of gaps between tree clusters increases and regeneration of climax tree species starts again.

An alternative view (HOLLING, 2001) suggests that the complexity of living systems of people and nature emerges not from a random association of a large number of interacting factors rather from a smaller number of controlling processes. These systems are self-organized, and a small set of critical processes create and maintain this self-organization. "Self-organization" is a term that characterizes the development of complex adaptive systems, in which multiple outcomes typically are possible depending on accidents of history. According to HOLLING (2001) there are three properties that shape the adaptive cycle and the future state of a system: wealth, controllability, and adaptive capacity. The adaptive cycle includes 4 phases: (1) long period of slow accumulation and transformation of resources, and (2) conservation (growing-up stage and optimum stage according to KORPEE, 1995), (3) shorter period of collapse that creates opportunities for (4) innovation (from release to reorganization), or decaying stage with regeneration phase according to KORPEL, 1995).

The European natural beech forests stand out due to a highly peculiar natural dynamism which is deter-

mined by the cycle of growth and decay of one single tree species, which is the beech. Old beech stands will regenerate with the crowns of individual trees gradually dying back to allow more light to the ground. Either there already is young beech wood that will now emerge, or the next generation of saplings will close the void within a period of a few years. The beech once again forms the upper crown canopy later on, thus resetting the cycle, which has been described as the small development cycle (Zukrigl et al. 1963, Leibundgut 1978, KORPEĽ 1989, 1995). In the wake of major disruptions, however, the cycle may also involve the formation of an early successional forest made up of pioneer species such as pines, birches, goat willows or rowans, which is later on infiltrated by medium-shade and shade tree species. This big successional cycle may take several decades longer than the small one. Variation incorporating elements of both big and small cycle are possible. This endogenous cycle of development meets the diversity of sites resulting from the glacial and postglacial periods, producing the considerable structural variety as basis for the species-rich, complex system. Rooted in the beech's enormous ecological plasticity, the high ecological stability results in a biodiversity-promoting continuity of the forest's character, which makes the dynamics of the beech forest persistently "predictable" for the forest dwellers. Old beech forests are, for example, home to a multitude of flightless ground beetles that would drop the ability to fly due to the habitat being continuously available or changing only at a small scale (BRITZ et al., 2009, PLACHTER et al., 2008).

A significant feature of the beech forests is decline in floristic diversity, which is a result of the history of flora and vegetation, from the former glacial refuges in Southern and Southeastern Europe up the northern and northwestern subterritories. Old beech trees can form a highly diverse habitat for fauna. The beech is a key species which creates its own internal forest climate and crucially influences soil formation, regeneration cycle, food chains and structures and reveals stonishingly specific diversity of plants, vertebrates, insects, molluscs and fungi. This diversity is described in terms of its ecological role in the ecological processes of beech forest ecosystems - trees and shrubs, mycorrhizae, geophytes, other herbaceous plants, lianas, herbivores, carnivores, dead wood inhabitants, destruens, etc. (ASSMANN et al., 2008).

As opposed to the climatic patterns of tropical rainforests, the climate of the temperate zone is distinguished by its seasonal changes together with the phenological floral cycle involved. From a physiognomic perspective, the most striking feature of deciduous trees is the fall of leaves, which will further accentuate the seasonal differences and conditions of the biotopes respectively. However, the foliage changing with the seasons does not take place abruptely. In pure beech forests this process is accompanied by unique changes in colour (KNAPP, 2011). The most dramatic consequence of leaf fall is the light climate's periodicity. This sets deciduous forests apart from all non-deciduous forest types, permitting the intermittent occurence of a herb layer that shows different specific adaptations. Spring geophytes exploiting the brief warm spring period prior to leafing for development are particularly well adapted and transform the soils of richer beech forests into a carpet of flowers. The association that has given rise to geophyte-rich beech forests is a result of ecosystemary continuity as well as the inner functional and structural differentiation of the development cycle of deciduous forests. In this particular shape, it is without paralel in the world (KNAPP, 2011).

A multitude of fungi are involved in dead wood decomposition, with a number of species being specialised in the metabolisation of specific wood types. The species of the genus Fagus are highly mycotrophic; in other words, much of their nutrient supply comes from fungi. Their survival is directly dependent on the mycobionts of ectotrophic mycorrhizae. The dominant mycorrhizal fungi associated with Fagus sylvatica are Agaricomycetidae, a subclass of Basidiomycetes (Homobasidiomycetes) from the genera Amanita, Boletus, Cortinarius, Inocybe, Laccaria, Lactarius, Tricholoma, Russula and Xerocomus. Soil acidity plays an important role in relation to the species spectrum of the mycorrhizal partners of Fagus sylvatica (Dörfelt, 2008). Species typical of the beech include Fomes fomentarius (wood-inhabiting fungi), Ganoderma applanatum (wood decaying), Neobulgaria pura, Oudemansiella mucida, which is indicative of extensive matured wood pools, and Hericium coralloides, which, although widespread throughout the northern hemisphere and also growing on other trees, is only found in very old, mature beech forests. Dead beechwood is colonised very swiftly by very many lignocolous fungi. Three phases characterise decomposition of beech stups: initial, optimal and final phase. There are more than 10 parasitic biotrophic fungi which infect Europe's beeches. A very large number of fungi are involved in the decomposition of fallen beech leaves, fruits, mast (cupulae) and twigs (Dörfelt, 2008). An especially important symbiosis has been evolved between fungi and plants in the rhizosphere, which is called mycorrhiza. Forests of the temperate zone are home to fungi that will enter into specific symbioses with one or few tree species (BRITZ et al., 2009).

Despite the beech's absolute dominance, the beech forests show outstanding diversification and are unique in function and structure (PICHLER et al., 2007a). Nortwithstanding the geologically short time of a few thousand years, a highly characteristic faunistic biocoenosis has been evolved postglacially which is just a globally unique as is the plant community. The fauna can exist in all its diversity, and the postglacial evolutionary processes can take place only if each forest development stage of the natural regeneration cycle is available – which is the case in the Primeval Beech Forests of the Carpathians (PICHLER, 2007; PLACHTER et al., 2008).

The Principles of Joint Management Plan

Long-term protection and management of the World Heritage Sites is ensured through national legal protection as national parks or core areas of a biosphere reserve. Effective implementation of the integrated management plan and the trilateral integrated management system is required to guide the planning and management of this World Heritage Sites.

The general objectives of the Integrated Management Plan are (PICHLER et al., 2007a):

- To ensure the most effective conservation of the WHS properties with all their abiotic and biotic components, geo- and biodiversity and ecological processes. To secure a lasting homeostasis and selfreproduction of the respective ecosystems and their protection both against anthropogenic factors.
- To maintain and expand the existing, ecologically connected complex of primeval and natural beech forests that encompass and connect (link) the WHS on both the Slovak and the Ukrainian sides – within the corridors connecting the WHS. Supporting the succession of managed beech semi-natural forests.
- To use WHS for scientific research in order acquire knowledge transferable and applicable on the level of sustainable. To use WHS for enhancement of landscape ecological stability.
- To use WHS for enhancement of ecological and environmental education, awareness of primeval forests chosen to maintain integrity and conservation of the existing sites, to preserve their naturalness and uniqueness.
- To support of traditional crafts, products and ecotourism.

Common elements of an effective management system could include: a) a thoroughly shared understanding of the property by all stakeholders; b) a cycle of planning, implementation, monitoring, evaluation and feedback; c) the involvement of partners and stakeholders; d) the allocation of necessary resources; e) capacity-building; and f) an accountable, transparent description of how the management system functions.

Conclusions

Joint World Natural Heritage "The Primeval Beech Forests of the Carpathians and the Ancient Beech Forests of Germany" is indispensable to understanding the history and evolution of the genus *Fagus*, which, given its wide distribution in the Northern Hemisphere and its ecological importance, is globally significant. These undisturbed, complex temperate forests exhibit the
most complete and comprehensive ecological patterns and processes of pure stands of European beech across a variety of environmental conditions and represent all altitudinal zones from seashore up to the forest line in the mountains. Beech is one of the most important elements of forests in the Temperate Broad-leaf Forest Biome and represents an outstanding example of the re-colonisation and development of terrestrial ecosystems and communities after the last ice age, a process which is still ongoing. They represent key aspects of processes essential for the long term conservation of natural beech forests and illustrate how one single tree species came to absolute dominance across a variety of environmental parameters.

Furthermore, it is not enough for a site to meet the World Heritage criteria, but it must also meet the conditions of integrity and/or authenticity and must have an adequate protection and management system to ensure its safeguarding.

Acknowledgement

This study was supported by the grant from the Slovak Grant Agency for Science VEGA no. 1/0364/10 and no. 1/0252/11.

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> Received December 17, 2012 Accepted February 14, 2013

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Book

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