

ACTA SILVATICA  
&  
LIGNARIA  
HUNGARICA



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HUNGARICA

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# ACTA SILVATICA ET LIGNARIA HUNGARICA

## Vol. 11, Nr. 1

### Contents

EREDICS, Attila – NÉMETH, Zsolt István – RÁKOSA, Rita – RASZTOVITS, Ervin – MÓRICZ, Norbert – VIG, Péter: The Effect of Soil Moisture on the Reflectance Spectra Correlations in Beech and Sessile Oak Foliage .....	9
SÁRÁNDI-KOVÁCS, Judit – LAKATOS, Ferenc – SZABÓ, Ilona: Post-epidemic Situation of a Previously <i>Phytophthora alni</i> -infected Common Alder Stand .....	27
PRIMUSZ, Péter – PÉTERFALVI, József – MARKÓ, Gergely – TÓTH, Csaba: Effect of Pavement Stiffness on the Shape of Deflection Bowl .....	39
FÜZESI, István – HEIL, Bálint – KOVÁCS, Gábor: Effects of Wood Ash on the Chemical Properties of Soil and Crop Vitality in Small Plot Experiments .....	55
LEBOCKÝ, Tibor – PETRÁŠ, Rudolf: Methods of Observation of Wild Boars Influence on the Growth of Forest Trees and Stands: a Case Study Based on Data from the Wild Boar Game Preserve .....	65
DELIISKI, Nencho – BREZIN, Veselin – TUMBARKOVA, Natalia: Modelling of the 1D Convective Heat Exchange between Logs Subjected to Freezing and to Subsequent Defrosting and the Surrounding Environment .....	77

# ACTA SILVATICA ET LIGNARIA HUNGARICA

## Vol. 11, Nr. 2

### Contents

SZŐCS, Levente – MELIKA, George – THURÓCZY, Csaba – CSÓKA, György: Parasitoid complex of leafminer <i>Pyllonorycter comparella</i> (Lepidoptera, Gracillariidae) in Hungary .....	91
NOWINSZKY, László – PUSKÁS, János: Sex Ratio Analysis of Some Macrolepidoptera Species Collected by Hungarian Forestry Light-Traps .....	99
FOLCZ, Ádám – BÖRCsök, Zoltán: Macrofungi in the Botanical Garden of the University of West Hungary, Sopron .....	111
KUČERA, Peter: On the Occurrence of Natural Norway spruce Woodland in the Pieniny Mts (Western Carpathians) .....	123
KESERŰ, Zsolt – BALLA, Ildikó – ANTAL, Borbála – RÉDEI, Károly: Micropropagation of Leuce-poplars and evaluation of their development under sandy site conditions in Hungary .....	139
VIZVÁRI, Zoltán – KISS, Tibor – MÁTHÉ, Kálmán – ODRY, Péter – VÉR, Csaba – DIVOS, Ferenc: Multi-Frequency Electrical Impedance Measurement on a Wooden Disc Sample .....	153
<b>Guide for Authors</b> .....	163
<b>Contents and Abstracts of Bulletin of Forestry Science, Vol. 5, 2014</b> The full papers can be found and downloaded in pdf format from the journal's webpage ( <a href="http://www.erdtudkoz.hu">www.erdtudkoz.hu</a> ) .....	165

# ACTA SILVATICA ET LIGNARIA HUNGARICA

## Vol. 11, Nr. 1

### Tartalomjegyzék

EREDICS Attila – NÉMETH Zsolt István – RÁKOSA Rita – RASZTOVITS Ervin – MÓRICZ, Norbert – VIG, Péter: Talajnedvesség hatása bükk és kocsánytalan tölgy lombzat reflexiós spektrumainak korrelációira .....	9
SÁRÁNDI-KOVÁCS Judit – LAKATOS Ferenc – SZABÓ Ilona: Egy korábban <i>Phytophthora alni</i> által fertőzött mézgáséger-állomány járvány utáni állapota .....	27
PRIMUSZ, Péter – PÉTERFALVI, József – MARKÓ, Gergely – TÓTH, Csaba: A pályaszerkezet merevségének hatása a behajlási teknő alakjára .....	39
FÜZESI, István – HEIL, Bálint – KOVÁCS, Gábor: Fahamu hatása a talaj kémiai jellemzőire és a termés vitalitására egy kisparcellás kísérletben .....	55
LEBOCKÝ, Tibor – PETRÁŠ, Rudolf: A vaddisznó hatása a fák és a faállomány növekedésére – egy vaddisznóskert esettanulmánya .....	65
DELIISKI, Nencho – BREZIN, Veselin – TUMBARKOVA, Natalia: Egydimenziós konvektív hővezetés modellezése fagyott és normál állapotú rönk és környezte között .....	77

**Vol. 11, Nr. 2**

SZŐCS Levente – MELIKA, George – THURÓCZY Csaba – CSÓKA György: A <i>Populus alba</i> -n élő <i>Phyllonorycter comparella</i> (Lepidoptera: Gracillariidae) levélaknázó magyarországi parazitoid együttese	91
NOWINSZKY László – PUSKÁS János: A magyar erdészeti fénycsapdák által gyűjtött néhány macrolepidoptera faj ivararányának elemzése	99
FOLCZ Ádám – BÖRCsök Zoltán: Nagygombák a Nyugat-magyarországi Egyetem Soproni Botanikus Kertjében	111
KUČERA, Peter: A közönséges luc természetes állománya a Pieninekben (Nyugati-Kárpátok)	123
KESERŰ Zsolt – BALLA Ildikó – ANTAL Borbála – RÉDEI Károly: Leuce-nyárok mikroszaporítása és homokterületen történő termesztésük kiértékelése Magyarországon	139
VIZVÁRI Zoltán – KISS Tibor – MÁTHÉ Kálmán – ODRY Péter – VÉR Csaba – DIVOS, Ferenc: Fakorongok multi-frekvenciás elektromos impedancia mérése	153
<b>Szerzői útmutató</b>	163
<b>Erdészettudományi Közlemények 2015. évi kötetének tartalma és a tudományos cikkek angol nyelvű kivonata</b> A tanulmányok teljes terjedelemben letölthetők pdf formátumban a kiadvány honlapjáról ( <a href="http://www.erdtudkoz.hu">www.erdtudkoz.hu</a> )	165

## The Effect of Soil Moisture on the Reflectance Spectra Correlations in Beech and Sessile Oak Foliage

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**Abstract** – Reflectance intensities of foliage are mostly due to biomaterials synthesised by plants. Adaptation to the continuously changing environment requires the regulated alteration of metabolic processes, which also influences the UV-VIS (Ultraviolet-Visible) and IR (Infra Red) spectra of leaves. For the calculation of various Vegetation Indices (VIs), e.g. NDVI (Normalized Difference Vegetation Index), the common practice is to use the reflectance spectrum of the whole foliage and when individual leaves of the same plant are sampled, an average VI is derived. On the contrary, our method exploits the small differences between individual leaves of the same plant, making use of the similar distributions of measured reflectance values. Using particular wavelength pairs, linear regressions of reflectance intensities have been investigated. The parameters of these regressions (slope and intercept) have been compared to the temporal variations of the environmental factors, such as temperature, vapour pressure deficit and soil moisture. By assessing the *sensitivity* of the regression coefficient (slope) to the changing environment, wavelength pairs can be selected whose *sensitivity* change reflects the effect of soil moisture deficit on the plant. Based on the state-dependent correlations of the reflectance spectra of plant foliage, a new concept is presented that is capable of indicating the level of environmental stress, e.g. drought stress.

**drought stress / state-dependent correlation / UV-VIS and IR spectrometry**

**Kivonat – Talajnedvesség hatása bükk és kocsánytalan tölgy lombzat reflexiós spektrumainak korrelációira.** A lombzat reflexiós intenzitásainak változását elsősorban a növény által előállított anyagok okozzák. A folyamatosan változó környezethez való alkalmazkodás megköveteli az anyagcsere-folyamatok szabályozott megváltoztatását, ami befolyásolja a levelek reflexiós spektrumát is az UV, látható és közeli infravörös tartományban. Különböző Vegetációs Indexek (VI) számításánál, mint pl. NDVI (Normalized Difference Vegetation Index), az általános gyakorlat szerint a teljes lombzat reflexiós spektrumát használják, vagy ha ugyanazon növény különböző leveleit mintázták, akkor egy átlagos VI-t származtatnak. Ezzel szemben módszerünk éppen a növény egyes levelei közötti kis különbségeket hasznosítja, kihasználva a mért reflexiós értékek hasonló eloszlását. Bizonyos hullámhosszpárokat kiválasztva, a mért reflexiós intenzitások lineáris regresszióit vizsgáltuk. A regressziós paramétereket (meredekség és tengelymetszet) összehasonlítottuk a környezeti hatások időbeli változásával, mint pl. hőmérséklet, légköri telítési hiány és talajnedvesség. A regressziós paraméter (meredekség) *érzékenysége*nek vizsgálatával olyan hullámhosszpárokat lehet

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kiválasztani, melyek *érzékenység* változása tükrözi a talajnedvesség hiány növényre kifejtett hatását. Bemutatjuk az új módszer alkalmazhatóságát, amely a növényi lombzat reflexiós spektrumainak állapotfüggő korrelációján alapul, és amely képes a környezeti stressz, pl. szárazságstressz kimutatására.

## **szárazságstressz / állapotfüggő korreláció / UV-VIS és IR spektrometria**

### **1 INTRODUCTION**

Water is not only the media for metabolism, but also one of the key substrates in the cells. The effect of its deficit on various plant tissues and on the whole plant is one of the most studied phenomena in plant science. Recent publications about plant drought stress focused on the genetic background of drought resistance, the modifications of plant hormone systems and enzyme activities, the changing accumulation of biomaterials and the improvement of drought tolerance, among others. The test plants were usually various agricultural crops e.g. wheat, rice, soybeans, grapes and tomatoes. On the contrary, publications studying the effect of drought on trees, particularly by spectrometric methods, are less frequent (e.g. Filella – Peñuelas 1994, Peñuelas et al. 1997a,b, Pu et al. 2003).

The water status of trees can be measured by various methods, e.g. by leaf sampling and a pressure bomb (Scholander et al. 1965), stem psychrometer (Dixon – Tyree 1984) or the recently developed leaf patch clamp (Zimmermann et al. 2008). These methods are able to describe the instantaneous turgor pressure but reveal less about the long term effects of the drought stress, and may be very sensitive to the sampling time and conditions. On the other hand, the overall effect of the unfavourable conditions may be estimated by analysing the bio-production of the plant, e.g. yield, growth or accumulated dry material, but it demands a longer period or a full growing season in order to obtain significant differences.

The measurement of the leaf reflectance spectrum is a very promising way to characterize drought stress, and many different methods have been proposed to interpret the reflectance data. Many “high spectral resolution reflectance vegetation indices” have been developed to monitor biomass, phenology or physiology (Peñuelas – Filella 1998). These methods usually measure the reflectance at a few selected wavelengths and calculate a Vegetation Index (VI) from these reflectance values.

For example, a Simple Ratio (*SR*) can be used to estimate green biomass:  $SR = R_{NIR} / R_{red}$ , where *R* is reflectance and *NIR* stands for near infrared (Jordan 1969). A more robust and widespread method is the Normalized Difference Vegetation Index (*NDVI*) which is calculated as:  $NDVI = (R_{NIR} - R_{red}) / (R_{NIR} + R_{red})$  and used not only at ground level but also by airborne or satellite remote sensing (Rouse et al. 1973). *NDVI* is mainly sensitive to the green biomass (Gamon et al. 1995), but decent correlations are found with many other environmental or physiological variables. The exact wavelengths of *SR* and *NDVI* are somewhat arbitrary, and vary according to the spectral sensitivity of the sensor that is used or the goal of the investigation. Many practical uses of the *NDVI* have already been proposed and also used in industry, e.g. irrigation scheduling (e.g. Hunsaker et al. 2007, Aldakheel 2011), soil salinity management (e.g. Li et al. 2014), yield prediction (e.g. Teal et al. 2006, Moriondo et al. 2007) or fertilizer dosage adjustment (e.g. Stone et al. 1996, Crain et al. 2012).

Not only red and near infrared light, but many other wavelengths have been investigated, and various other VIs have been proposed for different purposes. *Table 1* summarises the most frequently used VIs along with the wavelengths they use. Many studies have been conducted to compare different VIs for specific application purposes, e.g. stress detection (e.g. Zarco-Tejada et al. 2004, Eitel et al. 2007, Pu 2008).

Table 1. The most frequently used Vegetation Indices along with their used wavelengths.

Vegetation Index (VI)	Abbrev.	Wavelengths	Reference
Simple Ratio	SR	NIR, red	Jordan (1969)
Normalized Difference Vegetation Index	NDVI	NIR, red	Rouse et al. (1973)
Normalized Difference Pigment Index	NDPI	430, 680	Baret et al. (1988)
Modified Chlorophyll Absorption in Reflectance Index	MCARI	550, 670, 700 or 550, 670, 800	Daughtry et al. (2000) or Haboudane et al. (2004)
Transformed CARI	TCARI	550, 670, 700	Haboudane et al. (2002)
Triangular Vegetation Index	TVI	550, 670, 750	Broge - Leblanc (2001)
Modified Triangular Vegetation Index	MTVI	550, 670, 800	Haboudane et al. (2004)
Renormalized Difference Vegetation I.	RDVI	670, 800	Roujean - Breon, (1995)
Soil Adjusted Vegetation Index	SAVI	670, 800	Huete (1988)
Improved SAVI with self-adjustment f.	MSAVI	670, 800	Qi et al. (1994)
Optimized Soil-Adjusted Vegetation I.	OSAVI	670, 800	Rondeaux et al. (1996)
Greenness Index	G	554, 677	-
Zarco-Tejada & Miller Index	ZMI	710, 750	Zarco-Tejada et al. (2001)
Simple Ratio Pigment Index	SRPI	430, 680	Peñuelas et al. (1995)
Normalized Phaeophytinization Index	NPQI	415, 435	Barnes et al. (1992)
3-band ratio	RATIO	975, 1200, 1750	Pu et al (2003)
Photochemical Reflectance Index	PRI	531, 570 or 528, 567	Gamon et al. (1992)
Normalized Pigment Chlorophyll Index	NPCI	430, 680	Peñuelas et al. (1994)
Carter ratios	Ctr	420, 695 or 605, 760 695, 760 or 710, 760	Carter (1994)
Lichtenthaler indices	Lic	450, 800 or 440, 690	Lichtenthaler (1996)
Structure-Independent Pigment Index	SIPI	445, 680, 800	Peñuelas et al. (1995)
Vogelmann indices	Vog	740, 720 or 715, 726, 734, 747 or 715, 720, 734, 747	Vogelmann et al. (1993)
Gitelson and Merzlyak	GM	550, 750 or 700, 750	Gitelson - Merzlyak (1997)
Water Index	WI	900, 970	Peñuelas et al. (1997b)
Normalized Difference Water Index	NDWI	860, 1240	Gao (1996)
Disease Water Stress Index	DSWI	547, 802, 682, 1657	Galvão et al (2005)

Not only indices can be calculated from individual reflectance values, but other properties of the spectra may also be analysed. The “Red Edge” method calculates the derivative of the reflectance spectra in the red - near infrared region, and seeks the wavelength of maximum slope. The Red Edge position and shape can be an indicator of the chlorophyll content of a plant and its water status, among others (e.g. Filella – Peñuelas 1994). Pu et al. (2003) assessed the water status of *Quercus agrifolia* leaves using different absorption characteristics, such as wavelength position, absorption depth and width, and also a 3-band ratio at 795, 1200 and 1750 nm.

A recent study by Tan et al. (2013) compared 54 different VIs to find the best predictor for the Fraction of Photosynthetically Active Radiation (FPAR) and found that not a single VI but a combination of two VIs showed the best results. The Index Database maintained by Henrich et al. (2012) currently (in 2015) registers 517 different indices used by remote sensing. Thirty three of them aim to assess the water content of vegetation and 10 are related to stress. Clearly, there is a lot of information buried in the reflectance spectra of leaves.

The method proposed by us offers a new approach to measure not only the short term, but also the cumulative effect of environmental stress with a potentially non-destructive method using the reflectance spectra of the leaves. It is based on the concept of the state-dependent correlation between biochemical variables by Németh et al. (2009a). If more leaves are sampled from the same plant for a VI measurement, the results of the individual leaves are not the same; therefore they are usually averaged to yield a single VI for the whole plant. On the contrary, our method exploits these small differences: the distribution of the measured reflectance values and the correlations between them.

## 2 THEORETICAL BACKGROUND AND RELATED WORK

Plants tend to adjust their physiological state to the actual conditions of their environment. The primary environmental factor is light intensity, which is different in the foliage from leaf to leaf; therefore the intensities of photosynthesis are different in the individual leaves. Thus, the concentrations of plant metabolites have a distribution pattern within the foliage. Environmental factors are able to modify the expected values and standard deviations of these metabolite distributions, and they can also distort the shape of these distributions. Thus, the effect of environmental conditions on foliage appears in the distribution functions of the metabolites. This means that the expected values and the standard deviations of the metabolites can change with alterations of environmental conditions; the distribution functions of the metabolites can move to and fro along their scales (Badáczy et al. 2011; Németh 2013).

If there are at least two metabolites in the metabolism whose amounts are regulated synchronously, then their types of distributions are necessarily the same, and the levels of metabolites correlate linearly to each other (Németh et al. 2009a). The theoretical equation of state-dependent correlation is deduced from the identity of the distributions of standardized metabolite levels, and this theoretical linear relationship can be approximated by regression analysis. The correlating metabolite concentrations provide a regression straight line. The slope and the intercept of this straight line are able to reflect alterations in the moments (expected value, standard deviation) of metabolite distributions induced by the environmental factors. Thus, the state-dependent regressions can indicate the modifications in environmental condition.

### 2.1 State-dependent regressions of the reflectance spectra

The reflectance spectrum, which is perceived as various colours in the visible range, pertains to the mixture of the different materials synthesized by leaf cells. Their amounts are the daily products of the photosynthesis that follow periodic alterations controlled by metabolic regulation. The diurnal variation of chlorophylls is one of the well-known cases of metabolic oscillations (e.g. Shimada 1958, Busheva et al. 1991). Similar diurnal changes have also been detected in other pigments, e.g. the xanthophylls (Adams – Demmig-Adams 1992).

Due to their significant light absorption in the visible and infrared regions, plant pigments, e.g. chlorophylls, carotenoids, xanthophylls, etc. are the main materials that dominate the reflection spectrum of the leaves. Since their distributions within the foliage are influenced by some environmental factors, the distributions of characteristic reflection (or absorption) intensities are also sensitive to environmental factors. Moreover, if the type of their distribution is the same, then the reflectance values belonging to various wavelengths will also linearly correlate and provide state-dependent regression straight lines. In conclusion, by measuring the spectra of leaves and deriving their state-dependent regressions, the alterations of the physiological states of the plants can be tracked.

If the synthesis and decomposition of two arbitrary metabolites in the plant cells are regulated synchronistically, the actual concentration values of such metabolites are not independent of each other. Moreover, if their synchronistic regulation has linear character, then their concentrations will also be related to each other in a linear manner. Absorbance values ( $A$ ) obtained by spectroscopic measurement at particular wavelengths ( $A_{\lambda_1}$  and  $A_{\lambda_2}$ ) are proportional to the concentrations of these synchronously regulated metabolites and thus they are also linearly linked.

If the reflectance spectra have two such wavelengths ( $\lambda_1$  and  $\lambda_2$ ), whose absorbance intensities ( $A_{\lambda_1}$  and  $A_{\lambda_2}$ ) have the same type of distribution, then the standardised values of those absorbance intensities must be identical:

$$\frac{A_{\lambda_1} - \mu_1}{\sigma_1} = \frac{A_{\lambda_2} - \mu_2}{\sigma_2} \quad (1)$$

Where  $\mu$  is the expected value and  $\sigma$  is the standard deviation of the  $A_\lambda$  absorbance intensity, measured at  $\lambda$  wavelength. Rearranging *Equation 1* produces the theoretical equation for the state-dependent correlation of reflectance spectra:

$$A_{\lambda_1} = \frac{\sigma_1}{\sigma_2} \cdot A_{\lambda_2} + \frac{\sigma_2 \cdot \mu_1 - \sigma_1 \cdot \mu_2}{\sigma_2} \quad (2)$$

We investigated those wavelength pairs, whose absorbance intensities satisfy the equivalence criteria of *Equation 2*. This equation can be approximated by a linear regression: by fitting a straight line to the corresponding absorbance values:

$$A_{\lambda_1} = m \cdot A_{\lambda_2} + b \quad (3)$$

Where  $m$  is the slope and  $b$  is the intercept of the regression straight line.

## 2.2 Sensitivity and response time

On the base of the analogy between technological and biological control systems, the concept of *gain* can also be applied to physiological processes (Németh 2009b). Adaptation to different environmental conditions appears in altering the parameters  $m$  and  $b$ , which is visible from the different  $A_{\lambda_1}$  to  $A_{\lambda_2}$  linear regression. The *sensitivity* of the state-dependent regression can be evaluated by comparing the magnitude of the environmental changes e.g. temperature, to the changes of regression parameters  $m$  and  $b$ . If the relationship is more or less linear, then the slope of this straight line is hereafter called “*sensitivity*” ( $S$ ). The control theory suggests that plants under stress exhibit greater *sensitivity* to disturbing environmental factors and thus  $S$  increases.

In a controlled environment such as a greenhouse, if we abruptly change (as a step function) only one influencing factor, e.g. temperature, we can measure the properties of the response: how intensely and how quickly the plant responds to reach a new quasi-stationer state. This measurement yields the *sensitivity* and the time constant of the system for the selected environmental parameter. However, under natural climatic conditions there are many influencing factors that change simultaneously and gradually. We can measure the response of the metabolic system, but the magnitude and duration of the influencing factor is not evident. To resolve this problem, the time series of the influencing environmental variables are needed with several samples at different times, which show the actual state of the metabolic system.

The response of the plant to the changing environment is relatively slow, so the physiological state of the plant does not closely follow e.g. the daily course of the temperature but responds to a long term trend. This behaviour dampens abrupt changes and provides a more stable biological system dynamics with less fluctuation. This means that the actual state of a plant does not entirely depend on the instantaneous value of the influencing parameter, but is determined by the cumulative effect of a previous period. The length of this influential time interval can be on the order of days, and is called hereafter the “*time of influence*” ( $t_{infl}$ ).

The length of  $t_{infl}$  can also change, depending on environmental factors. To solve this problem, we calculated  $S$  as the linear regression of  $m$  against the environmental variable with many different  $t_{infl}$  to find that time interval which minimizes the variance, and therefore best describes the perceived change (see section 3.5.5).

### 2.3 The influence of the environment on the sensitivity

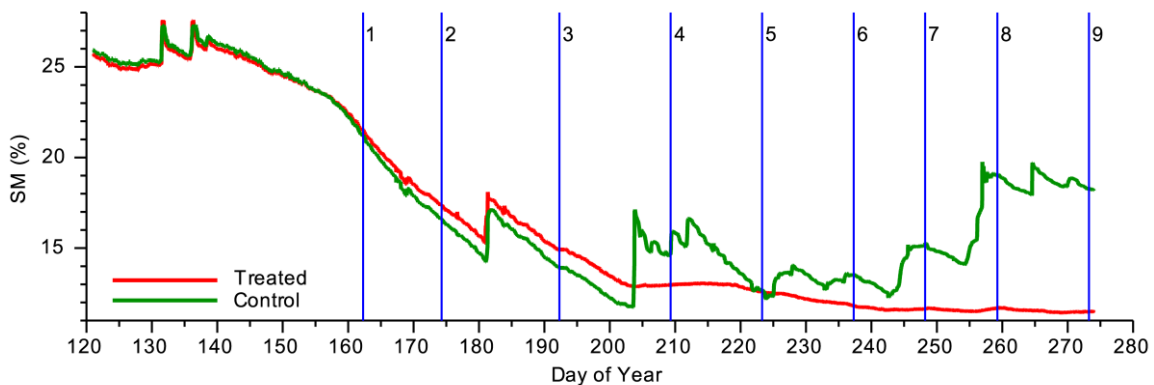
If the *sensitivity* ( $S$ ) changes over time, we can relate its change to the course of other environmental factors. With sufficient correlation and if logic doesn't prove otherwise, we may assume that the influencing factor has been found (or at least one of them). In this paper we demonstrate that soil moisture has a good agreement with *sensitivity* changes at some selected wavelength pairs and thus these wavelengths are suitable for the indication of drought stress.

## 3 MATERIALS AND METHODS

### 3.1 Site conditions

Leaf samples were collected at the Magasbérc Artificial Drought Experiment in the Sopron Mountains (N47°39'14", E16°27'34"), Hungary, from two 64-year-old beech trees (*Fagus sylvatica*) and two sessile oak trees (*Quercus petraea*). Slight drought (soil moisture deficit) was simulated at a portion of the area by building a roof of plastic foil on wooden frames 1 m above the ground, which excluded precipitation. The treated area was already separated in 2012 below ground by plastic sheets buried in a 1 m deep trench, to cut far reaching roots and prevent possible horizontal water movement. In addition, the site is located on a hilltop where not much surface or subsurface flow is expected so that the treated trees could be isolated from any water supply.

We collected leaf samples 9 times between 11<sup>th</sup> of July and 30<sup>th</sup> of September 2014, with a sampling interval of about 2 weeks. The covering roof was assembled after the third sampling and thus, after that time, the treated area received no precipitation, which resulted in continuously decreasing soil moisture. In contrast the control area received the subsequent rainfalls and its soil moisture recovered after an intense decrease in July, thanks to the rainy and cold weather in August (*Figure 1*). Except for soil moisture, all the other environmental factors acted equally on the trees.



*Figure 1. Time series of total soil moisture (SM) at the Treated and the Control plot. Blue vertical lines show the time of sample collection.*

### 3.2 Sampling protocol

We collected the leaf samples in the morning from the upper part of the canopy, using rope technique. We cut a small branch containing at least 7 healthy leaves from 1 treated and 1 control tree of both species, hereafter denoted as TB (Treated Beech), CB (Control Beech), TO (Treated Oak) and CO (Control Oak). The cut branches were placed immediately into water and processing began no more than 1 hour after sample collection. Experience shows that this procedure minimizes the impact of cutting off the branches on the measurement results.

### 3.3 Environmental data

Meteorological parameters and soil moisture were continuously monitored with a 5 minute data logging interval at the site and a nearby (400 m) micrometeorological tower. Soil moisture was measured by EC-5 sensors (Decagon Devices Inc., USA) close to the sampled trees, buried at 5 depths (10-20-30-50-90 cm). We calculated the total soil moisture ( $SM$ ) by a weighted mean of the 5 data. Air Temperature ( $T$ ) and relative humidity was measured by HMP155 sensors (Vaisala, Finland) above the canopy. From this data we calculated Vapour Pressure Deficit ( $VPD$ ) after Hardy (1998), which is the difference between the actual water vapour pressure and the saturation water vapour pressure at a particular temperature. Unlike relative humidity, Vapour Pressure Deficit has a simple nearly straight-line relationship to the rate of evapotranspiration: as  $VPD$  increases the plant needs to draw more water from its roots, therefore it is a good indicator of the evaporative demand.

### 3.4 Reflectance spectra

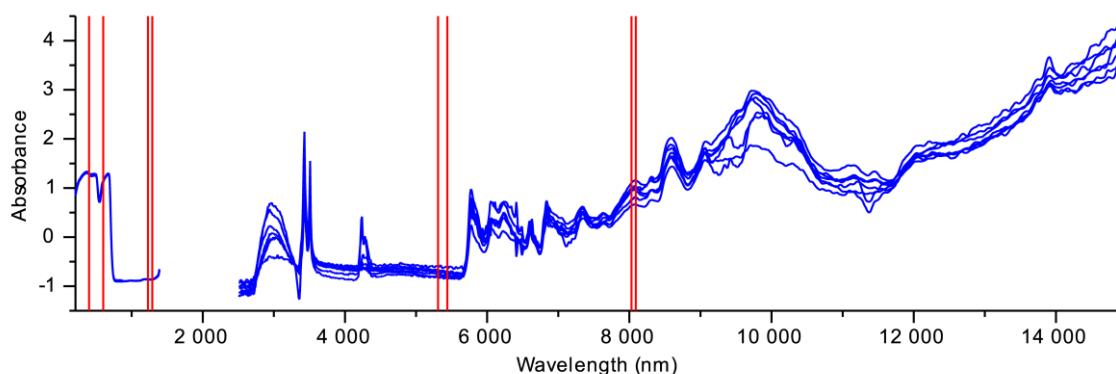
We measured the reflectance spectra of the leaves (7 by each tree) in the UV-VIS range of 200-1400 nm and 2 nm resolution with a Shimadzu UV-2600 spectrophotometer, and in the IR range of 700-400  $0\text{cm}^{-1}$  and 1  $\text{cm}^{-1}$  resolution with a Shimadzu IRAffinity-1 spectrophotometer equipped with an HATR-10 total reflection accessory (Shimadzu Corp.). The wavenumber scale ( $\text{cm}^{-1}$ ) of this second instrument was converted to nm scale (2500-14940 nm) to avoid the confusion of scales on the graphs. The reflectance spectra of each leaf was smoothed by a 10 nm wide moving average, and the Standard Normal Variate (SNV) transformation was used to pre-process the data.

### 3.5 Data evaluation

Data processing and evaluation were done in Scilab 5.4.1 (Scilab Enterprises), and the covariance analysis and grouped linear regression were carried out in StatsDirect.

#### 3.5.1 Selection of wavelength pairs

A sample set of 7 reflectance spectra (collected at the same time from the same tree) is shown in *Figure 2* with the marks of the wavelengths demonstrated in this paper.



*Figure 2. Sample reflectance spectra set of 7 leaves, collected from the Control Beech, at the first sampling. Red vertical lines mark the demonstrated wavelengths.*

Using a 10 nm resolution, the combined UV-VIS-IR spectra provide 928,884 possible pair combinations. Of course, many neighbouring wavelengths show the effect of the same material and not all the materials show linear correlation, which we looked for. To find promising wavelength pairs, we calculated the correlation matrix of the spectra, from the 7 leaves sampled at the same time. The Pearson  $R$  correlation coefficient shows the strength

and sign of the linear correlation. On these autocorrelation ‘maps’ (Figure 3) those regions have been separated that show remarkable linear correlation. In each region, wavelength pairs have been selected that exhibit the highest average correlation throughout the 9 samples. However there is no guarantee at this point that these pairs carry useful information, as e.g. they may represent two characteristic wavelengths of the same material. During the evaluation process detailed below, the useless pairs were filtered out gradually. In the following sections we demonstrate the workflow using the example of the 400-600 nm wavelength pair.

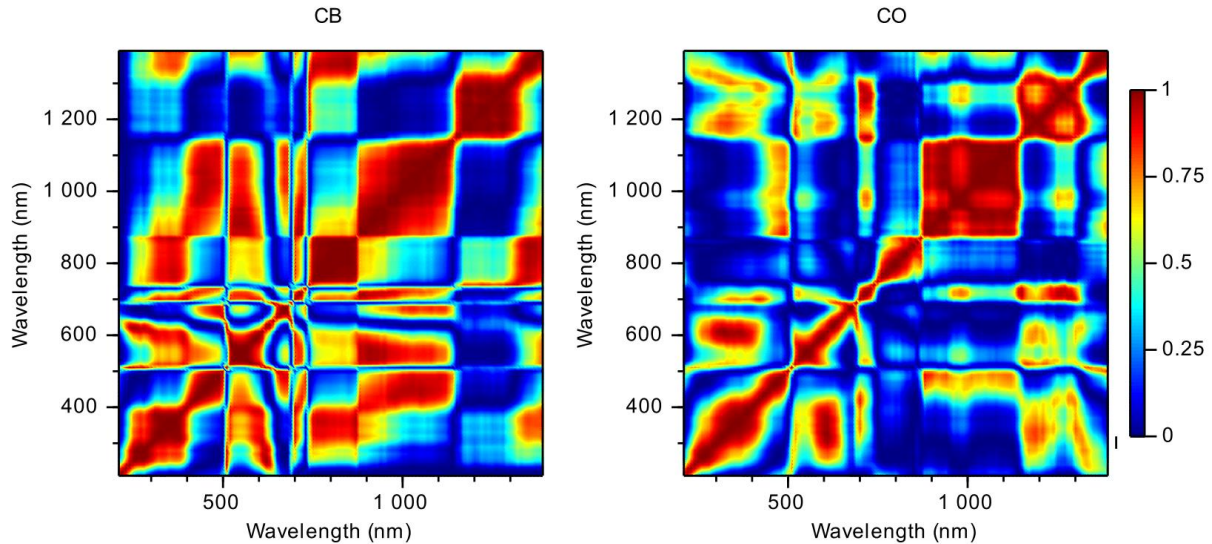


Figure 3. Two examples of autocorrelation ‘maps’ in the 200-1400 nm range, computed from 7 leaves of the first sampling. CB: Control Beech, CO: Control Oak.

The colour shows the squared correlation coefficient ( $R^2$ ):  
more red means stronger linear correlation.

### 3.5.2 State-dependent regressions

The linear regressions of the absorbance values (Equation 3) of the previously selected wavelength pairs were calculated for all the 9 samplings that produced a slope ( $m$ ) and an intercept ( $b$ ) for each sample (Figure 4).

Table 2. Pair wise slope and intercept comparison of the regressions for 400-600 nm absorbance values of the Control Beech at the 9 sampling times. The upper triangle of the table contains the probability limits of significance of slope comparisons; the lower one belongs to the intercept comparison. Darker colours show more significant differences.

		Slope comparison								
		sample 1	sample 2	sample 3	sample 4	sample 5	sample 6	sample 7	sample 8	sample 9
Vertical line separation	sample 1	-	0.5284	0.8538	0.0332*	0.1125	0.8961	0.8779	0.6364	0.7618
	sample 2	0.3650	-	0.7548	0.1246	0.3792	0.6735	0.5839	0.4124	0.8676
	sample 3	0.3156	0.8349	-	0.0841	0.2599	0.8340	0.8041	0.6272	0.9091
	sample 4	0.2279	0.6401	0.7820	-	0.4450	0.1312	0.0642	0.0361*	0.1214
	sample 5	0.0069**	0.0405*	0.0701	0.1574	-	0.3175	0.1954	0.1149	0.3475
	sample 6	0.0269*	0.1017	0.1449	0.2697	0.8116	-	0.9770	0.9064	0.7779
	sample 7	0.0005**	0.0029**	0.0063**	0.0221*	0.3104	0.2481	-	0.8377	0.7357
	sample 8	0.0002**	0.0003**	0.0005**	0.0029**	0.0765	0.0595	0.4210	-	0.5762
	sample 9	<0.0001**	<0.0001**	0.0005**	0.0022**	0.0473*	0.0451*	0.3138	0.8569	-

With ANCOVA (Analysis of Covariance) homogeneity test, those wavelength pairs were rejected that showed no or very little variation in the parameters  $m$  and  $b$ . The reason is that although they showed a good linear correlation, they were not sensitive to external environmental factors. Table 2 shows an example of the significance levels of a pair wise slope and intercept comparisons. The majority of compared pairs of the regressions are below the significance criteria, therefore these regression pairs can significantly be distinguished by their slopes or their intercepts (or both).

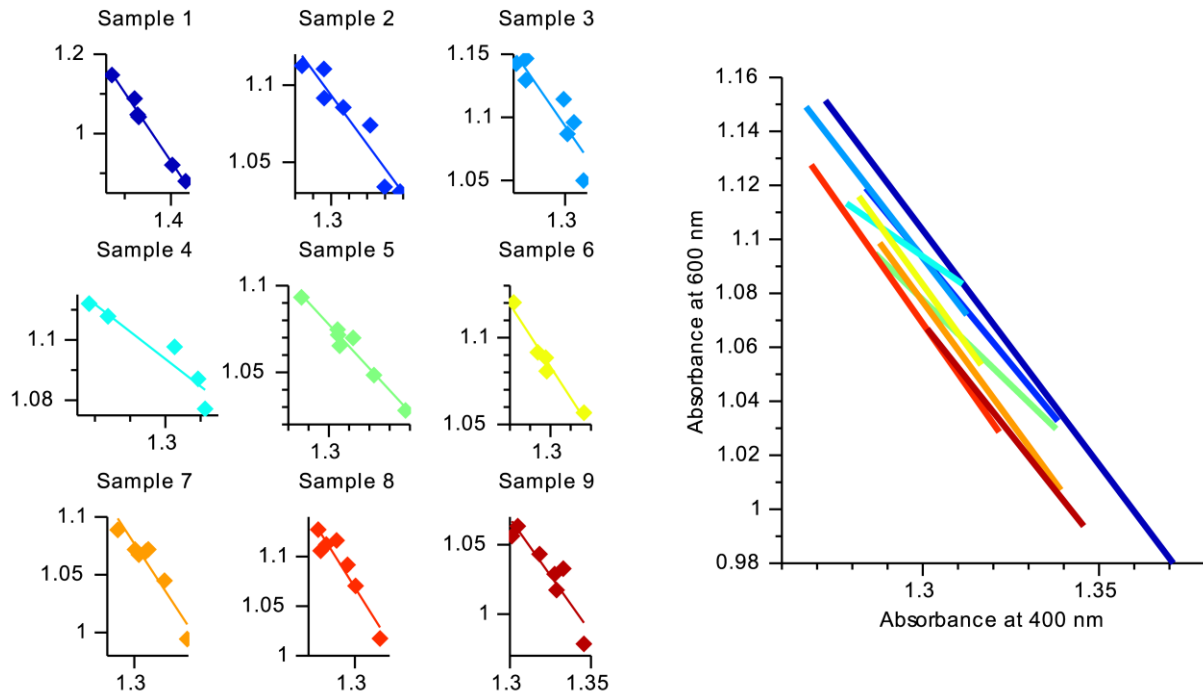


Figure 4. Example of the straight line regressions for 400-600 nm absorbance values of the Control Beech at the 9 sampling times.

### 3.5.3 Distribution test

The absorbance values of the selected wavelength pairs were standardized with the group means and standard deviations and plotted against each other. Ideally, the scatter plot should have a unity slope and zero intercept ( $m = \pm 1$ ,  $b = 0$ ) and the histograms must show similar distributions (but not necessarily a normal distribution) in order to satisfy the equivalence criteria of Equation 1. Figure 5 demonstrates that initial assumptions are strengthened and thus the state-dependent correlation theory can be applied.

### 3.5.4 Environmental data processing

It was necessary to characterise the effect of the continuously changing environment during the influencing time intervals ( $t_{infl}$ ) before each sampling. Therefore, two characteristic values were calculated: the mean ( $T_{mean}(t_{infl})$ ,  $VPD_{mean}(t_{infl})$ ), and the average rate of change, which is obtained by fitting a straight line to the time series, and used the slope (or derivative) of this line as an indicator of the intensity of the change ( $T_{change}(t_{infl})$ ,  $VPD_{change}(t_{infl})$ ). E.g.  $T_{change}(3) = -1.5$  °C/day means, that during the influencing 3 days before sampling, the cooling was an average 1.5 °C per day. But the exact length of the time interval was not previously known and therefore representative values (mean and change) were calculated for a series of different intervals, e.g.  $T_{change}(t)$ , where  $t$  ranges between 2 to 12 days with 1 hour resolution, which includes every possible value of the parameter  $t_{infl}$ .

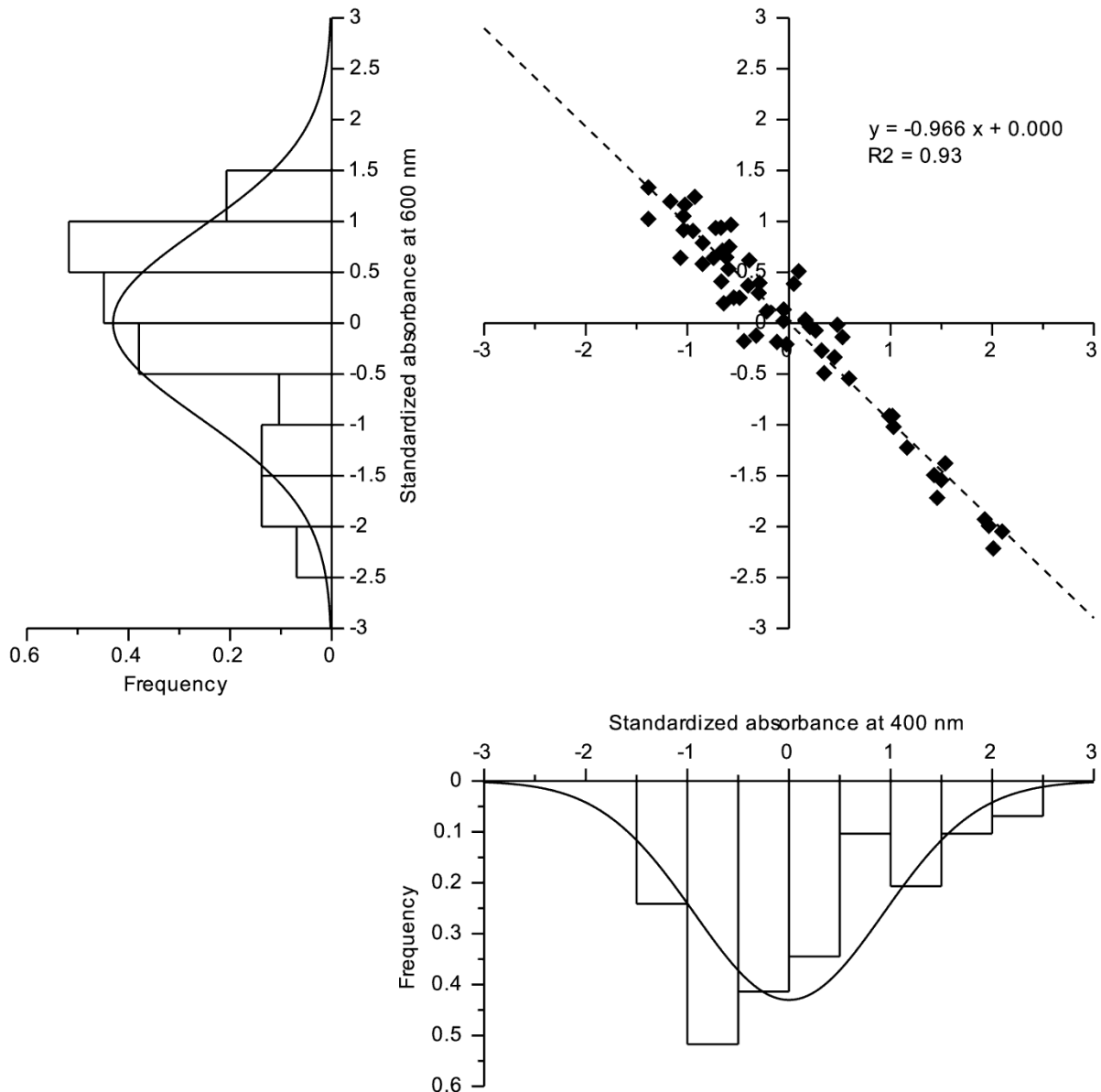


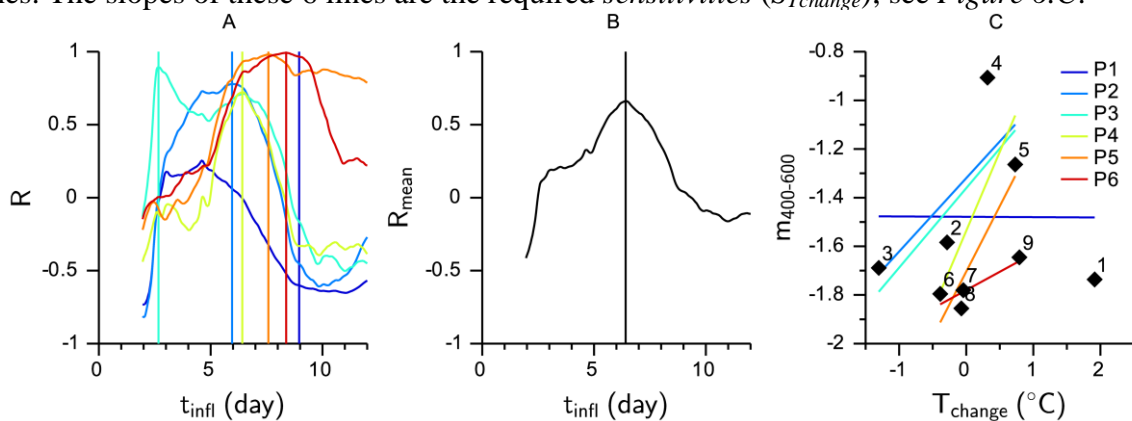
Figure 5. Control Beech 400-600 nm standardized absorbance values and their relative frequency distribution.

### 3.5.5 Sensitivity ( $S$ ) and time of influence ( $t_{infl}$ )

As defined before, the  $S$  is the *sensitivity* of the regression parameters ( $m$  and  $b$ ) to environmental conditions. Assuming constant sensitivity, if the slope  $m$  of a state-dependent regression is plotted against the environmental factor e.g.  $T_{change}$ , a derived straight line may be obtained. The slope of this straight line is the *sensitivity* related to temperature change ( $S_{Tchange}$ ). It shows how the state-dependent regression of the selected absorbances changes, when the weather is warming or cooling. If all the slopes of the state-dependent regressions fit to a common line, then the light absorbances at those wavelengths are not sensitive to any additional environmental stress factor, because the *sensitivity* is stable ( $S_{Tchange} = const.$ ). On the contrary, significant changes of  $S$  have been sought that can be used as an indicator of environmental stress. To investigate the possible changes of  $S$  as time passes, the samplings were grouped into shorter *periods*, and the  $S$  of each *period* was computed individually. Since there were only 9 samplings, 6 overlapping *periods* ( $P$ ) were created from groups of 4 subsequent samplings, e.g.  $P_1$ : sample 1-2-3-4,  $P_2$ : sample 2-3-4-5,  $P_3$ : sample 3-4-5-6, etc.

The *time of influence* ( $t_{infl}$ ) is the time interval before the sampling that describes the observed state with the highest probability. To find this time, for each *period* the Pearson  $R$  correlation coefficient was calculated for all the cases of  $m$  versus e.g.  $T_{change}(t)$ , where the latter is the characteristic value of the environmental parameter calculated at every possible  $t_{infl}$  (2-12 day). Graphically, it is equivalent to fitting a straight line to the measured  $m$  vs.  $T_{change}(t)$  at all the possible  $t_{infl}$ , and  $R$  describes the goodness of fit. The change of this correlation coefficient ( $R$ ) against time ( $t$ ) is shown in *Figure 6.A* for all 6 *periods*. The maximum (or minimum) of this curve shows the time of the best fit and thus this time is the  $t_{infl}$ . The same concept can also be applied for other environmental variables (e.g.  $VPD$ ) and both the *mean* and *change* values.

The  $t_{infl}$  values calculated for the different *periods* sometimes show good agreement, but many times there are some *periods* out of the 6, which produce extremely low or high values (e.g. 2.6 day in *Figure 6.A*). In theory the *time of influence* is not a fixed value, but may change as  $S$ , since  $t_{infl}$  is also a biological response parameter. But these extremes are probably due to a special pattern of the weather alteration that misguides the algorithm. To safely calculate the real  $t_{infl}$  for each *period*, more samplings were needed. To avoid the disturbing effect of these outliers, we assumed a constant  $t_{infl}$  during all the *periods*. This is based on the mean  $R$  of the 6 *periods*: the time of the maximum (or minimum) of this  $R_{mean}$  indicates the presumed common  $t_{infl}$  (*Figure 6.B*). Graphically, this  $t_{infl}$  ensures the best common fit of the 6 lines. The slopes of these 6 lines are the required *sensitivities* ( $S_{Tchange}$ ); see *Figure 6.C*.



*Figure 6. Control Beech 400-600 nm response to  $T_{change}$  variations.*

A.) *The correlation coefficients ( $R$ ) vs. all possible  $t_{infl}$  for the 6 periods ( $P$ ).*

*The maximum (or minimum) of each curve indicates the assumed  $t_{infl}$ , marked with a vertical line.*

B.) *The mean of the 6 correlation coefficients ( $R_{mean}$ ). Its maximum indicates the common  $t_{infl}$ .*

C.)  *$m$  vs.  $T_{change}(t_{infl})$  with the regression lines of the 6 periods. The slopes of these 6 lines are the sensitivities of the periods ( $S_{Tchange}$ ).*

### 3.5.6 Changing sensitivity in response to environmental stress

We needed to describe the overall effect of environmental conditions which may induce a stress syndrome in the plants. We calculated the mean values of the main environmental variables ( $SM$ ,  $T$  and  $VPD$ ) during the presumed common  $t_{infl}$  before every sampling, and from these ones, the *period means* were computed for the 6 *periods*. In this way, for every *period* 3 environmental factors ( $SM_{period-mean}$ ,  $T_{period-mean}$ ,  $VPD_{period-mean}$ ) and a *sensitivity* ( $S$ ) were obtained. Plotting  $S$  against the *period means* of the environmental factors, characteristic curves are revealed. The first graph in *Figure 6* shows that with decreasing soil moisture the *sensitivity* increases (negative slope decreases) which can also be seen in *Figure 5.C* as more steep lines. Similarly, with increasing soil moisture the *sensitivity* decreases as expected. On the contrary, the *sensitivity* shows no clear connection to the temperature or the vapour pressure deficit (*Figure 7. T and VPD*).

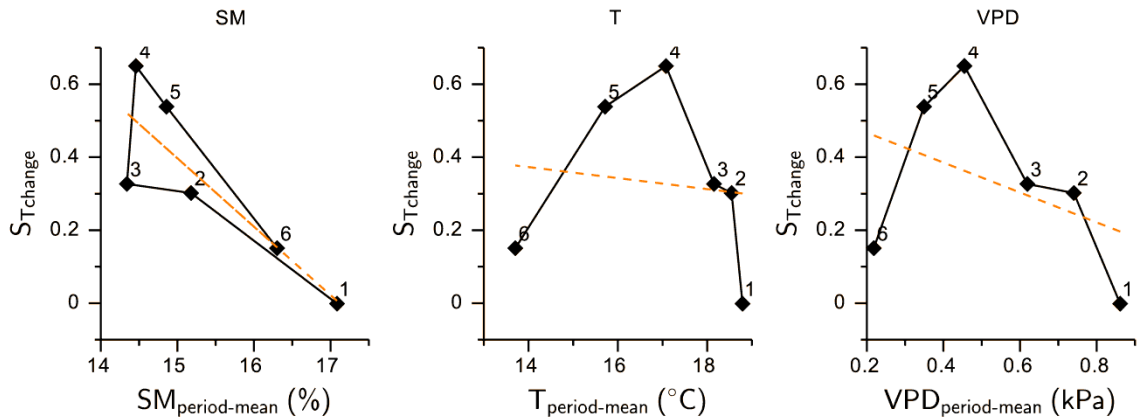


Figure 7. Control Beech 400-600 nm sensitivity for temperature change ( $S_{Tchange}$ ) during the 6 periods (identified by numbers) vs. the environmental variables: SM - Soil moisture, T - Mean temperature, VPD - Vapour Pressure Deficit.

#### 4 RESULTS AND DISCUSSION

Applying the previously described method, wavelength pairs that showed systematic *sensitivity* changes under drought stress were selected.

There are wavelengths which work well for both species, e.g. 400-600 nm, which is presented in *Figure 8*. Here, the regression parameter ( $m_{400-600}$ ) varies in response to the average temperature change ( $T_{change}$ ). The *sensitivity* of the regression in the 6 periods is expressed by the *sensitivity for temperature change* ( $S_{Tchange}$ ). This  $S_{Tchange}$  is plotted against the average soil moisture ( $SM_{period-mean}$ ), which shows that in beech trees (*Figure 8 left*) the *sensitivity* increases with decreasing soil moisture (increasing drought). This means if the trees are under stress, the change of  $m_{400-600}$  is bigger for the same amount of temperature change. The *sensitivities* of control beech (CB) show a nearly linear relationship with the soil moisture. The *sensitivities* of the treated beech (TB) show less change for a while, but below a certain soil moisture content (< 13%) they increase considerably.

The same seems to be true for the oaks (*Figure 8 right*); the only difference is the sign of the *sensitivity*. This negative sign means that for temperature increase (warming), oak trees raise the absolute magnitude of  $m_{400-600}$ . On the contrary, beech trees lessen the absolute magnitude of  $m_{400-600}$  under the same circumstances. What matters is not the sign, but the magnitude of the *sensitivity*, which increases with the decreasing soil moisture as expected, signalling greater stress. If the wavelengths for the calculation of  $m$  are simply exchanged (e.g. not 400-600 nm, but 600-400 nm), the sign of  $S$  reverses.

Regressions of wavelength pairs, which can express *sensitivity* to the  $VPD_{change}$  are also be found. An example is shown with the beech trees in *Figure 9*, measured at 1230-1290 nm.

Not only the visible or near infrared range can be informative, but sensitive wavelengths in the middle infrared region can also be identified. An example for the oak trees at 5310-5440 nm (1883-1838  $cm^{-1}$ ), and for the beech trees at 8030-8090 nm (1245-1236  $cm^{-1}$ ) is depicted in *Figure 10*. Both regressions of wavelength pairs are *sensitive* to the rate of temperature change ( $T_{change}$ ). These regressions show behaviour similar to the previously detailed examples.

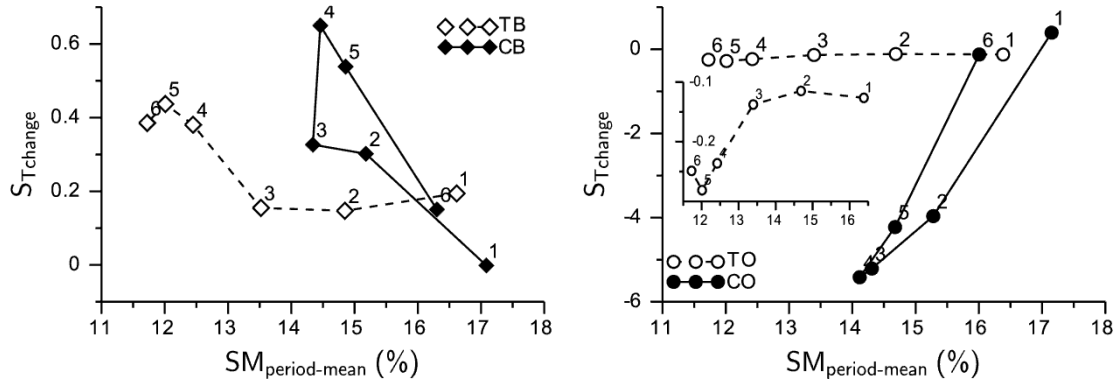


Figure 8. Beech (left) and Oak (right) 400–600 nm sensitivity ( $S_{Tchange}$ ) vs. average Soil Moisture ( $SM_{period-average}$ ) during the 6 periods.  $S_{Tchange}$  is the sensitivity of  $m_{400-600}$  to  $T_{change}$  variations. TB: Treated Beech, CB: Control Beech, TO: Treated Oak, CO: Control Oak.

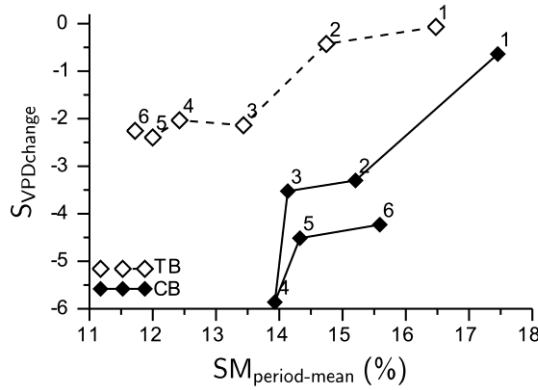


Figure 9. Beech 1230-1290 nm sensitivity ( $S_{VPDchange}$ ) vs. average Soil Moisture ( $SM_{period-average}$ ) during the 6 periods.  $S_{VPDchange}$  is the sensitivity of  $m_{1230-1290}$  to VPD change variations. CB: Control Beech, TB: Treated Beech.

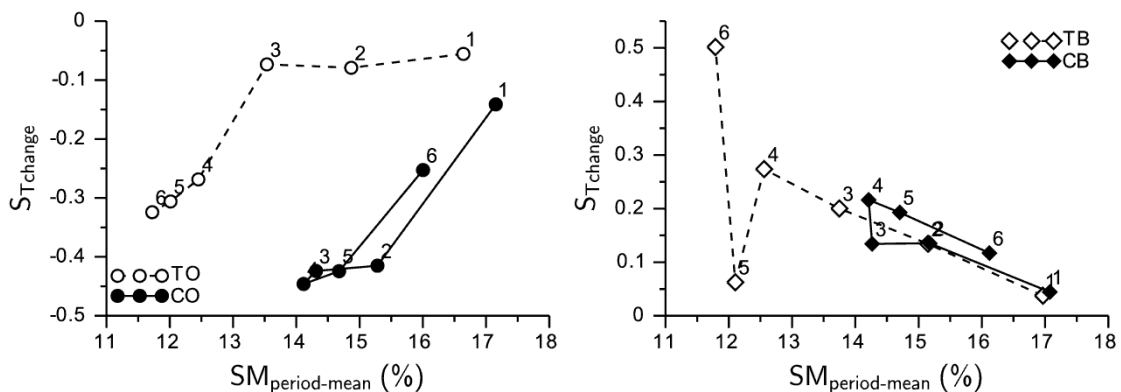


Figure 10. Oak 5310-5440 nm (left) and Beech 8030-8090 nm (right) sensitivity ( $S_{Tchange}$ ) vs. average Soil Moisture ( $SM_{period-average}$ ) during the 6 periods.  $S_{Tchange}$  is the sensitivity of  $m_{5310-5440}$  and  $m_{8030-8090}$  to  $T_{change}$  variations, respectively. TO: Treated Oak, CO: Control Oak, TB: Treated Beech, CB: Control Beech.

At 400 nm and 600 nm wavelengths the absorbance intensities are mainly affected by the photosynthetic pigments (chlorophylls, xanthophylls, carotenoids, etc.). At 400 nm almost all of the pigments contribute to the resultant absorption, but at 600 nm the absorbance is determined mainly by chlorophylls. Thus the  $m_{400-600}$  slope changes of the state-dependent regressions indicate the variation in the ratio of chlorophylls compared to the total pigment content, and this variation is sensitive to the environmental parameters such as temperature and soil moisture. Between 1200 and 1300 nm, water and the lingo-cellulose system are the significant influencing factors for the light absorption. At even higher wavelengths, the overtones of vibration resonances of the molecules prevail the absorbance spectra.

However, to successfully use the state-dependent regression method, the exact knowledge of the underlying materials is not required and in many cases it is not even possible due to the high number of different bio-materials that shape the spectra at a given wavelength.

The wavelengths presented in this paper are not meant to be a process description, but to serve as the proof of concept. To fine tune the method for practical application more investigation is needed.

## 5 SUMMARY

Middle-aged beech and sessile oak trees have been investigated while slight drought (soil moisture deficit) has been simulated at half of the trees. Environmental parameters have been continuously monitored and the foliage has been sampled regularly to obtain UV-VIS-IR reflectance spectra of the leaves.

The state-dependent correlations at particular wavelength pairs in the reflectance spectra of foliage are sensitive to environmental variations. Changes in the environmental parameters, such as those of temperature or vapour pressure deficit, cause significant variations in the slope of the regression straight lines. The *sensitivity* of this variation, e.g. how big the slope change is for a given temperature rising, can be expressed by the *sensitivity for temperature change*. This *sensitivity*, in turn, varies with the soil moisture; the bigger the water deficit, the greater the absolute magnitude of the *sensitivity*. Therefore the slopes of the state-dependent regressions of some selected wavelengths are good indicators of the available soil moisture.

If the selected wavelengths are not directly sensitive to the water content of leaves, but measure some synthesized bio-materials, then the slopes represent the underlying metabolic regulation. In this way, the changes of *sensitivity* reflect the changes of metabolic regulation and its stability. The state and the stability of the regulation define the level of drought stress, summing up the long term effects of the environment.

This method applies the approach of state-dependent correlation for data analysis. This kind of correlation, due to the synchronous regulation of certain bio-material pairs, can be found in many biochemical processes. The exact qualification of materials influencing the reflectance spectra may be desirable but is not necessarily required. This is because it is not the material qualities but their amounts that determine the regression relationship at the selected wavelengths. The existence of state dependent correlation can be proved statistically without a priori knowing the materials involved. Moreover, as previous studies suggest, it might be utilized to assess not only drought stress, but many other environmental stress factors, too. However, it requires further investigation.

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## Post-epidemic Situation of a Previously *Phytophthora alni*-infected Common Alder Stand

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**Abstract** – This paper reports on the current situation of the *Phytophthora* species occurring in a declining common alder (*Alnus glutinosa*) stand in North-West Hungary. The stand was affected by a severe epidemic caused by *Phytophthora alni* in the late 1990s. The authors evaluated the health condition of the forest stand and collected soil samples from the rhizosphere of twenty selected trees two times per year in 2011 and in 2012 in order to isolate *Phytophthora* species. A diverse *Phytophthora* community was found in the soil consisting of eight species with different aggressiveness and with different ecological demands. Pathogenicity tests confirmed the role of the collected strains in the decline of the alder stand.

**Root and collar rot of alder / soilborne pathogen / *Phytophthora alni* / *Phytophthora taxon Raspberry* / *Phytophthora lacustris***

**Kivonat** – Egy korábban *Phytophthora alni* által fertőzött mézgáséger-állomány járvány utáni állapota. A tanulmány egy pusztuló nyugat-magyarországi mézgáséger- (*Alnus glutinosa*-) állományban jelenleg előforduló *Phytophthora*-fajokról, és azok faállományra gyakorolt hatásáról tudósít, egy korábban lezajlott, *Phytophthora alni* okozta súlyos járványt követően. A szerzők vizsgálták a faállomány egészségi állapotát, illetve talajmintákat gyűjtöttek a fák gyökérszójából a *Phytophthora*-fajok kitenyésztése céljából. A vizsgálatokat 20 megjelölt fán végezték, 2011-ben és 2012-ben, évente 2–2 alkalommal. Az állományban változatos, eltérő agresszivitású és környezeti igényű fajokból álló *Phytophthora* közösséget találtak. A patogenitási tesztek igazolták a gyűjtött izolátumok szerepét az erdőpusztulásban.

**éger gyökér és gyökfő betegsége / talajlakó kórokozó / *Phytophthora alni* / *Phytophthora taxon Raspberry* / *Phytophthora lacustris***

### 1 INTRODUCTION

A severe epidemical decline of common alder (*Alnus glutinosa*) trees was observed in South-England in the summer of 1993. An unknown *Phytophthora* species was isolated from the plant tissues and rhizosphere soil samples (Brasier et al. 1995). The previously unknown ‘alder *Phytophthora*’ spread epidemically throughout Europe in the last decade of the 20<sup>th</sup> century (Cech 1998, Werres et al. 1998, Szabó et al. 2000, Jung – Blaschke 2004, Cerny et al. 2008).

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The ‘alder *Phytophthora*’ is an interspecific hybrid, a series of phenotypically diverse, heteroploid genotypes coming from asexual hybridisation (Brasier et al. 1999). The species was described in 2004 as *P. alni* and the different variants were described as subspecies: *P. alni* ssp. *alni*, *P. alni* ssp. *multiformis* and *P. alni* ssp. *uniformis* (Brasier et al. 2004). The disease can occur in various habitats and in trees representing different ages. However, the primarily source of invasion might be the planting of infected saplings (Oszako 2010).

Other *Phytophthora* species were also isolated from the necrotic tissues or from the rhizosphere soil of infected alder trees. *P. citricola*, *P. cactorum*, *P. gonapodyides*, *P. megasperma* and *P. pseudosyringae* were isolated from necrotic tissues while *P. inundata*, *P. lacustris*, *P. plurivora*, *P. gregata*, *P. taxon hungarica* and *P. polonica* were isolated from rhizosphere soil (Szabó et al. 2013, Marçais – Husson 2014, Belbahri et al. 2006). These species may contribute to the decline of the alder stands (Szabó et al. 2013). However, they should not to be the main reason for the epidemic alder decline (Marçais – Husson 2014).

In 1999, a severe alder decline was observed near Csorna, North-West Hungary (Szabó et al. 2000). *P. alni* ssp. *uniformis* was isolated both from rhizosphere soil samples and necrotic root tissues (Szabó et al. 2000, Nagy et al. 2003). By 2003, the health condition of the alders improved gradually (Szabó et al. 2013). Instead of *P. alni*, clade 6 *Phytophthoras* were isolated from the rhizosphere soil samples (Szabó et al. 2013). This time, alder decline was observed at several other locations in Hungary (Koltay 2007). However, in addition to *P. alni*, other *Phytophthoras* were prevalent at these sites, too (Szabó et al. 2013). In the beginning of the 21<sup>st</sup> century, frequently isolated *Phytophthora* species from alder stands were *P. gonapodyides*, *P. plurivora*, *P. inundata*, *P. megasperma*, *P. lacustris*, *P. gregata* and two undescribed taxa, *P. taxon hungarica* and *Phytophthora* sp. 1. (Szabó et al. 2013).

Although, alder decline caused by the *P. alni* subspecies is well-studied, there are still some unanswered questions, especially about the ecology and evolution of the subspecies. What happens in the surviving forest stands afterwards is also an important question.

A diverse *Phytophthora* community occurs in the soil of the alder stands because of the favourable wet conditions (Szabó et al. 2013). However, we also lack information about the impact of other *Phytophthoras* on alder trees. Monitoring the health condition and *Phytophthora*-composition in these stands is necessary to predict a potential epidemic in time.

Herein, we present the results of a post-epidemic resurvey that was carried out in an alder forest severely attacked by *P. alni*. We also report on the differential pathogenicity of various *Phytophthora* species on alder saplings as a result of two kinds of inoculation methods.

## 2 MATERIALS AND METHODS

### 2.1 Evaluation of the health condition

Twenty common alder trees were selected for sampling in a thirty-three-year-old marshland forest near Csorna, West Hungary (*Figure 1*). The investigated forest stands grow on fen soil with permanent water effect. The evaluation of the health condition and the sampling was done in June and September of 2011 and 2012.

Specific symptoms of *Phytophthora* root and collar rot of alders, like bark necrosis often with a dark exudate, usually appear at the base of the trunk. The health condition of trees was evaluated based on the specific collar and secondary crown symptoms. To evaluate the severity of the crown and collar symptoms, five-point scales were used (*Figure 2*).

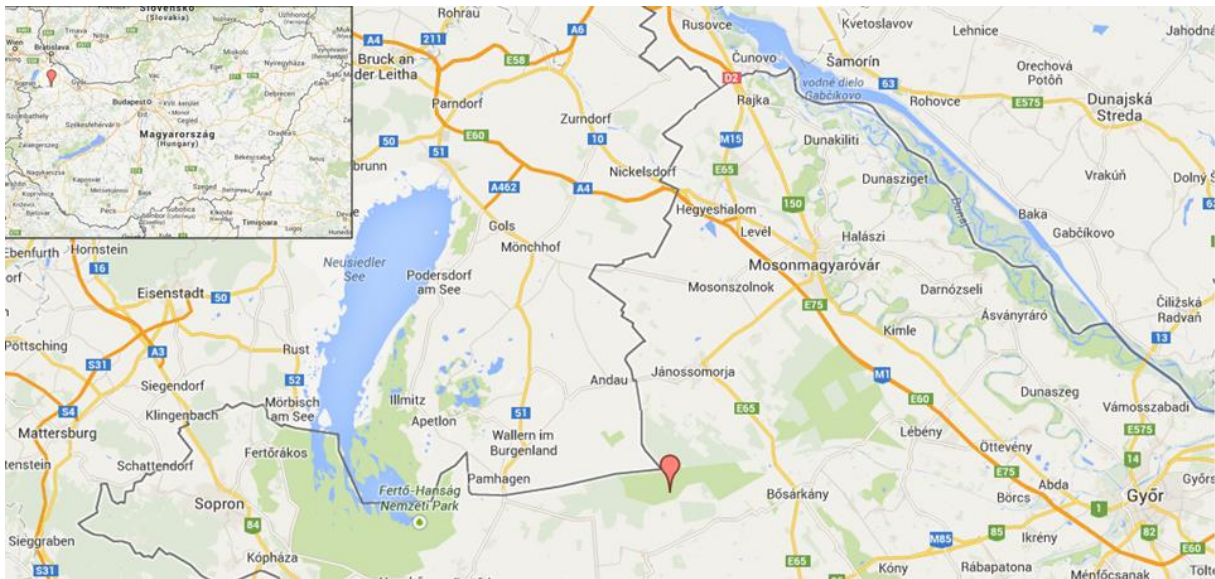


Figure 1. The sampling site. Source: Google Maps.



Figure 2. A1-A5: Health condition categories based on collar symptoms:

A1: Asymptomatic tree; A2: Bark necrosis covers less than 10% of the girth of the trunk horizontally and spreads vertically to a maximum 10% of the total height of the tree; A3: Bark necrosis covers 10–30% of the girth and height of the tree; A4: More than 30% of the total girth and height of the tree is damaged; A5: Dead tree.

B1-B5: Health condition categories based on crown symptoms:

B1: Healthy crown; B2: Less than 20% of the crown is dying. Some leaves are yellowish; B3: 20–50% of the crown is dying. Leaves with yellowish discoloration occur in groups. B4: More than 50% of the crown is dead. Yellow leaves in large groups, or the remaining foliage is yellow; B5: Dead tree.

## 2.2 Sampling, isolation and species identification

The method of soil sampling, sample processing and isolation were the same as reported earlier (Kovács et al. 2013). Identification of the collected isolates was based on morphological characteristics and specific molecular markers as described earlier (Kovács et al. 2013).

### 2.3 Pathogenicity tests

The pathogenicity of the following four most important species (collected in 2011) was tested: *P. alni*, *P. taxon Raspberry*, *P. inundata*, *P. lacustris*. Additionally, a *P. gonapodyides* strain was also tested because this species often occurred earlier in the rhizosphere soil of alders (Szabó et al. 2013). One representative isolate per *Phytophthora* species was used for inoculation. Inocula were collected from 14-day-old colonies grown on potato-dextrose agar plates (39g/L, Microtrade Ltd., Budapest, Hungary) in Petri dishes with a diameter of 90 mm.

One-year-old common alder saplings grown in plastic containers were infected. The soil for planting was tested for the lack of *Phytophthoras* using a leaf-baiting method.

Seventeen saplings per each isolate and corresponding controls (altogether 93 saplings) were wound-inoculated on April 26 and 27, 2013. A narrow wound with a length of approximately 5 mm was made on the base of the stem with a sterile scalpel and a 5 mm × 5 mm pathogen-infested agar plug was inserted in the wound. The wound was then sealed with Parafilm® (Pechiney Plastic Packaging Company). Control saplings had non-infested agar pieces in their wounds and were sealed as above. Another 17 saplings per isolate were used for root infestation (altogether 94 saplings including the control saplings). Altogether two pathogen colonies per sapling were put into the soil of the saplings at four positions around their stem.

The saplings were watered when necessary, and maintained under natural environmental conditions. After five months of incubation, disease severity of the saplings was evaluated based on five-point scales, 1 and 5 expressing no disease and lethality, respectively. In the case of the stem inoculations, the length and width [mm] of the bark lesion were measured. The average lesion size was calculated from the measured data based on the formula of an ellipsoid [mm<sup>2</sup>].

The health condition of the root system was evaluated based on a 5-point scale (Figure 3).



Figure 3: The scale for evaluating the health condition of the root system:

1. Healthy root system;
2. Root loss is below 30%;
3. 30–50% of the root system is lost;
4. Root loss is more than 50%;
5. Completely dead saplings.

The health condition of the shoots was evaluated based on a similar scale: 1. Symptomless sapling; 2. Some leaves are smaller than usual, occasionally with yellowish discolouration; 3. 30–50% of the potential crown is dead; 4. More than 50% of the potential crown is dead; 5. Completely dead saplings.

## 2.4 Data analysis

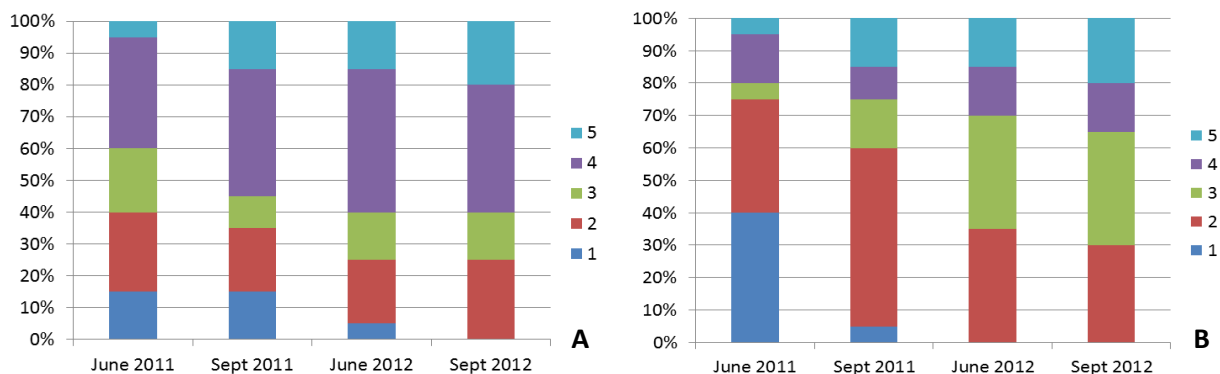
The health condition datasets were analysed with the Software STATISTICA (Ver. 11, StatSoft Inc. 2012). We used Kruskal-Wallis and Mann-Whitney tests to see the change in the health condition of our sampling site.

Data resulted from the pathogenicity tests were also analysed with STATISTICA. The Gaussian distribution of data sets was tested with the Shapiro-Wilks method. The homogeneity of variances of the variables was tested with Levene statistic, based on the median. Because the lesion size dataset does not have Gaussian distribution based on the results of the Shapiro-Wilks method, non-parametric tests were used to evaluate the pathogenicity. Kruskal-Wallis non-parametric ANOVA was used to compare multiple independent groups, and the Mann-Whitney U-test was used for pairwise comparisons of the treatment groups at  $\alpha=0.05$  significance level.

## 3 RESULTS

### 3.1 Changes in the health condition

The health condition of the trees based on the bark symptoms (*Figure 4A*) did not change significantly during the experiment. Active lesions with fresh exudate could be observed altogether three times. Each time only one tree had fresh collar symptoms. However, the health condition of the trees based on the crown symptoms (*Figure 4B*) became considerably worse ( $p=0.0053$ ) according to the Kruskal-Wallis test. Significant differences were observed between the results of June 2011 and 2012 ( $p=0.0061$ ), and between the results obtained in June 2011 and September 2012 ( $p=0.0029$ ). Changes are conspicuous in the decrease of the number of symptomless trees (from eight in June 2011 to zero in June and September 2012), and in the increase of the number of dead trees (from one in June 2011 to four in September 2012).



(1: best – 5: worst, details in the text)

*Figure 4 A: Changes of the health condition based on the health condition categories of root collar and bark symptoms.*

*Figure 4 B: Changes of the health condition of the sampling site based on the health condition categories of crown symptoms.*

### 3.2 Observed *Phytophthora* species composition

*Figure 5* shows the observed species composition at different sampling times. Altogether nine *Phytophthora* taxa were isolated during the two-year survey: *P. alni* ssp. *multiformis*, *P. lacustris*, *P. taxon Raspberry*, *P. inundata*, *P. plurivora*, *P. polonica*, *Phytophthora* sp. *oaksoil*, *P. gonapodyides* and *Phytophthora* taxon *hungarica*. The most diverse species composition was observed in June 2011. Five different species, *P. alni* ssp. *multiformis*, *P. lacustris*, *P. taxon Raspberry*, *P. sp. oaksoil* and *P. inundata* were isolated at that time. Only

one species, *P. lacustris* was present at all times. *P. taxon Raspberry* was collected two times, in June 2011 and September 2012. The other species were found only once out of the four sampling times. They were present in the soil samples with low frequency.

Despite its low frequency, the presence of the highly aggressive *Phytophthora alni* in the rhizosphere soil of the trees is of great importance. At the very first sampling time, the *P. alni* ssp. *multiformis* was detected in the rhizosphere of two trees. However, at the other sampling times, we were unable to isolate this pathogen again.

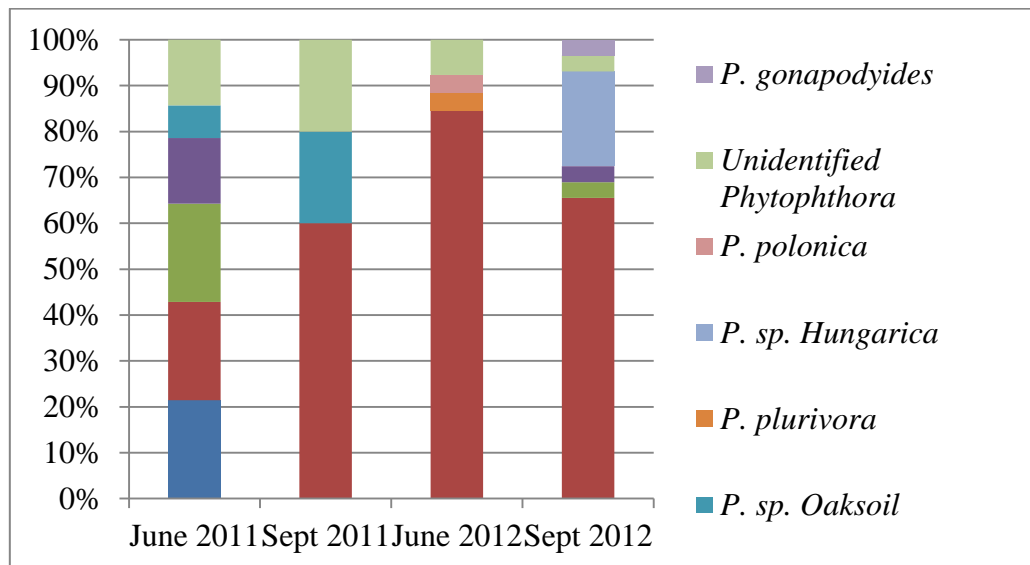


Figure 5: The observed species composition at the different sampling times.

### 3.3 Pathogenicity

#### 3.3.1 Soil infestation test

Two out of 94 infected saplings died during the test period. Both saplings were infected with *P. alni* ssp. *multiformis*.

According to the results of the Kruskal-Wallis nonparametric ANOVA, the health condition of the roots of the saplings (Figure 6A) was significantly different between the groups with different treatments ( $p=0.0000$ ). The soil infestation test did not result in significant differences in the health condition of the shoots during the test period ( $p=0.0513$ ).

Based on the pairwise comparisons with the Mann-Whitney U-test, the health condition of the root system was worse in every infected group than it was in the non-infected control group. The most substantial damage was caused by the *P. alni* ssp. *multiformis* isolate ( $p=0.0000$ ). Similar severe damage was caused by the *P. lacustris* ( $p=0.0001$ ). The root system of the saplings infected with *P. taxon Raspberry* or *P. inundata* was also considerably weaker than the health condition of the roots of the control saplings ( $p=0.0014$  and  $p=0.0177$ ). However, the root system of *P. gonapodyides*-infected saplings was not significantly damaged in comparison with the root system of control saplings ( $p=0.1497$ ). *P. alni* damaged the root system more severely than *P. taxon Raspberry* ( $p=0.0293$ ), *P. inundata* ( $p=0.0002$ ) or *P. gonapodyides* ( $p=0.0001$ ). Damage caused by *P. inundata* was milder than that caused by *P. lacustris* ( $p=0.0020$ ), but was not much different from the damage caused by *P. gonapodyides* ( $p=0.4745$ ). *P. lacustris* caused significantly more severe damage to alder roots, than *P. gonapodyides* did ( $p=0.0006$ ). The damage caused by *P. taxon Raspberry* did not differ greatly from the damage caused by *P. inundata* ( $p=0.1729$ ), *P. lacustris* ( $p=0.1434$ ) or *P. gonapodyides* ( $p=0.0665$ ).

### 3.3.2 Stem inoculation test

Four out of 93 saplings, i.e. those inoculated with the *P. alni* ssp. *multiformis* isolate, died during the test period.

According to the results of the Kruskal-Wallis non-parametric ANOVA, the treatment groups differ to a great extent in terms of the health condition of the shoots of saplings ( $p=0.0011$ ). As to the condition of the root system of saplings, there are also pronounced differences between the groups ( $p=0.0007$ ), albeit the planting media of these saplings was not infected with Phytophthoras. The lesion sizes (Figure 6. B) were also significantly different ( $p=0.0000$ ) between the treatment groups.

Based on the pairwise comparisons with the Mann-Whitney U-test, the health condition of the shoots was the poorest in the *P. alni*-infected group. It was considerably poorer than in the non-infected control saplings ( $p=0.0244$ ), as well as in the *P. taxon Raspberry*-infected ( $p=0.0081$ ), the *P. gonapodyides*-infected ( $p=0.0027$ ) and the *P. inundata*-infected ( $p=0.0114$ ) groups.

However, based on the symptoms of the *P. alni*-infected group, the health condition of the saplings did not differ significantly from the health condition of the *P. lacustris*-infected group ( $p=0.5228$ ). The health condition of the shoots in the *P. lacustris*-infected group were damaged much more than the shoots in the *P. taxon Raspberry* ( $p=0.0380$ ), *P. gonapodyides* ( $p=0.0131$ ) and *P. inundata*-infected ( $p=0.0487$ ) groups.

Although only wound inoculation was carried out, there were notable differences in the health condition of the roots between the treatment groups. Based on the health condition of the root system, the most severe damage was caused by the *P. alni* ssp. *multiformis* isolate. The root systems of the saplings in the *P. alni* ssp. *multiformis*-inoculated group were significantly more degraded than the roots of the control saplings ( $p=0.0114$ ), the *P. taxon Raspberry*-infected ( $p=0.0401$ ) saplings and the *P. inundata*-infected ( $p=0.0309$ ) saplings. Furthermore, *P. lacustris* caused substantial damage to the roots compared to the control saplings ( $p=0.0177$ ). However, its impact was not significantly different from that of the other three Phytophthoras used for the inoculation test. *P. alni* ssp. *multiformis* and *P. lacustris* seem to be able to colonize roots secondary through the transport vessels of the sapling.

Every *Phytophthora* species used for the stem inoculation proved to be pathogenic to alder saplings ( $p=0.0001$ ). The minimum, average, and maximum area of the lesions caused by the isolates used are summarized in Table 1. The lesions caused by the *P. alni* ssp. *multiformis* strain were considerably larger than those caused by *P. lacustris* ( $p=0.0309$ ), *P. taxon Raspberry* ( $p=0.0031$ ), *P. gonapodyides* ( $p=0.0031$ ) and *P. inundata* ( $p=0.0008$ ). The lesions caused by *P. lacustris* were also ascertainably larger than those caused by *P. taxon Raspberry* ( $p=0.0027$ ), *P. gonapodyides* ( $p=0.0040$ ) and *P. inundata* ( $p=0.0001$ ).

Table 1: Average size of lesion caused by the species used for the wound inoculation experiment

Species	Area of lesion (mm <sup>2</sup> )		
	minimum	average	maximum
<i>P. taxon Raspberry</i>	7.85	56.51	188.50
<i>P. alni</i> ssp. <i>multiformis</i>	0.00	502.32	1350.90
<i>P. gonapodyides</i>	0.00	45.61	
<i>P. inundata</i>	0.00	33.68	66.76
<i>P. lacustris</i>	31.42	132.59	522.29
Control	0.00	0.00	0.00

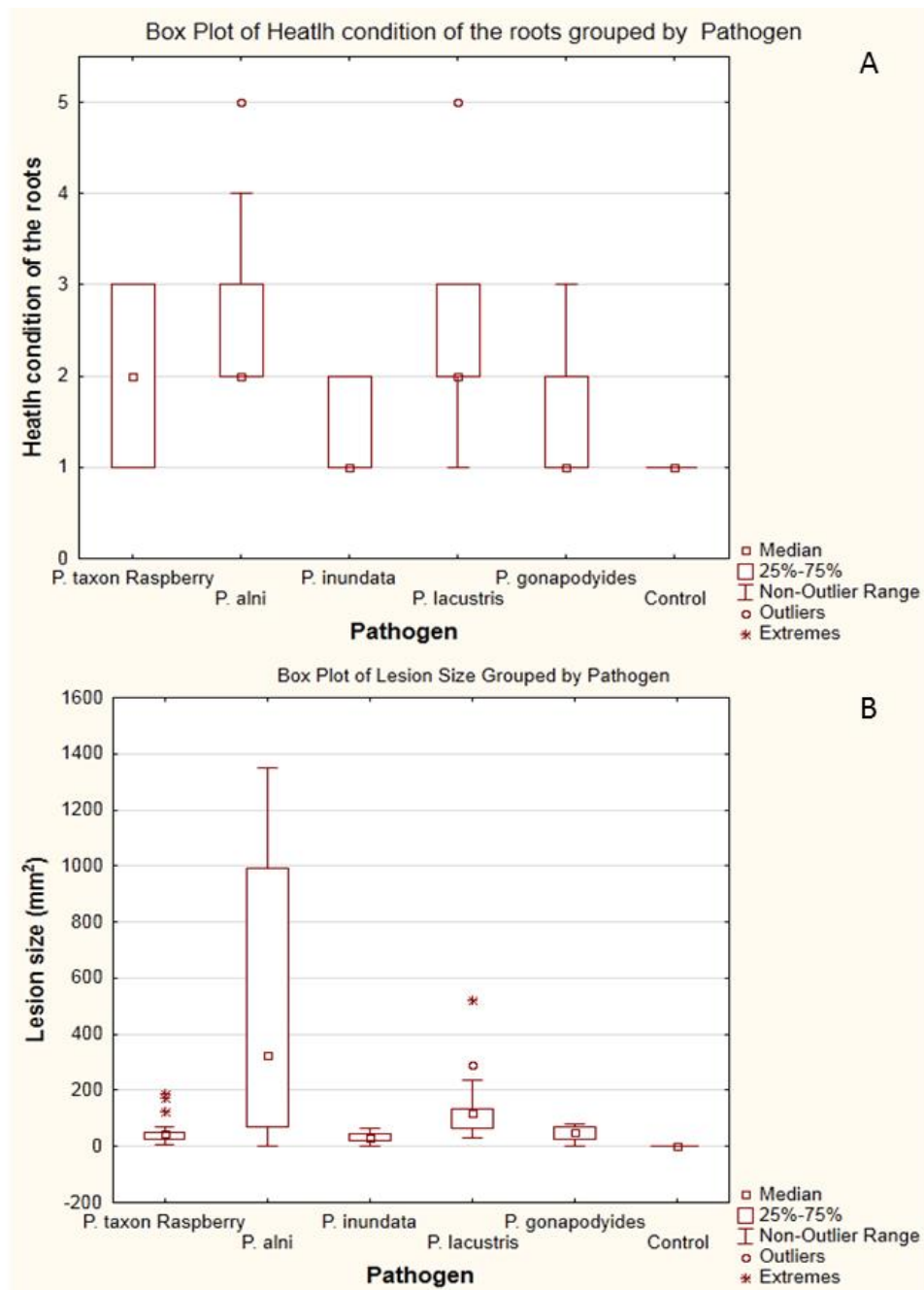


Figure 6: A: Health condition of the roots (soil infestation). B: Average size of the stem lesions (mm<sup>2</sup>) caused by the different pathogens.

#### 4 DISCUSSION

The collar symptoms of alder trees observed at the sampling site predominantly included cankers and tarry spots remaining from an earlier *Phytophthora alni*-infection. Active collar symptoms with fresh exudates were observed altogether on three different trees, at one sampling time each. This explains that the health condition of the trees seems to stagnate based on the dataset referring to collar symptoms. The observed crown symptoms are secondary symptoms ensuing from a potential root infection. The negative tendency of the health condition based on the crown symptoms, and the increasing number of dead trees may be in correlation with the *Phytophthora* species found in the rhizosphere of the investigated trees.

Both morphological and molecular methods were used to identify the collected isolates. Changes in the local composition of *Phytophthora* species were observed during the monitoring period. Weather extremities and changes in the soil moisture content may correlate stronger than seasonality to the observed alterations in species composition. Regarding *P. alni*, it has been isolated from the rhizosphere of two trees only, although it was not present in the soil between 2003 and 2008 (Szabó et al. 2013). In recent studies, we have isolated *P. alni* ssp. *multiformis*, whereas before 2003 *P. alni* ssp. *uniformis* was present in the stand. Based on the study of Marcais – Husson (2014), the presence of *P. alni* suggests that all trees on the studied site might again be attacked by *P. alni*. Conclusively, completely symptomless alder trees might harbour *P. alni* in their root system and as such they become the main inoculum source (Elegbede et al. 2010). Other *Phytophthoras*, like those in clade 6, are consistently dominant in forest soil with the permanent presence of water as was the case with our sampling area. However, *P. gonapodyides*, a species which had been multitudinous, was only once isolated during recent surveys while *P. lacustris* became the most frequently isolated *Phytophthora*. *P. lacustris* seems to be significantly more aggressive than *P. gonapodyides* on common alder. The new appearance of *P. alni* and the continuous presence of *P. lacustris* may explain the considerable condition decline based on crown symptoms of the trees in 2011.

The *Phytophthora* species chosen for the pathogenicity tests are soil-borne pathogens. They primarily damage the root system. Symptoms in the aboveground parts usually appear with a delay. This is why the soil infestation tests showed no significant differences in the health condition of the shoots, but they did reveal significant differences in condition of the root system between the treatment groups.

Among the studied *Phytophthoras*, the aggressive *P. alni* is able to infect trees through wounds or lenticels of the trunk. In this case, it damages the transport vessels in the roots and stem. The change of water and assimilates fails between the roots and the crown. The process leads to the death of the tree (OBwald et al. 2014). This may explain the decline of root conditions in the *P. alni*-infected group in comparison with the other groups by the wound-inoculation test.

In a pathogenicity test Szabó and Lakatos (2008) used only the stem inoculation method on alder saplings with one isolate each of *P. gonapodyides*, *P. alni* ssp. *uniformis*, *P. inundata* and a then unknown taxon similar to *P. lacustris*. The average lesion size caused by that *P. gonapodyides* isolate was more than two times larger (93.49 mm<sup>2</sup>) than the lesions caused by our isolate. This suggests quite a high intraspecific variation in aggressivity. Our *P. alni* ssp. *multiformis* isolate caused lesions that were almost three times larger than the ones caused by *P. alni* ssp. *uniformis* isolate (172.79 mm<sup>2</sup>). The average lesion size caused by their *P. inundata* isolate (36.76 mm<sup>2</sup>) is comparable to ours. Brasier and Kirk (2001) used, among other *Phytophthora* spp., *P. alni* and *P. gonapodyides* isolates for wound inoculations. In accordance with our results, they found that *P. alni* was the most aggressive *Phytophthora* species on common alder, while lesions caused by *P. gonapodyides* did not differ considerably from those on the control logs. Furthermore, Nechwatal et al. (2013) carried out both stem inoculations and soil infestations. In the case of root infection, both *P. gonapodyides* and *P. lacustris* significantly reduced the total dry mass of the roots of common alder saplings (Nechwatal et al. 2013). Both species caused similarly small lesions. The lesions did not differ much from those on the non-infected control saplings, while they were significantly smaller than the lesions caused by *P. alni* in their case study (Nechwatal et al. 2013). Our results with *P. gonapodyides* are well matched with their results, but with *P. lacustris*, our results suggest a higher intraspecific variability in pathogenicity.

*P. inundata* seems primarily to be a parasite of riparian woody species but also of some tree species like *Juglans regia* or *Prunus* spp. under horticultural circumstances (Brasier et al.

2003). In forest ecosystems, it was affiliated with root rot of olive (*Olea europea*) and species of the genera *Fagus*, *Castanea*, *Salix* and *Alnus* (Brasier et al. 2003, Safaiefarahani et al. 2013, Szabó – Lakatos 2013). It may cause the death of the host trees after heavy rainfalls and floods (Brasier et al. 2003). Therefore, after the extremely wet year of 2010, it is not surprising that we found *P. inundata* at our very first sampling.

*P. taxon Raspberry* is a clade 6 taxon, like its close relatives *P. lacustris*, *P. gonapodyides* and *P. inundata*. Data on pathogenicity tests with this species, however, are not available. Still, it is suggested that it may be an opportunistic pathogen similar to the other clade 6 species (Jung et al. 2011). The results of our pathogenicity tests support this hypothesis.

## 5 SUMMARY

A diverse *Phytophthora* community was present in the rhizosphere of the tested common alder trees. Of the eight species we found, only *P. lacustris* was isolated continuously. *P. sp. 'oaksoil'* and *P. taxon Raspberry* were found twice, while the other five species were only found once each. The highly aggressive *P. alni* was detected at the site again after more than ten years. At the very first sampling time, *P. alni* ssp. *multiformis* was isolated from the rhizosphere of two trees. However, earlier *P. alni* ssp. *uniformis* caused epidemic in the stand. Instead of *P. gonapodyides*, the more aggressive *P. lacustris* became the dominant *Phytophthora* species. *P. inundata*, *P. plurivora* and *P. taxon hungarica* were present in the soil, as earlier. However, instead of *P. megasperma*, *P. polonica* and *P. sp. 'oaksoil'* could be isolated.

Although there was no *Phytophthora*-epidemy observed at our representative sampling site, the decline of the trees was significant. Active collar symptoms were observed only in a few cases, while the number of dead trees increased and the health condition of the trees decreased, especially in the first year of our study.

Both pathogenicity tests confirmed the different pathogenicity of the isolated *Phytophthoras* on common alder saplings. Based on our results, the most aggressive *Phytophthora* pathogen of common alder is *P. alni*, while the second most aggressive is *P. lacustris*, followed by *P. taxon Raspberry* and *P. inundata*. *P. gonapodyides* was not confirmed to be pathogenic to alder roots. However, with its small lesions it proved to be slightly pathogenic to common alder saplings upon wound inoculations. Our results suggest that the appearance of *P. alni* and the continuous presence of *P. lacustris* may correlate with the decline of alders.

Our pathogenicity tests conclusively demonstrate the different pathogenicity of *Phytophthora* spp. on young alder saplings. However, their aggressiveness might be weaker on mature trees, thus the development of decline may be long-continued.

These results are based on a two-year-long field experiment, conducted in a representative sampling site of a marshland common alder forest. The changes observed during the survey period might also be true for similar forest stands surrounding the sampling site.

In order to fully understand the epidemiological and ecological aspects of alder decline, it might be necessary to correlate the abiotic environmental conditions of the sampling site with the observed results.

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## Effect of Pavement Stiffness on the Shape of Deflection Bowl

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**Abstract** – The paper introduces a new method for calculating the elastic moduli of pavement layers. The method requires only two input parameters: the thickness of the upper „bound” layer and the Falling Weight Deflectometer (FWD) or Improved Benkelman Beam Apparatus (IBBA) measurement data. The authors developed a continuously differentiable regression function, which can be applied to describe the shape of the deflection bowl. Additional parameters of the deflection bowl (e.g. radius of curvature, position of inflexion point) can be calculated based on the regression function. FWD measurements were simulated running the BISAR (Bitumen Stress Analysis in Roads) software on different pavement variations. Outputs of the simulations were further processed with self-developed software. As a result, a series of diagrams were elaborated, by which the elastic moduli of the pavement layers can be determined.

**Stiffness / pavement layers / elastic moduli / deflection bowl / BISAR**

**Kivonat** – A pályaszerkezet merevségének hatása a behajlási teknő alakjára. Útpályaszerkezetek esetében a megfelelő rehabilitációs eljárás kiválasztása igen nagy gazdasági jelentőséggel bír. Ezért a szerkezetek állapotának megfelelő ismerete nélküli döntéshozatal igen költséges lehet. Emiatt különösen fontos, hogy az FWD (Falling Weight Deflectometer) vagy IBBA (Improved Benkelman Beam Apparatus) eszközzel mért elmozdulások elemzésével olyan többletinformációhoz jussunk, ami a döntést megkönnyíti a gyakorló mérnök számára. Az FWD vagy IBBA eszközzel mért deformációs vonalra illesztett függvényből levezetett görbületi sugár ( $R_0$ ) és a burkolatvastagság ( $h$ ) ismeretében a kötött rétegek alján jelentkező megnyúlásokat jól lehet becsülni. A BISAR (Bitumen Stress Analysis in Roads) programmal végzett számítások statisztikai elemzése pedig azt mutatja, hogy a  $D_0$  (központi behajlás) és  $R_0$  (görbületi sugár) paraméterek ismeretében lehetőség nyílik a kötött és szemcsés rétegek modulusának visszaszámolására.

**Merevség / pályaszerkezet rétegek / rugalmassági modulus / behajlási teknő / BISAR**

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## INTRODUCTION

Forest roads with asphalt pavement represent the basis of the forest road networks in Hungary. Properly maintained asphalt pavements offer a high level of service. While traffic load of forest road networks have grown, expenses for their maintenance remained lower than required in the last three decades. As a result, these roads are in poor condition, generally. Renovation projects demand the knowledge of the roads' bearing capacity. The term "bearing capacity", although widely used at pavement management projects, is hard to define. In fact, direct measurement of bearing capacity is impossible. Instead, one can measure the deflection caused by a known load, and calculate the bearing capacity afterwards.

The traffic transfers its loads to road pavements through the tyres of the vehicles. Due to this, shearing stresses originate from vertical loads (pressing, beating, shaking, bending etc.) and horizontal stresses (braking, accelerating, wearing) (Kosztka 1978, 1986). These stresses affect each pavement layer differently, such as the elastic and plastic (permanent) deformation, the break and the structural realignment (Boromisza 1976). All these structural changes appear on the surface of the pavement as deformations, and the so-called deflection bowl or deformation surface forms.

To measure the evolving deformations several methods have been elaborated. Currently the measuring procedures based on absorbed oscillation are widely used. These are called Falling Weight Deflectometers, FWD. These deflectometers, operating with impulses, often drop a given weight from a given height onto a disc with anti-shock – using the potential energy – then they record the evolving displacements (Kosztka et. al. 2008). Researchers of the Institute of Geomatics and Civil Engineering at the University of West Hungary developed a new instrument to measure the full deflection bowl with the Benkelman beam (Markó et. al. 2013). The development was based on the Benkelman beam, extending its properties with automated data logging and the ability to measure multiple points of the deflection bowl. The Improved Benkelman Beam Apparatus (IBBA) continuously measures the vertical displacement of one point on the surface of the pavement, together with the horizontal position of the truck (*Figure 1*).



*Figure 1. Falling Weight Deflectometer (left) and Improved Benkelman Beam Apparatus (right) in action.*

The deflection bowl recorded during the test provides much more information about the current state of the pavement structure than the central deflection in itself. Therefore we can define its bearing capacity, remaining lifetime, and the thickness of the needed strengthening layer more precisely. Choosing the applicable rehabilitation procedure in the case of a given pavement structure has a really great economic significance. Without appropriately knowing the condition of the pavement, decisions could become very expensive. This is why it is so important to gain additional information by analysing the deflections, which makes it easier for the practising engineers to make decisions. We started our work with this approach, and summarized our results in this paper.

## 1 MATERIAL AND METHOD

### 1.1 Estimating the deflection bowl with functions

When deflection is measured to define the bearing capacity, displacements are measured and recorded only in certain distances from the load. This makes it necessary to fit functions onto the discrete measurement points to get the complete plot of all the evolved deflections. It is practical to apply functions describing the deflection bowl, because this way the geometrical attributes that are important regarding the stressed pavement can be defined with comparatively easy calculations.

Because of the surface sinking caused by mining (e.g. tunnel building), functions were already elaborated long time ago. Most authors (Aversin, Martos, Beyer, Bals etc.) suggested functions similar to the Gaussian bell curve (Fazekas 1978). Suggestions can be found to describe the deformation curve of pavements in Hothan and Schäfer's (2004) summary work.

Hossain (1991) used exponential function to estimate the deformation caused by external load:

$$D(x) = ae^{bx} \quad (1)$$

where  $x$  : distance from the centre of the load [m] and  $a, b$  : parameters.

On the basis of their examinations the effect of the upper and stiffer layers can be experienced in the decrease of the "a" parameter, while the effect of the lower high-solidity layers causes increasing "b" parameter value. The "a" and "b" parameters depend on the strength of materials characteristic of the pavement. The exponential function is able to estimate the FWD or IBBA measurements with high correlation, though it cannot reproduce the natural shape of the deflection bowl. Therefore it is not suggested to use it despite the high correlation coefficient.

Jendia (1995) tries to describe the whole deformation curve by substituting the exponential function in the central range of the deformation line  $0 \leq x \leq r$  with a hexic polynomial:

$$D(x) = \begin{cases} c_3x^6 + c_2x^4 + \\ +c_1x^2 + c_0 & 0 \leq x \leq r \\ ae^{bx} & x \geq r \end{cases} \quad (2)$$

Jendia first defines "a" and "b" values of the unknown parameters. He specifies the second derivative's equality at the joint of the functions, that is, the continuity of the curve. Therefore, there are three constraints for the  $c_3, c_2, c_1$  and  $c_0$  parameter.

He creates the last independent variant iteratively by minimizing the difference between the values measured on the second and third sensor of the FWD device and the calculated deflections. The method of Jendia can reach only low equality with the data points in spite of its high demand of calculation (*Figure 2*).

Grätz (2001) makes it possible to describe the deflection bowl with a single function:

$$D(x) = \frac{w_a + w_b x^2}{1 + w_c x^2} \quad (3)$$

With the help of the suggested rational fractional function<sup>1</sup> the three factors that describe the whole bowl can be defined (*Figure 2*, Grätz (1)). The correspondence with the measured results can be further increased if a fourth degree of the polynomial is used:

<sup>1</sup> The rational fractional function is a mapping of the set of scalars, where we give the association with the quotient of two polynomials.

$$D(x) = \frac{w_a + w_b x^2 + w_c x^4}{1 + w_d x^2 + w_e x^4}. \quad (4)$$

Using the altered function higher correlation can be achieved, though the layer parameters cannot be concluded from the equation's factors as the coefficients depend on all the layers differently (Figure 2. Grätz (2)). To describe the deformation curve in practice, it is suggested to apply functions with which one bowl parameter can be deduced, which describes a special layer of the pavement (e.g. radius of curvature).

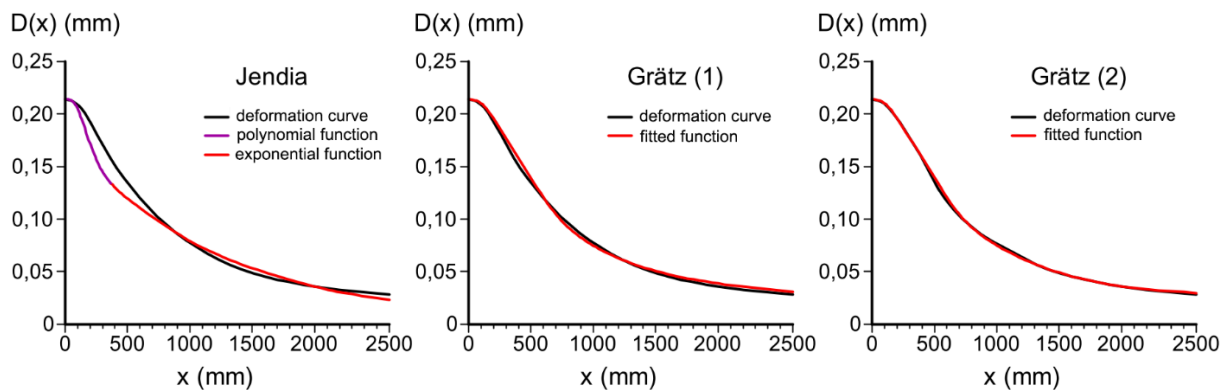


Figure 2. Comparing different deflection bowl functions (Hothan and Schäfer 2004)

In his study, Daehnert (2005) introduces two function types from the French literature (Ph. Leger and P. Autret) which shows good correspondence with the theoretical deformation curve:

$$D(x) = D_0 e^{-(x^2b)} \quad (5)$$

and

$$D(x) = D_0 \frac{a}{x^2 + a} \quad (6)$$

where  $D_0$  the maximum deflection in the axis of the load [m].

The function (6) was originally developed to process the Lacroix deflectograph data. In its structure, it is similar to the Agnesi<sup>2</sup> Witch Curve (Scharnitzky 1989). Cser (1961) uses function (6) to model the evolving deformations directly under wheel load with the substitution of  $a = 3r^2$

$$D(x) = D_0 \frac{3r^2}{x^2 + 3r^2} \quad (7)$$

where  $r$  the radius of the loaded surface considered to be evenly dissolved and circle shaped [m]. The curve has an inflexion point at the edge of the wheel load ( $x = r$ ). The function can follow the evolving deformations only in a restricted extent, as the inflexion point is fixed.

<sup>2</sup> Maria Gaetana Agnesi (Milan, May 16th, 1718 – Milan, January 9th, 1799), Italian linguist, mathematician and philosopher, the honorary member of the University of Bologna.

## 1.2 Estimating the deformation curve on the basis of mechanics functions

Starting with the Boussinesq stress formulas, the value of  $D_0$  deflection under the centre of the  $d = 2r$  diameter flexible circle plate can be deduced (Papagiannakis and Masad 2008):

$$D_0 = \frac{2pr}{E_e}(1 - \mu^2) \quad (8)$$

where

$D_0$  : vertical deflection measured in the load axis [mm],

$E_e$  : the modulus of the flexible halfspace [MPa],

$p$  : surface distributed load [MPa],

$r$  : radius of the loading plate [mm],

$m$  : the Poisson factor [-].

Beside the central deflection, Odemark calculated the deformation curve of the flexible halfspace with  $E_e$  modulus, loaded in the usual way using the  $y = f(p, r, E_e)$  function. The second differential at its  $x = 0$  point estimates the curve's value very well. The  $R_0$  radius of curvature, in the case of one-layer half-space, can be calculated with the following formula (Nemesdy, 1985):

$$R_0 = \frac{E_e r}{p(1 - m^2)}. \quad (9)$$

Both functions give the same result in the case of homogeneous, infinite halfspace, so it is obvious that there is functional relation between the central deflection and the radius of curvature. Take the quotient of the equivalent modulus provided by the two equations:

$$c = \frac{2r^2}{R_0 D_0} \quad (10)$$

where "c" factor is the quotient of the two moduli, which is  $c = 1$  in the case of homogeneous infinite half space.

It requires very long calculations to define deformations on the surface of the homogeneous half-space using Boussinesq's theory. Therefore, to simplify this, it is practical to take an estimating function. When determining the estimation function it is necessary to start from the geometric restrictions that are the boundary conditions. On the base of the above functions the following conditions can be stated:  $x = 0$ , where  $D(x) = D_0$ , and the second derivative  $D''(x) \gg 1/R_0$  of the requested  $D(x)$  function at  $x = 0$ . Additionally, the defined  $D_0$  and  $R_0$  values require mechanics conditions (10), too. Searching for the function that satisfies the conditions we start with the function type suggested by Cser (1961):

$$D(x) = D_0 \frac{d^2}{c \times x^2 + d^2} = D_0 \frac{1}{c \left(\frac{x}{d}\right)^2 + 1} \quad (11)$$

In the suggested function "c" is the so-called *shape factor*, which influences the shape of the deformation curve (Primusz – Tóth 2009; Primusz – Markó 2010). It can easily be seen that the estimation function really satisfies the  $x = 0$  and  $D(x) = D_0$  conditions. After defining the shape of the deflection bowl we can define the radius of curvature (*Figure 3*).

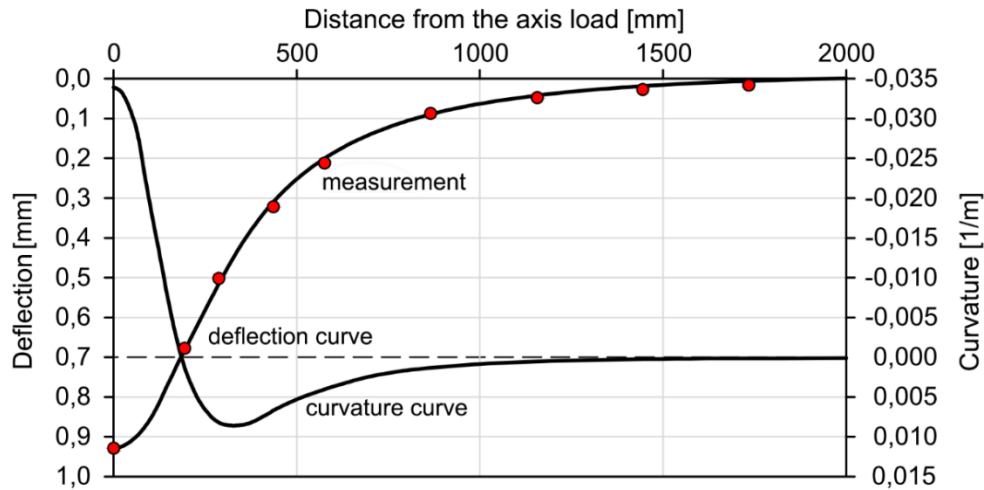


Figure 3. Curve of deflection bowl and curvature under load

We estimated the radius of curvature of the oscular circle belonging to an optional point of the  $D = D(x)$  function with the  $k(x) \gg D''(x)$  function:

$$k(x) \approx \frac{\vartheta^2}{\vartheta x^2} \left( \frac{D_0 4r^2}{cx^2 + 4r^2} \right) = 8 \frac{D_0 r^2 c (3cx^2 - 4r^2)}{(cx^2 + 4r^2)}. \quad (12)$$

The negative sign of the curve means that in the case of positive bending moment, the centre (0 point) of the oscular circle described with the curve radius is located on the  $-D$  driven side of the axis. The curve alteration is presented in Figure 3. The minimal radius of curvature at  $x = 0$  is

$$R_0 = \frac{2r^2}{D_0 c}. \quad (13)$$

It can be seen that the mechanics condition (10) is also satisfied, so the function is a good estimation of the mechanically defined deformation curve.

### 1.3 Estimation of the strain rising at the bottom of the bound layer

Knowing the radius of curvature derivated from the fitted function on the measured deformation points, and the thickness of the overlay, the strains rising at the bottom of the bound layers can be estimated with the following formula:

$$\varepsilon = \frac{h}{2R_0} = c \cdot D_0 \frac{h}{4r^2} \quad (14)$$

where

- $\varepsilon$  : strain in the load axis,
- $h$  : thickness of the bound layer,
- $R_0$  : radius of curvature in the load axis.

The conditions defined on the bound layer are satisfied if Hooke's law is present and the elastic modulus is equal for compression and for tension (Primusz – Tóth 2009).

### 1.4 Computer simulation with the BISAR software

The simulation is basically an examination where the expected and real behaviour of the system is being studied through the physical or computer model of a process.

Applying the simulation model we are able to provide appropriate inputs for the model of the system, operate it and observe the outputs.

Through the simulation of the pavements, we can observe what deformations evolve at the places of the sensors that record the deflections under external load, typical of the FWD devices, and how much stress evolves in each structural layer. Using the simulation, the deflection curve recorded on the pavements can be provided with more information, therefore, more exact pavement diagnostics are possible (Huang 2003).

#### 1.4.1 Setting the simulation model

Nowadays, the most popular and most accepted method of defining the stresses evolving in pavements is the application of computer software. One of the oldest and most referred software is the BISAR (Bitumen Stress Analysis in Roads), developed by the SHELL Research Center. The software can calculate stress, strain, and deflection in an elastic multilayer system loaded with vertical load. The layers are characterized by their layer thickness, elastic modulus, Poisson factor and the adhesion defined at the boundaries of the layers. The whole system is supported by an infinite elastic half-space.

We used the DOS version of the BISAR software to carry out the simulation, because this way – after generating the starting data files – we could run batched calculations. The data files contained the structure of the pavements to be calculated in one procedure (number, thickness, modulus of layers etc.), the rate and place of load, and the coordinates of points where the calculation of stresses and strains are needed. The BISAR simulation ran in the case of two- and three-layer systems.

#### 1.4.2 Pavement models used in simulation

The layers of the pavements can basically be divided into three groups: subgrade (together with frost protecting and/or improving layers), base layer, and overlay. Each group can be divided into further layers, so an average real pavement can be built of 3–5 layers (*Figure 4. a*). As to their material, the layers can be bound with bitumen (sometimes hydraulic) or can be unbound. As most pavement models are able to consider the material characteristics with the help of the elastic modulus and the Poisson factor, it is suggested to close up the unbound and bound layers and handle them as a whole instead of increasing the number of layers (Yoder – Witzak 1975).

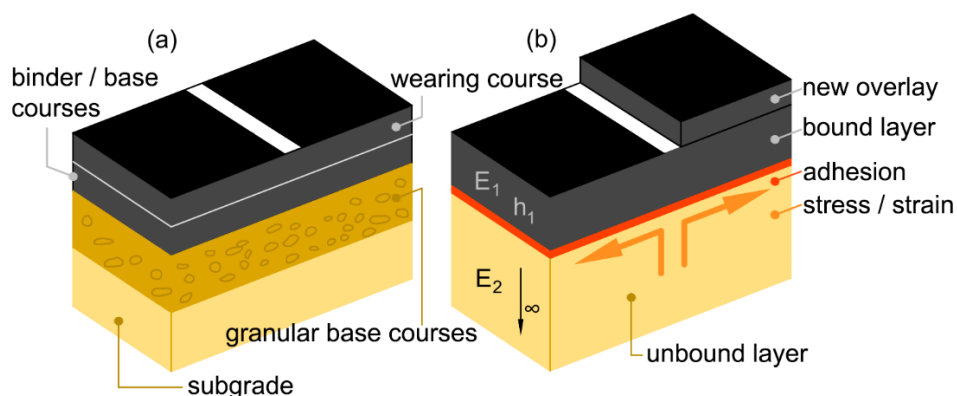


Figure 4. The structure of the pavement models in the simulations

Accordingly, bound and unbound pavement layers can be distinguished (*Figure 4. b*). The pavement behaviour models are able to consider the collaboration defined between the layers. Full slip should be assumed between the unbound granular layers and the bound overlays, while in the case of reinforcement – between the old and new overlays – full

adhesion should be expected, even if it causes smaller stress to the old asphalt layers. During simulation we examined the behaviour of the existing pavements with two-layer systems, while we used three-layer systems in the case of reinforced pavements. The two-layer systems are the idealized models of the existing pavements, in which the bottom layer refers to the unbound granular layers and subgrade, while the top bound layer refers to the overlays. There is probable frictional adhesion between the bound and unbound layers. In the case of the two-layer pavement models, the modulus of the top bound layer is between 1000 and 8000 MPa, the modulus of the bottom unbound half-space varied between 20 and 500 MPa. The thickness of the top bound layer varied between 50 and 500 mm, and we divided the examined range logarithmically examining 7 different values. Thus, we examined the two-layer systems in  $12 \times 12 \times 7 = 1008$  combinations (Table 1).

Table 1. The parameters of the pavement models examined during simulation

Model	Nr.	Modulus	Var.	Poisson	Thickness	Var.	Adhesion	Sum
Two-layered	1	20 – 500	12	0,5	Infinite	–	1	1 008
	2	1000 – 8000	12	0,5	50–500	7		
Three-layered	1	20 – 500	12	0,5	Infinite	–	1	15 552
	2	1000 – 8000	12	0,5	50–300	6		
	3	5000 – 15000	3	0,5	20–120	6	0	

Notes: Layer ordinal (Nr.) from bottom to top, Layer-modulus (Modulus [MPa]), Variation (Var.), Poisson-factor [–], Thickness [mm], Adhesion [0: full adhesion, 1: full slip], All variations (Sum)

The three-layer models evolve from the two-layer systems with the addition of a reinforcement layer; this reinforcement layer can help in the examination of the bearing capacity of pavements. During calculations the rigidity modulus of the new asphalt layer used for reinforcement was 5000, 10000 and 15000 MPa. We then increased the thickness of the reinforcement layer by 2 cm up to 12 cm, so altogether 15552 variations evolved (Primusz – Markó 2010). Beside the layer modulus and thickness, the Poisson factor gets different values in the case of different materials; however, its practical definition is rather difficult as its value depends on tension and temperature (Pethő 2008; Szentpéteri – Tóth 2014). In the case of general road construction materials its value is usually between 0.2 and 0.5. The effect of the cross contraction factor's changes was examined in detail by De Jong, Peutz and Korswagen (1973), and Tam (1987). The researchers stated that changing the Poisson factor had little effect on the primary design parameters (strain, stress, deflection). That is, the effect on deflections caused by changing the Poisson factor is rather small compared to the layer thickness or the layer modulus. If the Poisson factor is increased from 0.2 to 0.5, the deflections will decrease by only few percent (Van Gurp 1995). Based on the above research in the BISAR simulation, we took each layer with the value  $\mu = 0.5$ . In this case, the analytical functions will largely be simplified; therefore, it is – in several respects – practical to choose this value.

### 1.4.3 Calculating stresses

The BISAR software is able to handle several loads and calculate their superposition. The loads and the examined points are placed in one frame of reference and can be arbitrarily defined by the  $x$ ,  $y$ ,  $z$  coordinate triplet. During the simulation we took size  $F = 50$  kN single wheel load, which affects the top layer vertically, and scatters evenly a radius  $r = 0.15$  m elastic circle plate ( $p = 0.707$  MPa). The distance of the examined points – measured from the

load axis – was equal to the usual sensor set-up of the FWD device. In respect to the asphalt overlays' lifetime, the most important stress is the strain of the bottom edge line caused by the vertical deflection load. Therefore, in the fixed positions we examined not only the vertical deflections, but the strains defined on the bottom plane of the bound layers. After running the BISAR software, we evaluated the result text files with a self-developed program. We considered the vertical deflections calculated by the BISAR as a result of an FWD measurement during our further analysis.

## 2 RESULTS AND DISCUSSION

During the first part of the evaluation we examined how much effect each layer had on the evolving deformations in the case of a given pavement. This question is described in detail in Van Gorp's study (1995).

Figure 5 shows how much the layers of a three-layer structure effect the surface deflections (Van Gorp 1995). Naturally, the distribution changes with the modifying of the layer thickness or stiffness. According to the study, the thicker and stiffer the upper layers are, the more important the darkened areas of Figure 5 become. The figure demonstrates well that the bearing capacity of subgrade has the greatest influence on the peak value of the surface deflections, and 900 mm from the load axis, the measured deflection represents the deflection of the subgrade in 100%. It can also be observed that if we consider the subgrade and base layer as one, then the effect of the top bound (asphalt) layer expands only 300 mm from the load axis, so it mostly affects the central deflections. This theory can be examined with the BISAR simulation.

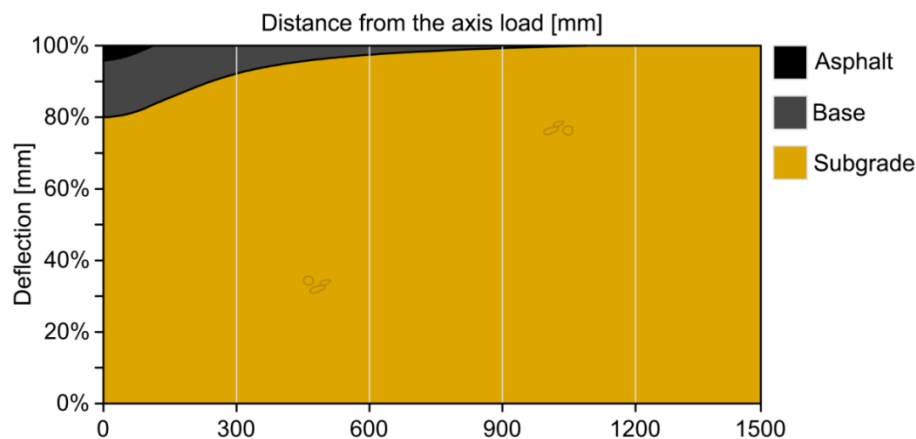


Figure 5. The effect of each layer on the surface deflection (Van Gorp 1995)

In the case of the examined pavement models, we calculated the maximum deflections on the surface, and also the partial deflections evolving at the bottom of the bound layer with the BISAR software. Thereby we had both deformation curves by structure. We fitted the function (11) onto these deflections, which resulted in two “c” shape factor values. By comparing the two values it can be stated that between the deformation curves evolving on the surface and at the bottom of the bound layer, the difference depends on the ( $h$ ) layer thickness. This relation is graphically presented in Figure 6; the surface deformation curve is marked as  $c_t$ , while the one being at the bottom of the bound layer is marked as  $c_b$  (Figure 6. a). The thicker the bound layer is ( $h$ ), the bigger the extent of the difference is (Figure 6. b). The differences of shape factors can be originated from the differences of deflections. Comparing the deflections of the 1008 two-layer systems, only the central deflections showed measurable differences in the range of 0–1 mm (70% of the  $D_0$  values fell in between).

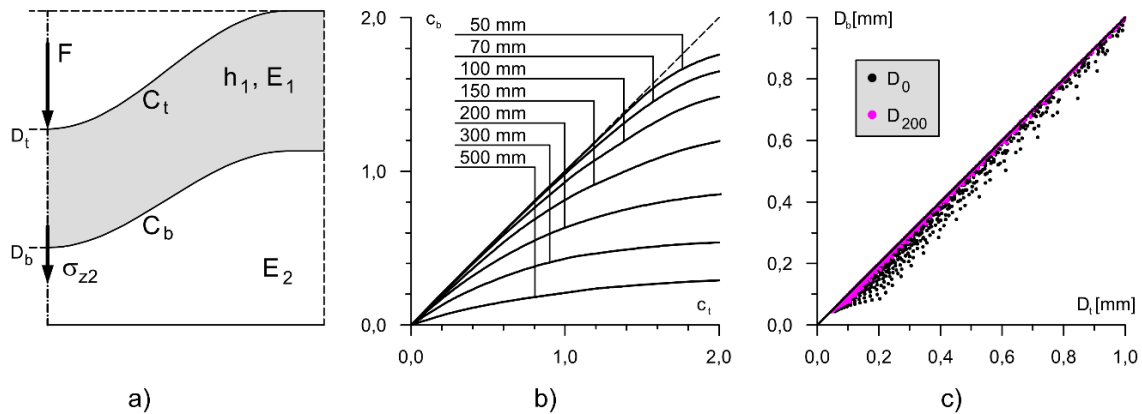


Figure 6. The changes of the “c” parameter of the function fitted onto the deformations evolving on the surface and at the bottom of the bound layer

Going farther from the load axis (200–300 mm) the deflections calculated on the surface and at the bottom of the bound layer were entirely the same (this result also confirms our statements at the beginning of this chapter). That is, the differences of shape factors can mainly be explained with the changes of the central deflection, as the thickness of the bound layer influences the compression of the layer itself (Figure 6. c).

Now, if we examine the results from the practical aspect and accept the assumption that the deflections measured on the pavement surface are nearly the same as the ones evolving at the bottom of the bound layer, that is  $D_t(x) \approx D_b(x)$ , then the shape factor ( $c_b$ ) that characterizes the bottom of the bound layer can well be estimated on the base of the surface measurement:  $c_t \approx c_b$ . As the practical measurements always have the possibility of mistakes, and there are several factors (e.g. temperature) that modelling cannot count with, hereafter we will not make any difference between the two shape factors.

## 2.1 Analysis of the two-layer system

Using the results of the BISAR software we looked for relationship between the parameters deduced from the shape of the deflection bowl (Primusz and Tóth 2009), and the layer parameters of the two-layer system.

The examination revealed that the “c” shape factor, the quotients of the layer moduli ( $K$ ) and the thickness of the bound layer ( $h$ ) have very close correspondence. The graphical evaluation of the results is shown in Figure 7.

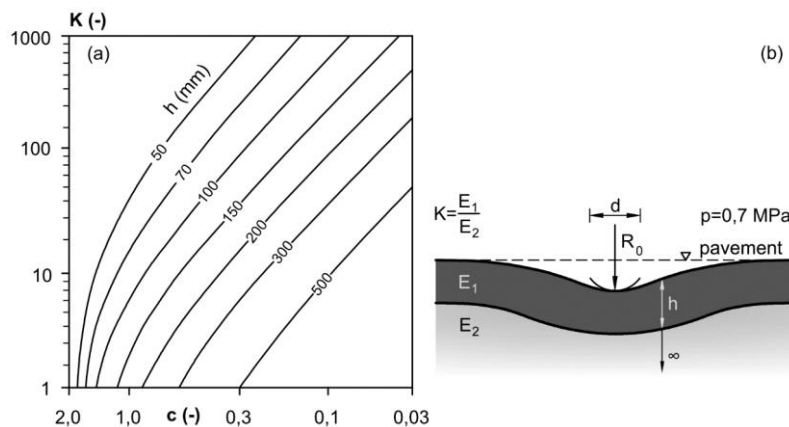


Figure 7. Function between the shape factor ( $c$ ) and the rate of the layer moduli ( $K$ )

With the help of the graph the idealized two-layer model of a given pavement – knowing the bound layer thickness – can be induced from the FWD or IBBA measurements. We made the statistic model of the  $K = f(c, h)$  function connection in two steps. The examinations showed that the rate of the  $K$  moduli and the  $\sigma_{z2}$  compressive stress evolving on top of the bottom layer are related similarly to the one in *Figure 7*. As the  $K$  and the  $\sigma_{z2}$  values do not depend on the accuracy of the function fitted on the deflections – that is the “c” shape factor – it is practical to first describe this relationship. We received the best result using the following model:

$$Y = \frac{a}{b \cdot X^c + 1} \quad (15)$$

with the next substitution:  $Y = \sigma_{z2}$  and  $X = h^d \sqrt{K}$ . We defined the model's  $a$ ,  $b$ ,  $c$  and  $d$  parameters with the STATISTICA program:

$$\sigma_{z2} = \frac{0,8}{20,816 \cdot (h^{1,4} \sqrt{K})^{1,393} + 1} = \frac{0,8}{20,816 \cdot h^{1,95} K^{0,70} + 1} \quad (16)$$

The accuracy of fitting is clearly shown by the very high  $R^2 = 0,9977$  value. Then we looked for a relation between the “c” shape factor and the  $\sigma_{z2}$  value. There was clear polynomial function with  $R^2 = 1$ :

$$\sigma_{z2} = 0,0392c^6 - 0,2749c^5 + 0,6907c^4 - 0,8332c^3 + 0,5424c^2 + 0,2588c \quad (17)$$

After plotting the value pairs, the hexic polynomial could be estimated with a line without the significant decrease of the fitting's rate ( $R^2 = 0,9954$ ):

$$\sigma_{z2} \approx 0,4205 \cdot c. \quad (18)$$

Substituting function (18) in function (16), we get the wanted relationship:

$$c \approx \frac{1,9}{20,816 \cdot h^{1,95} K^{0,70} + 1} \quad (19)$$

or reordering to  $K$ :

$$K \approx 0,0131 \left[ h^{-1,95} \left( \frac{1,9}{c} - 1 \right) \right]^{1,428} \quad (20)$$

the  $K$  factor here shows the stiffness of the layers correlated with each other.

## 2.2 Estimating the modulus of the granular layers

According to the study of Hoffmann (1988), if the pavement and the subgrade are considered as a two-layer system, knowing the radius of curvature and the central deflection, the E-modulus of the subgrade can be directly calculated. This statement can be checked knowing the results of the BISAR simulations. Using the deformation curves of the 1008 two-layer systems, the radius of curvature of the systems can be defined with the function (13). The related  $D_0$ ,  $R_0$  and  $E_2$  data rows are graphically presented in *Figure 8*.

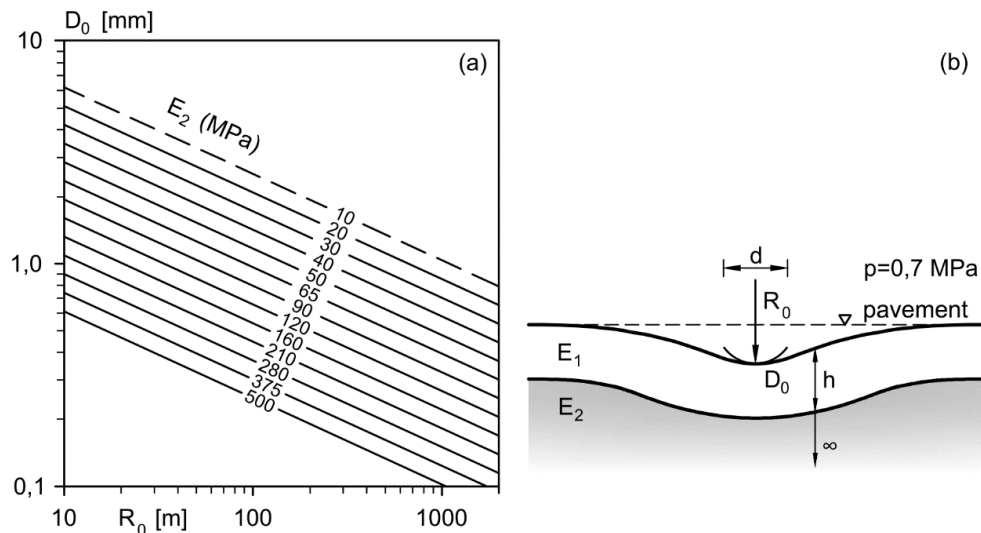


Figure 8. The E-modulus of the bottom layer can be defined by the centre deflection and the radius of curvature

Figure 8 shows that the data rows define a plane in dual logarithmic frame of reference. So the E-modulus of the bottom layer can be defined by the following function relation:

$$E_2 = a \cdot D_0^b \cdot R_0^c \quad (21)$$

The linear figure of the function:

$$\log(E_2) = \log(a) + b \log(D_0) + c \log(R_0) \quad (22)$$

The following general equation can be derived with the  $Y = \log(E_2)$ ,  $X_1 = \log(D_0)$ ,  $X_2 = \log(R_0)$ ,  $b_0 = \log(a)$ ,  $b_1 = b$  and  $b_2 = c$  substitution:

$$Y = b_0 + b_1 X_1 + b_2 X_2 \quad (23)$$

Here  $b_0$  represents the intersection, while  $b_1$  and  $b_2$  show the partial slopes. The regression factors can be defined with the least square method as before (Orbay 1990). The results of the calculations made with the STATISTICA program are shown in Table 2.

Table 2. The statistic characteristics of the function constants fitted onto the two-layer system

h (cm)	N = 1008	$\beta$	Deviation ( $\beta$ )	B	Deviation (B)	t(1005)	p
Intersection		–	–	3.08794	0.000767	4024.03	0.00
5–50	Direction tangent (1)	–1.56581	0.000598	–1.62284	0.000620	–2617.99	0.00
	Direction tangent (2)	–1.05669	0.000598	–0.62894	0.000356	–1766.77	0.00

$R^2 = 0.9998$ ,  $F(2.1005) = 3515439$ ,  $p < 0,0000$ ,  $\alpha = 0,05$  and the residual deviation: 0.00525

According to the examination, there is very strong correspondence between the E-modulus of the bottom layer, the vertical deflection interpreted at the load axis, and the radius of curvature. The high  $R^2$  also shows this. So the  $E_2$  modulus can be estimated with the following function from the evolving deflections:

$$E_2 = 1224,45 \cdot D_0^{-1,623} R_0^{-0,629} \quad (24)$$

To avoid big numbers we gave the  $D_0$  in millimetres, the  $R_0$  radius of curvature in metres, while the joint modulus of the granular layers  $E_2$  in MPa in the equation. According to the function (13)  $R_0$  depends on the “c” shape factor, therefore, the (24) can be changed as follows:

$$E_2 = 111,73 \cdot D_0^{-0,994} \cdot c^{0,629} \quad (25)$$

According to the statistical model, it is not necessary to know the bound layer thickness to estimate the joint modulus of the granular layers, so it can be defined from the FWD or IBBA measurements without any destruction. The defined condition parameter may be useful especially for the Pavement Management Systems (PMS).

### 2.3 Estimating the modulus of the bound layers

The definition of the modulus of the bound layers is done using the following simple formula:

$$E_1 = K \cdot E_2 \quad (26)$$

where

- $E_1$  : modulus of the bound layer [MPa],
- $E_2$  : modulus of the unbound granular layer [MPa],
- $K$  : rate of the layers compared to each other [-].

In the function (26),  $K$  is the rate of the bound and unbound layers compared to each other, which is calculated with the formula (20). To estimate the  $E_2$  modulus we use the formula (24). Therefore we proved that in the case of two-layer pavement models, the moduli of the layers can unequivocally be calculated back from the deformation curve, so it is not necessary to use the iterative backcalculation methods.

### 2.4 Analysing the three-layer system

With the help of the BISAR software we modelled 15552 three-layer pavement variants. We calculated the evolving stresses and strains at the bottom of the reinforcement layers ‘built onto’ the original pavements and the whole bound layer thickness. The calculation was based on Ambrus’s (2001) former results. He demonstrated that at the bottom of the reinforcement layer of pavements having the same deflection curve but different structure, the same strains evolve in every case. That is, if the pavement deflection curve (its radius of curvature) is known, then the rate of the necessary reinforcement can be directly estimated.

We could not find regression relationship between the  $R_0$  radius of curvature of the deflection curves calculated with the BISAR software and the strains evolving directly at the bottom of the reinforcement layer. The reason for this is that we assumed full adhesion between the two layers, so it actually behaved as one layer. Therefore, we later only dealt with the strains evolved at the bottom of the whole bound layer thickness. We managed to draw the following statistic model:

$$\log(\varepsilon_b) = -0,522 \cdot \log(R_0) - 0,533 \cdot \log(\Delta h) - 0,189 \cdot \log(E_{AC}) + 5,088 \quad (27)$$

or

$$\varepsilon_b = 122463 \cdot R_0^{-0,522} \cdot \Delta h^{-0,533} \cdot E_{AC}^{-0,1888} \quad (28)$$

where

- $\varepsilon_b$  : strain evolving at the bottom of the bound layer after the reinforcement [ $\mu\varepsilon$ ],
- $R_0$  : the radius of curvature of the pavement before the reinforcement [m],
- $\Delta h$  : the thickness of the reinforcement layer between 20 and 120 mm,
- $E_{AC}$  : the modulus of the reinforcement layer between 5000 and 15000 MPa.

The results of the calculations carried out with the STATISTICA software are shown in Table 3. Knowing  $\varepsilon_b$ , it is possible to define the necessary asphalt reinforcement layer. To do this, the asphalt fatigue functions used in asphalt mechanics have to be applied. The principle of the method is that the material is able to tolerate a certain strain during limited load repetitions without failure. That is why the evolving  $\varepsilon$  strain is equivalent with a repetition number, such as a unit axis crossing number (Ambrus 2001). The fatigue function of the material should be defined with laboratory examinations, though today, several estimating functions can be used (Bocz 2009).

Table 3. The statistic characteristics of the function constants fitted onto the three-layer system

h (cm)	N = 15 552	$\beta$	Deviation ( $\beta$ )	B	Deviation (B)	t(15548)	p
	Intersection	–	–	5.08800	0.01863	273.09	0.00
2–12	Direction tangent (R)	–0.83663	0.002617	–0.52207	0.00163	–319.69	0.00
	Direction tangent (H)	–0.42522	0.002617	–0.53302	0.00328	–162.49	0.00
	Direction tangent (E)	–0.11296	0.002617	–0.18882	0.00437	–43.16	0.00

$R^2 = 0.8935$ ,  $F(3.15548) = 43491$ ,  $p < 0.0000$ ,  $\alpha = 0.05$  and the residual deviation : 0.10747

Currently in Hungarian road maintenance practice, the critical strain is defined directly at the bottom of the reinforcement layer. One reason for this is that the old asphalt layer becomes cracked, so we cannot count on its long-term load-bearing ability. This approach sometimes results in exaggeration, as it expects only the new layer to resist the external loads, while the old asphalt layers are still able to participate in the force-game. Counting with the existing asphalt layers is also hampered by the fact that only the fatigue ability of loose asphalt mixtures could be examined with 2- or 4-point bending test. It is very circuitous to make a test piece out of the core samples drilled out of existing pavement for these examinations. Today, the cracking-drawing test (Indirect Tensile Test, ITT) makes it possible to use samples directly drilled out of the pavement and define its fatigue characteristics (Pethő – Tóth 2012). The old asphalt material's fatigue criterion should be defined with the least squares method from the results of the laboratory experiment:

$$N_f = k \cdot \left( \frac{1}{\varepsilon_0} \right)^n \quad (29)$$

where

$N_f$  : the entire load repetition number,

$k, n$  : material constants,

$\varepsilon_0$  : horizontal strain in  $\mu\varepsilon$  in the centre of the test piece.

Based on the function (29) a statement can be made in connection with the strain of the old asphalt layer. The importance of the function (28) is that the strain can be estimated at the bottom of the existing asphalt layers after the reinforcement. Comparing the two functions the base function of a design procedure can be deduced, which will design upon the fatigue characteristics of the old rather than the new asphalt.

### 3 SUMMARY

The function suggested by us can be fitted not only onto deflection curves calculated with the FWD or IBBA, but also the ones calculated with the BISAR software. We showed that by knowing the deflection curve and the thickness of the bound layer, without using further

iteration procedures (backcalculation), we could define the modulus of the examined pavement's layers. The modulus calculated this way can certainly not be matched with the result of any laboratory tests. The practical benefit of the procedure is that with the defined moduli, we can create a pavement model whose behaviour – shape alterations under wheel load – well approximates the real pavement.

Knowing the radius of curvature we can calculate the strain of the bottom of the bound layer; knowing the strain, we can calculate the existing pavement's lifetime. The analysis of the three-layer models made it possible to estimate the strains evolving at the bottom of the existing asphalt layer after building the reinforcement layer, and so we can establish the theoretical possibility of a harmonic and economic reinforcement design method. The elaborated modelling procedure on the network level has the capability to be the base of a pavement management system. On the project level, the appropriately parameterized two-layer pavement model can help plan more professional reinforcement layers.

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# Effects of Wood Ash on the Chemical Properties of Soil and Crop Vitality in Small Plot Experiments

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**Abstract** – Wood-burning power plants and heating plants produce a great amount of wood ash as a by-product of the combustion process. In 2009 we launched an experiment in which we examined the composition of ash, the nutrient supplying capacity of soil mixed with ash, and the availability of its constituents. In the spring of 2010, we conducted small plot experiments using wood ash applications equivalent to 0; 1; 2.5; 5 and 10 t of wood ash/ha, on slightly acidic clay loam soil using white mustard and rye grass as the test plants. The pH value of the soil rose in a statistically verifiable way as a result of the ash treatments. After the application of ash, the P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O-content of the soil rose significantly; the treatments also increased the magnesium and sulphur content of the arable soil as well as the level of Zinc among the microelements. However, none of the wood ash applications caused verifiable changes in the number of shoots, in the green mass, or in the height of test plants. The increased nutrient supply of the soil through the treatments was not reflected in the nutrient content of the plants during the first year.

**wood ash load / small plot experiment / nutrient supply / heavy metal content**

**Kivonat** – Fahamu hatása a talaj kémiai jellemzőire és a termés vitalitására egy kisparcellás kísérletben. A fatüzelésű erőművekben és fűtőművekben az égetés melléktermékeként nagy mennyiségben keletkezik fahamu. 2009-ben indított kísérletsorozatunkban vizsgáltuk a hamu összetételét, a hamuval kevert talaj tápanyag-szolgáltató képességét, alkotórészeinek felvehetőségét. 2010 tavaszán szabadföldi kisparcellás kísérletet állítottunk be 0, 1, 2,5, 5, 10 t fahamu/ha-nak megfelelő dózissal fehér mustár és angol perje tesztnövényekkel, gyengén savanyú, agyagos vályogtalajon. A talaj vizes szuszpenzióban mért pH-értéke statisztikailag igazolhatóan növekedett a kezelések hatására. A hamu kijuttatásakor jelentősen emelkedett a talaj P<sub>2</sub>O<sub>5</sub>- és a K<sub>2</sub>O-tartalma. A kezelések növelték a termőtalaj magnézium- és kén tartalmát, valamint a mikroelemek közül a cink mennyiségét. A tesztnövények kelésszámában, zöldtömegében és magasságában egyik fahamu dózis sem okozott igazolható változást. A kezelések hatására a talajban megnövekedő tápelem kínálatot a növények tápanyagtartalma az első évben nem mutatta.

**fahamu-kijuttatás / kisparcellás kísérlet / tápelem-ellátottság / nehézfém-tartalom**

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## 1 INTRODUCTION

Interest in using biomass for energy has grown recently because of the reduced availability of fossil fuels. In recent years, a partial or complete switch to biomass heating solutions has happened in Hungary, both in regular households and in many coal-fired power plants such as the ones in Pécs and Ajka. (Tóth et al. 2011). For fuel, these power plants use firewood or sawdust and scrap wood from forestry and the wood industry.

The by-product of wood combustion is ash. The amount of combusted firewood is growing every year, which is consequently reflected in the increasing amount of wood ash. The annual estimated mass of wood ash in Hungary is 30 thousand tonnes; its volume is 50 thousand m<sup>3</sup> (Tóth et al. 2011). Currently, wood ash is treated as waste and is most often used for filling former mine-shafts or it is simply landfilled. The growing expenses associated with landfilling and the reluctance to open new waste landfill sites have brought about increasing interest in alternative methods of disposal. Among these is the use of wood ash for the purposes of soil amelioration and soil refill in agriculture, horticulture, and forestry (Demeyer et al. 2001; Steenari – Lindqvist 1997; Zimmermann et al. 2010).

A variety of factors can influence the composition of wood ash. The quality of the combusted wood – whether it was scrap wood or of higher quality – can play a role as can the combustion technology used. (Campbell 1990). Which parts of trees are combusted can also influence the ash significantly; the nutrient content of roots and branches is much higher than the nutrient content of logs, for example. The difference between species can have significant effects, in many cases even within the same genus. In certain species the chemical compound of ash is normally influenced by the soil properties and the climate (Werkelin et al. 2005). The differences in the chemical composition of identical wood species from different stocks with acidic or calcareous soil may be bigger than in different wood species from stocks with identical soil qualities (Ulrich 1990). In addition to nutrients, ash might contain other elements such as heavy metals. Consequently, the data in literature concerning the chemical composition of wood ash is considerably diverse (Demeyer et al. 2001).

The application of wood ash can cause rapid changes in the chemical properties of the soil, especially in the top layer. The treatment can enhance the pH of the top layer by 0.3–2.4 units in a load of 1–7 t/ha (Mandre et al. 2006; Ozolincius et al. 2007; Perucci et al. 2008). In the deeper layers, the neutralizing effects are far more moderate, or they do not appear at all (Arvidsson et al. 2002). Oxides, hydroxides, hydrogen carbonates and carbonates are responsible for the rapid change in the pH level. The rate of hydroxide/hydrogen carbonates/carbonates ions can vary to a great extent, and consequently the alkalisation effects of different ashes can also be diverse (Etiegni – Campbell 1991).

Ash contains all the components of wood in a concentrated form, except for carbon, hydrogen and nitrogen which evaporate during the firing of wood. The mean concentration of major nutrients in wood ash is as follows: 0.06% N, 0.42% P, 18% Ca, 0.97% Mg. Also, 2.27% of K for wood-fired boiler ashes (Vance 1996) and 1.57% P, 18.5% Ca, 2.86% Mg and 3.52% K for bark ashes have been reported (Someswhar 1996). During its application in soil, wood ash behaves like fertilizers with a low nitrogen content (NPK: 1:10:50) (Park et al. 2004). Its K-content perfectly dissolves in water, which explains its sensitivity to leaching (Demeyer et al. 2001; Odlare – Pell 2005). The greatest part of potassium gets bound in the soil both physically and chemically, and only 20 to 40% of potassium is available (Naylor – Schmidt 1989) when the potassium content in the soil is low. The calcium-oxide content of soil turns into calcium-hydroxide in the presence of water. Calcium-hydroxide reacts with the carbon-dioxide in the air, which results in the formation of calcium-carbonate. As a result, wood-ash enhances the lime content of the soil (Steenari et al. 1999).

The microelement concentration of wood ash is diverse. According to prior research, the heavy metal content of wood ash is typically low (Someshwar 1996). The increased pH-level in the soil will cause a further decrease in the mobilization of certain heavy metals. Despite the low concentration and the differences in mobility, heavy metals (Cd, Cr, Cu, Mn, Ni, Pb, Zn) may cause changes in the soil, vegetation and consequently in the composition of ground water (Ozolincius et al. 2007). Repeated spillage of ash may cause an accumulation of heavy metals. By lowering the soil pH, the mobilization of heavy metals will increase, which is a potential threat for the environment. However, wood ash has just the opposite effect on soil because it reduces the mobility of heavy metals (Vance 1996; Campbell 1990). According to the latest surveys, the toxic trace element load of the ash can fluctuate, so its Cd-content can reach a concentration of 20 mg/kg (Omil et al. 2007), like when the wood used comes from the soil of a spoil bank with high cadmium content and high acidity, for instance. In cases other than that, the accumulation of cadmium to such an extent is rare. Nevertheless, ash still needs to be used carefully in order to prevent heavy metal contamination or its incidental negative effects on terrestrial and aquatic ecosystems (Narodoslawsky – Obernberger 1996).

Application of ash on soil can be performed in several ways, and each way impacts the environment differently. Raw ash (as a strong alkaline) is difficult to manage as its particulates tend to dissolve easily. The particulate size of stabilized ash is larger, and as its pH-value is about 10.5, it is easier to manage, but at the same time – as it has a tendency to carbonation – the dissolution of nutrients is slow (Steenari – Lindqvist 1997). The pH of ash granules is lower, about 9, and its elements dissolve slowly due to the bigger particulate size (Callesen et al. 2007). Extruded, pelleted wood ash is also easy to manage thanks to its size, however, the dissolution of the elements is limited (Emilsson 2006; Csiha et al. 2007).

Wood ash is regarded as non-hazardous waste of non-agricultural origin, thus its use for agricultural purposes is an activity subject to prior authorisation. According to Act CXXIX of 2007, an application for an authorisation certificate must be submitted to the Directorate for Plant and Soil Protection of the relevant government office of a given county.

To examine the effect of wood ash on soil and vegetation we conducted a small plot experiment launched in May 2010. Within this framework we have been examining the composition of wood ash, the mobilization of its constituents, and its nutrient-supplying capacity. We have been studying the changes in the chemical properties of the soil as a result of the treatment, as well as the effect of the ash on the number of shoots, the growth and the element content in the test plants.

## 2 MATERIALS AND METHODS

The wood ash used in the experiment was collected in March 2010 from the ADA Hungária Bútorgyár Kft [ADA Hungária Furniture Factory Ltd.] in Körmen where scrap wood is disposed of through burning. Before the experiment was launched, the wood ash was stored in sealed plastic bags. We chose a slightly acidic soil for the experiment due to the alkalizing effect of wood ash. The agricultural plot selected for the experiment had an area of 100 m<sup>2</sup> and was located in Tanakajd, in Vas County. The designated land was divided into forty plots (1 m x 1 m / plot) to allow for the investigation of the effects of different loads of wood ash on ryegrass (*Lolium perenne*) and white mustard (*Sinapis alba*). The treatment groups were composed of a treatment-free control group and groups based on 1, 2.5, 5 and 10 tonnes of wood ash/ha with four replications for each treatment for both plant species (i.e. four replications per treatment per plant species). The treatment plots were arranged according to the laws of a randomized block design (Sváb 1981).

Of the two test plants, ryegrass responds intensely to changes in nutrient quantity, with rapid initial growth and strong germination. Ryegrass is commonly used for the testing of fertilizing effects for this reason. Seeds of white mustard show a rapid and nearly 100 per cent germination, which is why it can be used for testing anti-sprouting and germination inhibiting effects.

The preparation of each plot was followed by the application of the required amount of ash load, with the ash evenly mixed into the upper 10 cm layer of the soil. The test plants were seeded in May 2010. White mustard seeds were seeded on the 1 m<sup>2</sup> small plots, with 200 germinative seeds/m<sup>2</sup> and 25 cm spacing between each line, at a depth of 2–3 cm. Ryegrass seeds were dispersed on the 1 m<sup>2</sup> small plots, (40 grams of germinative seeds/m<sup>2</sup>) onto the surface of the soil, and subsequently worked into the soil to a depth of 1–2 cm. Rainy weather assured the water needs of the plants and no additional watering was required.

On the seventh day after planting, the number of sprouted white mustard plants was recorded for each plot. The white mustard and ryegrass plants were harvested in August (95 days after planting), at which point the number of plants per plot was recorded. In addition, the average height of the white mustard plants was determined based on the height of five randomly chosen plants. In the case of the ryegrass, we measured the average height of the plants. After the plants were cut off at the base, the above-ground biomass was measured per each plot, and subsequent to the harvest, we took samples of the white mustard test plants. Average samples from the replications were created for laboratory testing per treatment. Soil sampling also took place during the harvest. The samples were taken from depth of 0–10 cm, and consequently one average sample per treatment and replication was created. The laboratory-based analyses on wood ash, soil and plant samples were performed at the accredited soil protection laboratory of the Directorate for Plant and Soil Protection of the Vas County Government Office. The „total” element content of the wood ash, of the soil and of the white mustard test plants was determined following cc. HNO<sub>3</sub> + cc. H<sub>2</sub>O<sub>2</sub> destruction, using Inductively Coupled Plasma. The soluble nutrient content of the soil was detected based on the method of Hungarian Standard MSZ 20135:1999.

Determination of easily soluble nitrogen fractions was made by means of extraction solvent 0.01M CaCl<sub>2</sub>, while determination of soluble potassium and phosphorus fractions was performed by ammonium lactate solution (AL). For the extraction of the copper, manganese and zinc content ethylenediaminetetraacetic acid was used.

The statistical analysis was performed using software StatSoft, Inc. (2012), STATISTICA (data analysis software system), version 11. To explore the differences in the treatment effects, a one-way analysis of variance (ANOVA) was performed.

### 3 RESULTS

The ash mixed during the experiment is strongly alkaline, with a pH of 13.0. Its phosphorus content is 0.37%, calcium content is 23.29%, magnesium content is 2.68% and potassium content is 5.42% by weight. The iron content of the ash is significant too, 13.46 g/kg (*Table 1*). According to the test results it can be stated that the phosphorus content of the ash is lower than the values given in the literature (Etiégni – Campbell 1991), and the toxic heavy metal load did not exceed the values in the literature, with the exception of chromium.

The mechanical composition of the soil mixed for the experiment had the physical consistency of clayey loam, with slightly acidic pH, with no soda lime content, with nearly 27.3 per cent of clay-content (< 0.002 mm), and over 60 per cent of mud sediments in it (< 0.002 mm). According to its physical properties, the hydraulic conductivity of the soil is

average, while its water holding capacity is good and its air metabolism is satisfactory. Its nutrient content, based on its humus concentration, is good too.

Table 1. Characteristics and content of nutrients and heavy metals of wood ash.

Tested parameters	Units	Results	Results given in the literature (Etiégni – Campbell 1991)
pH (H <sub>2</sub> O)		13.0	13.1 – 13.3
dry matter	% w/w	99.9	–
volume/mass	kg/dm <sup>3</sup>	0.926	–
P	g/kg d.m.*	3.7	14.0
K	g/kg d.m.	54	41.3
Ca	g/kg d.m.	233	317
Mg	g/kg d.m.	27	22.5
Na	g/kg d.m.	5.8	3.4
Al	g/kg d.m.	18	23.65
Fe	mg/kg d.m.	13.5	19.5
Cd	mg/kg d.m.	6.5	21
Cr	mg/kg d.m.	182	86
Cu	mg/kg d.m.	110	145
Hg	mg/kg d.m.	< 0.5	–
Ni	mg/kg d.m.	58.9	–
Pb	mg/kg d.m.	85.0	130
Zn	mg/kg d.m.	496	700

Note: d.m.\* - dry matter

The soil pH is favourable for the exposure to nutrients and thus for the nutrient uptake of plants, as this is a suitable pH-range for active biological life, with no extreme chemical conditions in the soil that would significantly lower the mineralization of any type of organic substances, macro- or micro-nutrients.

Due to the sum of the exchangeable cations in the soil, the overall cation exchange capacity of the soil (T-value) is good, 23.2 meq/100 g of soil. The good adsorption properties are formed as a result of the clay content and the humus content of the soil (Table 2). It is the colloids of the soil that are responsible for the water and nutrient absorption, as the amount of water stored in soil and available for uptake depends on the quantity and quality of colloids.

Table 2. Major characteristics of the soil used in the green-house experiments

Parameters	Results
pH (H <sub>2</sub> O)	6.6
pH (KCl)	5.6
Plasticity index according to Arany (K <sub>A</sub> )	43
Percentage of humus	2.6%
Percentage of carbonated lime	< 0.1%
Cation exchange capacity (T-value) (meq/100 g of soil)	23.2

In the case of the white mustard test plants, the treatments didn't cause a significant difference either in the number of all the emerged plants recorded on the seventh day, or in the number of plants per plot at the end of the trial period (Table 3).

Table 3. Effect of wood ash treatment on test plants.

Wood ash treatment (t/ha of soil)					Significant difference (5%)	Average
0	1	2.5	5	10		
White mustard, number of shoots (pc)						
182	181	184	180	179	10	181
White mustard, number of stocks (pc)						
180	179	186	183	183	7	182
White mustard, height (cm)						
120	121	122	120	121	8	121
White mustard, green mass (g)						
2543	2705	2430	2713	2663	919	2611
Ryegrass, height (cm)						
27	27	28	28	27	1.94	28
Ryegrass, green mass (g)						
802	830	821	848	864	107	833

According to the weight measurement of the different plots, the ash treatments provided a higher weight of organic material in both plant species on all plots, with the exception of one. There was no difference between the average growth in height within a species: white mustard reached an average height of 120–122 cm in each plot, while ryegrass reached 27–28 cm. The positive effects of wood ash treatment were manifested in the organic material content of the above-ground parts. When the white mustard was compared to the control group, the treatment of 1 t/ha caused 6.3% increase in weight, the treatment of 5 t/ha caused 6.7% increase, while the 10 t/ha treatment caused a growth of 4.7%. In the 2.5 t/ha treatment the green mass was lower than that of the control. A very similar tendency occurred with the ryegrass, too: when compared to the control, the 1 t/ha treatment resulted in a growth of 3.5%, the 2.5 t/ha treatment in a growth of 2.4%, the 5 t/ha treatment in a growth of 5.7%, while the 10 t/ha treatment caused an 8% growth in the above-ground organic matter. No significant change in the height and green mass of the test plants could be detected in any of the cases.

The ash treatment enhanced the soil pH measured in aqueous suspension. The pH value of the average soil-sample taken from an identically treated soil rose for both white mustard and ryegrass; after the application of a 10 t/ha dose, the pH level rose to 7.6, compared to the pH 6.4 level of the control. The values measured in the 1 mole KCl-suspension followed the pH-changes of water. No statistically verifiable differences in the pH-changes of the soil were uncovered for the two test plants: the pH values measured both in aqueous suspension and in suspension of potassium chloride seem to form a saturation curve (*Figure 1*).

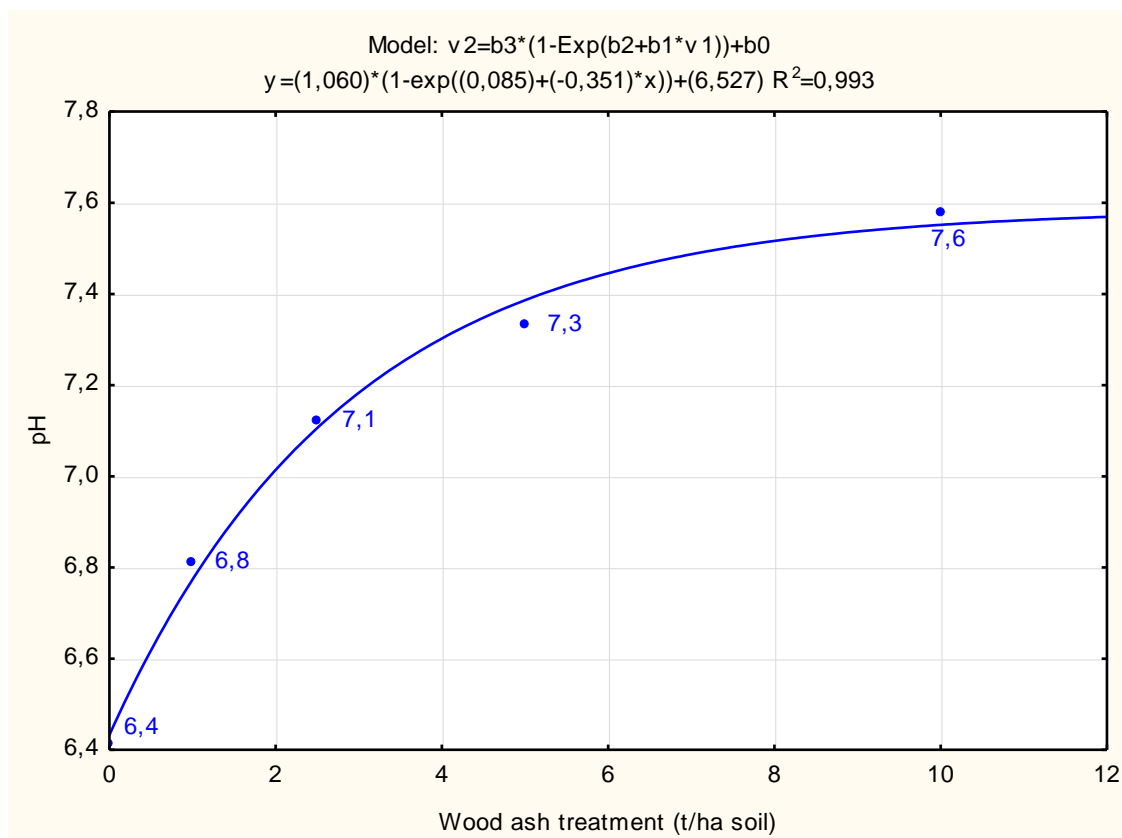


Figure 1. The pH-change in the soil of white mustard caused by the wood ash treatment

The ash treatment increased the carbonated lime content of the soil as well as the amount of potassium, phosphorus, magnesium and zinc, among the other nutrients. The carbonated lime content of the lime-defective soil rose to 0.8 mass% during the treatment of 10 t/ha. The treatment did not affect the amount of nitrogen, as this substance essentially evaporates during wood combustion.

The potassium content of the wood ash used in the experiment is equivalent to 653 kg of  $K_2O$  active substance per hectare. The AL-soluble  $K_2O$  content compared to the value of 301 mg/kg of the control increased up to 792 mg/kg, in the case of the highest dose in the 0–10 cm layer, so the potassium content of the originally well-supplied soil showed a further increase (limit value: 301–500 mg/kg).

The amount of phosphorus applied together with wood ash is equivalent to 85 kg  $P_2O_5$  of active substance per hectare. The treatment caused a change in the  $P_2O_5$  content from the value of 760 mg/kg to 1144 mg/kg in the 0–10 cm layer, thus the amount of phosphorus continued to increase in the especially well-supplied soil (limit value: 161–360).

The dosage of 10 t/ha enhanced the sulphate content of the top layer; its value changed to 35.6 mg/kg from the control's 11.9 value.

As a result of the ash treatment, the amount of magnesium in the soil with optimum magnesium supply (limit value: 100 <) increased to 398 mg/kg, from the initial 141 mg/kg (Buzás 1983). The EDTA-soluble zinc content of the control soil changed from the value of 5.3 mg/kg to 10.5 mg/kg after the wood ash treatment. In the case of copper and manganese no increase in concentration could be detected, while the amount of AL-soluble sodium content significantly rose after the wood ash treatment, reaching the value of 127 mg/kg, compared to the 36.0 mg/kg of the control. As to heavy metals, an increase in the amount of cadmium could be detected, with a change from 0.3 mg/kg to 0.5 mg/kg in the soil (Table 4).

Table 4. Effects of the wood ash treatment on the lime-, nutrient- and heavy metal-content in average soil samples

Parameters	Units	Doses of ash (t/ha)				
		0	1	2.5	5	10
CaCO <sub>3</sub>	% w/w	0.0	0.0	0.3	0.3	0.8
NO <sub>3</sub> <sup>-</sup> +NO <sub>2</sub> <sup>-</sup> -N	mg/kg	8.2	8.4	11.6	11.6	11.6
P <sub>2</sub> O <sub>5</sub>	mg/kg	760	888	906	999	1144
K <sub>2</sub> O	mg/kg	301	383	468	579	792
Na	mg/kg	36.0	56.0	71.0	95.0	127
Mg	mg/kg	141	148	237	305	398
SO <sub>4</sub> <sup>2-</sup> -S	mg/kg	11.9	13.4	19.7	22.6	35.6
Ca	mg/kg d.m.	4648	5115	6187	6696	7435
Cd	mg/kg d.m.	0.3	0.4	0.4	0.5	0.5
Cr	mg/kg d.m.	37.2	38.5	38.2	38.2	40.1
Cu	mg/kg d.m.	21.5	22.5	22.8	23.3	23.3
Hg	mg/kg d.m.	< 1.5	< 1.5	< 1.5	< 1.5	< 1.5
Ni	mg/kg d.m.	25.7	27.1	26.5	25.8	27.3
Pb	mg/kg d.m.	28.9	30.4	30.7	30.8	31.8
Zn	mg/kg d.m.	82.7	87.5	89.0	90.6	93.3

The particulate content of white mustard test plants did not change significantly, which proves that the amount of nutrients in the untreated soil was initially suitable for the development of the plants. The amount of heavy metals in test plants did not change following the treatment (Table 5).

Table 5. Effects of the wood ash treatment on the average element content of the white mustard test plants.

Parameters	Units	Doses of ash (t/ha)				
		0	1	2.5	5	10
N	% w/w d.m.	2.6	1.9	2.3	1.6	1.5
P	% w/w d.m.	0.3	0.2	0.3	0.2	0.2
K	% w/w d.m.	2.1	2.1	2.0	1.9	1.6
S	% w/w d.m.	0.9	0.8	1.0	0.6	0.5
Cd	mg/kg d.m.	0.2	0.4	0.3	0.2	0.2
Cr	mg/kg d.m.	< 1.5	< 1.5	< 1.5	< 1.5	< 1.5
Cu	mg/kg d.m.	3.4	2.7	2.7	2.0	2.1
Hg	mg/kg d.m.	< 0.5	< 0.5	< 0.5	< 0.5	< 0.5
Ni	mg/kg d.m.	< 7.5	< 7.5	< 7.5	< 7.5	< 7.5
Pb	mg/kg d.m.	< 3.0	< 3.0	< 3.0	< 3.0	< 3.0
Zn	mg/kg d.m.	31.8	36.1	34.1	24.9	21.0

#### 4 EVALUATION OF RESULTS AND CONCLUSION

During our research we examined the properties and chemical composition of wood ash. We studied its effect on the chemical condition and nutrient supplying properties of soil. Using white mustard and perennial ryegrass as test plants, we examined the effect of wood ash on the growth of test plants.

The laboratory tests revealed that wood ash has fairly high potassium, phosphorus and magnesium content, and its pH is highly alkaline. With one exception, the toxic heavy metal content of wood ash did not exceed the average values cited in literature (Etiégni – Campbell 1991). Wood ash has apparent effects on the soil. In the case of both test plants the dose of 10 t/ha raised the soil pH by approximately 1 unit. Regarding the nutrients, the nitrogen content of the soil did not change; however its potassium, phosphorus, sulphur, magnesium, calcium and zinc content increased following the ash treatment, as well as the lime content of the lime deficient soil. Among heavy metals, a slight increase in the cadmium amount could be detected, while the raised sodium content of the soil calls for further research.

The raised Na-content is unfavourable for cultivated plants, and its value exceeding 60 mg/kg is a sign of unfavourable salinization.

The treatment did not cause any statistically verifiable differences in the number of shoots and stocks or in the height and in the green mass of the test plants. Despite the fact that the above-ground organic matter grown on the different plots could not be detected with 95% level of significance, their amount increased by 3–8%, which means the plants were able to produce a greater amount of organic material from the excess amount of nutrients during a representative period of time. The plants experienced no toxic effects even with a maximum amount of ash-dose applied. The nutrient supply enhanced by the wood ash treatment was not reflected in the nutrient content of the plants. The test plants must have had an optimum nutrient supply at the time of the control, which would explain why the treatments did not cause significant differences. No significant change could be detected in the heavy metal content of the plants as a result of the wood-ash treatment.

Based on the conducted trials it can be stated that wood ash can effectively be used for agricultural purposes of amelioration of acidic soils instead of liming in order to reduce the acidification of soils with characteristics similar to those examined in the experiment. The experiments have proved that wood ash can successfully be used for purposes of nutrient supply. The ash must be applied and evenly mixed with the upper level of the soil before sowing; the recommended wood ash dose is 1 to 5 t/ha. With higher doses the  $P_2O_5$  and  $K_2O$ -values measured in the soil would exceed the optimum values, which would cause the applied nutrients to be washed out of the topsoil in default of the needed colloid-content. In nitrogen-deficient soils, it is recommended to combine the application of wood-ash with N-fertilization.

Moreover, the agricultural application of wood ash has significant economic importance too. Application of ash on arable lands may lower the expenses of landfilling ash as waste. Utilization of wood ash, which is available in huge amounts, may partially replace expensive soil conditioners and fertilizers.

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# The Influence of Wild Boars on the Growth of Forest Trees and Stands: A Case Study of a Wild Boar Game Preserve

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**Abstract** – This research investigated methods for determining and quantifying the impact of wild boars on the increment and growth of forest trees and stands. The influence of wild boars on stand variables was observed in a wild game preserve established in central Slovakia in 2000 practicing intensive wild boar management. Long-term measurements obtained from two long-term research plots of sessile oak trees established in 1969 were used to monitor stand growth. Increments of trees were observed on tree ring cores coming from trees surrounded by differently damaged soil surfaces. Wild boars rooting the soil surface proved to have neither a positive nor negative influence on the mean diameter and height of the forest stands. Analysis of radial increments in 9 trees growing on sites with more intense, deeper, and permanent rooting in the soil profile located near a larger mud bath was also carried out. A more distinctive increment depression was found on one oak near the mud bath and on one beech where deeper soil surface rooting occurred.

**wild boar game preserve / game damage / tree ring analysis / radial increment**

**Kivonat** – A vaddisznó hatása a fák és a faállomány növekedésére – egy vaddisznóskert esettanulmánya. Módszertani vizsgálatokat végeztünk annak megállapítására, hogy a vaddisznók jelenléte milyen hatással van a fák és a faállomány növekedésére. Vizsgálatainkat egy 2000-ben létrehozott vaddisznóskert területén végeztük. Kiindulási alapnak a területen 1969-ben létrehozott két kocsánytalan tölgy hosszútávú kísérleti terület adatait használtuk fel. Olyan mintafákból vettünk évgyűrű mintákat, melyek környezetében a vaddisznók a talajfelszín különböző mértékben kársították. A vaddisznók túrásának nem volt sem pozitív, sem negatív hatása a faállomány növekedésére. Kilenc további fa vizsgálatát is elvégeztük, melyeknél a talajfelszín túrása sokkal intenzívebb volt, illetve közelebb helyezkedtek el egy dagonyához. Itt egy tölgy és egy bükkfán figyeltünk meg jelentősebb növekedés csökkenést.

**vaddisznóskert / vadkár / évgyűrű elemzés / átmérő növekedés**

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## 1 INTRODUCTION

Forest stands are multifunctional communities of which wild game is an integral part. Though game populations and migrations generally occur naturally in forests, there are cases where game is unnaturally introduced into and supported within a restricted area. Game preserves are a good example of this. Economic considerations are at the forefront of most game preserves, but consequent increased game concentration per unit area and the possible negative influence on growth and production of forest stands must also be considered when introducing game into an environment. Published reports on forest stand damage caused by larger rodents (brown hare and wild rabbit) and also primarily by bigger mammals from the genus *Cervidae* (red deer, roe deer and fallow deer) (Ueckermann 1986, Náhlik 1995, Reimoser – Gossow 1996, Find’o 1998); Find’o – Petráš (2007, 2011) summarized current knowledge in this field. Therein, they provided a detailed ecological assessment of game living freely in natural environments including the feeding habits of the game and their negative influences on forest biotopes. These authors defined the extent of negative influence of game on forest stands and they instituted a methodology for damage calculation. Being cognizant of the latest knowledge on volume and value production of forest stands (Petráš – Mecko 1995, Petráš et al. 1996, Halaj – Petráš 1998), these authors also formulated the basis for this methodology. This was supplemented by experimental results of increment loss quantification following the browsing of young forest stand crowns (Find’o 1998) and also browsing and peeling of the bark from tree stems (Jöbstl 1987, Petráš 1996, Náhlik – Walter – Illés 1998, Čermák et al. 2009).

Although direct damage to crowns and stems by forest game has been relatively well studied, indirect methods of quantifying soil surface rooting and scarification of considerable portions of the soil profile were previously lacking. Wild boars are typically culprits of scarification; (Náhlik – Sándor 2003, 2005, 2012, Jánoska – Varju 2008). Find’o – Petráš (2007, 2011) quote this game on their list of animal species decreasing natural stand regeneration by excessive mast feeding. Within the concept of soil surface rooting, this game is more likely to be useful rather than harmful. These authors contend that better seed germination and better forest regeneration result in more favourable soil surface humidification. Although the intensity of soil surface rooting is not taken into consideration with this statement, this intensity certainly differs between wild boars living freely in the wild and those living in game enclosures.

The aim of this work is to investigate methods which will determine and quantify the impact of wild boars on the increment and growth of forest trees and stands. Specifically, in a wild boar game preserve with intense management. The focus is on observation of the wild boars’ influence on tree increments of more tree species and with respect to soil surface damage intensity in immediate proximity to those trees.

## 2 MATERIAL AND METHODS

The material for this study emanates from two sources. The first source consists of repeated measurements on two permanent sample plots established in 1969, and currently located within the game preserve. The second source is composed of tree ring cores taken from the most attacked sites of trees growing in the game preserve.

### 2.1 The wild boar preserve

The wild boar preserve is located in the central part of Slovakia, on a forest enterprise area owned by the Technical University in Zvolen. Its total area is 254 ha; 243 ha of which are

afforested. It was established in 1966–1971 to keep fallow deer, but in 2000 it was reworked to house wild boars. The population density of wild boars was increased gradually. In the first year there were only 6 boars, but after 5 years, their population increased to 215. The population is currently around 210–240 individuals, making the density 1 wild boar per 1.1–1.2 ha. As a result, the influence wild boars exert on forest stands was expected to occur after the period of 2000–2005.

The total area of the wild boar preserve is fenced. The game preserve is located at an altitude varying from 320 to 490 m. The growing season lasts 224 days, while the mean annual temperature is 10.9°C. This area receives 700 to 800 mm annual precipitation. SE to SW exposures prevail. The geological base is formed from andesite. There are middle-deep soils with 25–30% proportion of soil skeleton which rises up to the surface in some sites of the game preserve. Soil surface is covered with a thin layer of fallen leaves and in some places with thermophile plants and sparse brush undergrowth. The composition of the trees is 80% *Fageto quercetum* (FQ), 18% *Querceto fagetum* (QF) and 2% *Fagetum pauper* (Fp). sessile oak (*Quercus petraea* Liebl.) and the turkey oak (*Quercus cerris* L.) that are more than 70-years-old prevail throughout the entire game preserve. These tree species combined represent 65% of the tree species in the game preserve.

## 2.2 Long-term research plots

Two long-term research plots (LTP) were established to conduct long-term research on forest production and for construction of domestic yield tables in the 1969 state-wide programme. The sample plots are situated at approximately 400 m altitude. LTP No. 100 is 0.33 ha in area, and LTP 105 has 0.24 ha. Intense rooting of the soil surface caused by wild boars searching for food was determined by the last two measurements on both LTPs in 2005 and 2010. The sessile oak is the main tree species on sample plots, together with rare mixture of hornbeam (*Carpinus betulus* L.) and small-leaved lime (*Tilia cordata* Mill.). All trees on the sample plots are permanently numbered and each was repeatedly measured in 5-year intervals for diameter, sociological tree position, quality and mechanical stem damage. Although tree heights were generally recorded only during the first and last measurements, additional measurements were taken for height curve construction. Models of the height curves were derived from the Michajloff (1943) function:

$$h(d) = 1.3 + a \cdot \exp\left(\frac{-b}{d}\right) \quad (1)$$

where:  $h$  and  $d$  are for tree height and diameter,  
 $a$  and  $b$  indicate regression parameters.

A deviated position in some repeated measurements was discovered during detailed tree height analysis. Some height curves intersected and the distance between neighbouring curves was disproportionately large or small. Therefore, the following equation of Michajloff function modified by Petráš et al. (2012) was used:

$$h(d, t) = 1.3 + p_1 \cdot t^{p_2} \cdot \exp\left(\frac{-p_3 \cdot t^{p_4}}{d}\right) \quad (2)$$

where:  $h$ ,  $d$  – tree height and diameter,  
 $t$  – stand age,  
 $p_1 - p_4$  – regression parameters.

For its derivation, parameters  $a$  and  $b$  in equation (1) were expressed with dependence on stand age ( $t$ ), and an independent regression model was derived from equation (2) for each LTP. The QC.Expert statistical package (Kupka 2003) was applied and the accuracy and precision of derived models was thus evaluated. Basic stand variables were calculated from

the number of trees in each 1 cm diameter class and from the height curve models. There were hectare variables such as the number of trees ( $N$ ), the basal area ( $G$ ) and the stand volume ( $V$ ). Mean stem variables such as the tree mean volume ( $v$ ), diameter ( $d_g$ ) and height ( $h_g$ ) and the same variables of upper stem were derived from those hectare variables. Their development was compared with the development of yield table models (Halaj – Petráš 1998).

### 2.3 Tree ring cores

Changes in tree growth were examined by tree ring series. *Table 1* shows that 9 examined trees comprised 6 sessile oak (*Quercus petraea* Liebl.) trees, 2 silver firs (*Abies alba* Mill.) and 1 European beech (*Fagus sylvatica*). While most of these trees were in a crown level position, with one being dominant and two intermediate, the crowns of two additional trees were cramped by neighbouring crowns. Tree rings were taken from trees growing on sites heavily attacked by wild boars. Seven trees are located at the position where wild boars both fed on supplementary given fodder and secured larger bedding. Another two trees are located at another site, near the bigger mud bath. One core was taken from each tree at breast height, and tree ring widths were measured by a digital positioner, connected on-line to a computer. Tree-ring series were synchronized, dated, and analysed in detail. Comparison was made between tree ring curves and variability of annual radial increments prior to 2000, before implementation of the game preserve for wild boars, and also after that period, when wild boars intensely influenced the plot. This research method was based on the work of several authors; primarily that of Fritts (1976), Schweingruber (1983), Pollanschütz (1986), Petráš et al. (1993) and Petráš – Mecko (2011). These authors confirmed that changes in growth conditions are best indicated by changes in radial increments; where trees form thinner tree rings with lower variability in worsening growth conditions, and vice versa.

*Table 1. Basic data on trees of which tree ring cores were taken*

Number of the tree	Tree species	Tree diameter d (cm)	Tree class	Crown	Notice
1	oak	47	2	free	
2	oak	35	2	suppressed	
3	fir	35	3	free	
4	oak	33	2	suppressed	
5	fir	72	1	free	
6	oak	43	2	free	
7	beech	39	3	free	
8	oak	46	2	free	at the mud bath
9	oak	41	2	free	at the mud bath

## 3 RESULTS AND DISCUSSION

### 3.1 Changes in oak stands growth

Measured tree heights were individually equalized for each LTP, dependent on their stand diameter and height, using nonlinear least squares in compliance with the model (2). One height measurement proved incorrect for each LTP. The following systematic methods were employed. (1) On LTP 100, tree heights were lowered on the fourth measurement by approximately 1.0–1.5 m, and (2) on LTP 105, higher heights were measured on the first occasion. In both cases, these measurements were excluded from the regression equation. The

equation parameters (Table 2) are very favourable. Determination coefficients  $R^2$  are relatively high and these reveal that the model explains 93–95% of observed height variability. Standard errors of the equation are relatively low  $s_h$  (1.13–1.32 m) for the same reason. The model height curves (Figure 1) have a typical fan shape, where tree heights increase non-linearly with broader diameter and higher stand age.

*Table 2. Parameters ( $p_1 - p_2$ ) and statistical characteristics of regression equation of height curves models*

Plot	$p_1$	$p_2$	$p_3$	$p_4$	n	R	$R^2$	$s_h$ (m)
LTP 100	2.1186	0.6390	0.5211	0.6732	905	0.9632	0.928	1.32
LTP 105	0.9495	0.7999	0.3479	0.7550	656	0.9761	0.953	1.13

Note: LTP – long-term research plots; n – number of measured heights; R – correlation coefficient;  $R^2$  – coefficient of determination;  $s_h$  – standard errors.

*Table 3. Development of basic stand variables from repeated measurements on LTP*

Year of measurement	Age	Values per 1 ha			Mean stem			Upper stem (10%)		
		V ( $m^3$ )	N (pcs)	G ( $m^2$ )	v ( $m^3$ )	d (cm)	h (m)	v ( $m^3$ )	d (cm)	h (m)
LTP 100										
1969	56	194	1318	27.26	0.147	16.39	18.50	0.385	24.18	21.37
1974	61	211	1179	27.67	0.179	17.50	19.53	0.488	26.33	22.68
1979	66	231	923	27.60	0.250	19.74	21.10	0.660	29.49	24.20
1984	71	268	801	29.70	0.335	21.97	22.55	0.837	32.23	25.58
1995	82	345	687	34.07	0.503	25.41	25.10	1.194	36.62	28.15
2005	93	435	638	39.36	0.682	28.31	27.29	1.619	40.88	30.60
2010	98	474	605	41.33	0.784	29.79	28.34	1.833	42.76	31.67
LTP 105										
1969	48	102	3881	26.30	0.026	9.70	12.10	0.109	15.90	15.28
1974	53	122	3522	27.29	0.035	10.50	13.01	0.153	17.77	16.66
1979	58	145	2312	26.48	0.063	12.60	14.77	0.237	20.75	18.35
1984	63	177	1723	27.64	0.102	14.75	16.53	0.341	23.61	19.95
1995	73	255	1314	32.96	0.194	18.29	19.37	0.545	27.77	22.63
1999	78	286	1032	34.02	0.277	20.76	21.10	0.648	29.44	23.85
2005	84	340	973	37.68	0.350	22.47	22.48	0.805	31.74	25.38
2010	89	379	898	39.71	0.422	23.98	23.69	0.934	33.39	26.57

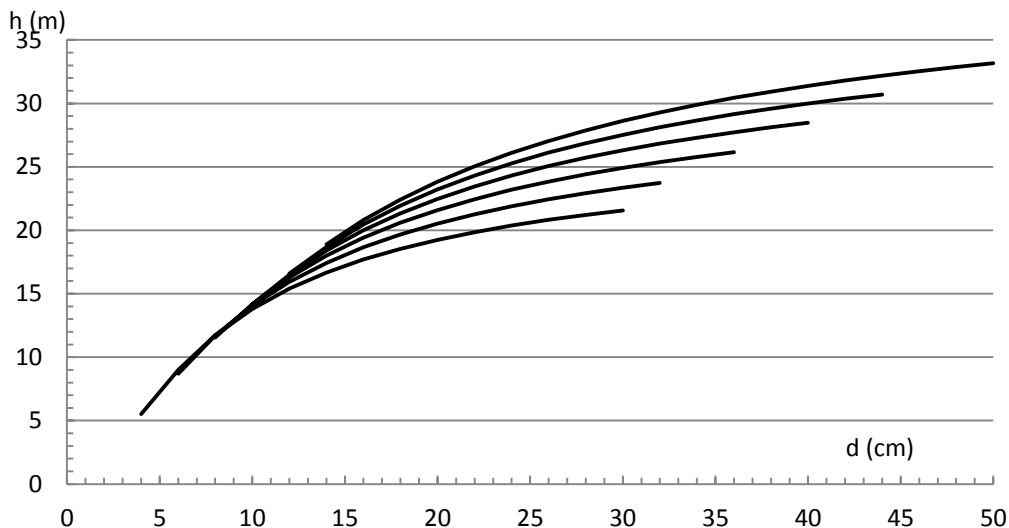


Figure 1. Height curves model for trees 50–100 years old on the LTP 100

Oak stand growth was evaluated by the continuous development of their growth variables (Table 3). Although hectare variables such as the number of trees ( $N$ ), the basal area ( $G$ ) and the stand volume ( $V$ ) result from growth processes, these also significantly depend on stand density. More remarkable negative influence of wild boars on the density of mature stands cannot be expected at a wild boar density of 0.83–0.94 per 1 ha of the game preserve, only on the growth conditions of individual trees, so their influence was assessed via changes in tree mean diameter ( $d_g$  in Figure 2) and mean height ( $h_g$  in Figure 3). Development of these characteristics on both LTPs was compared with model development yield tables (Halaj – Petráš 1998), and thus LTP 100 was assigned to site class 26 and LTP 105 to class 22. However, the actual mean diameters, and especially the mean heights, displayed a more abrupt course on these LTP than that in the model yield tables. Following the establishment of the game preserve in the year 2000, increased significant influence of wild boars on the stands growth was expected. At that time, LTP 100 was approximately 90-years-old and LTP 105 was 80. However, comparison of the actual and the model curves failed to definitively prove that wild boars negatively or positively influenced the growth process of oak stands through trampling effects on the soil surface.

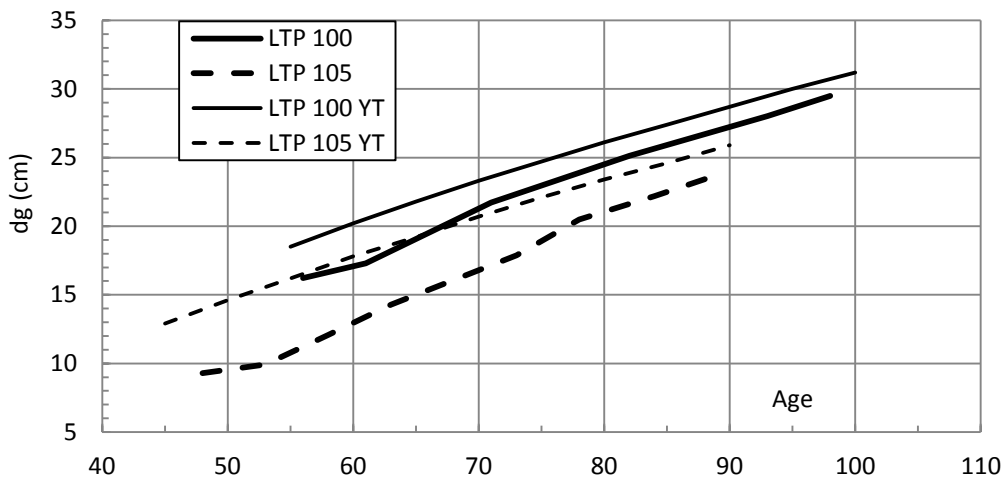


Figure 2. Comparison of mean diameter  $d_g$  development (bold) on LTP 100 and 105 with the model one (thin) by yield tables (Halaj – Petráš 1998)

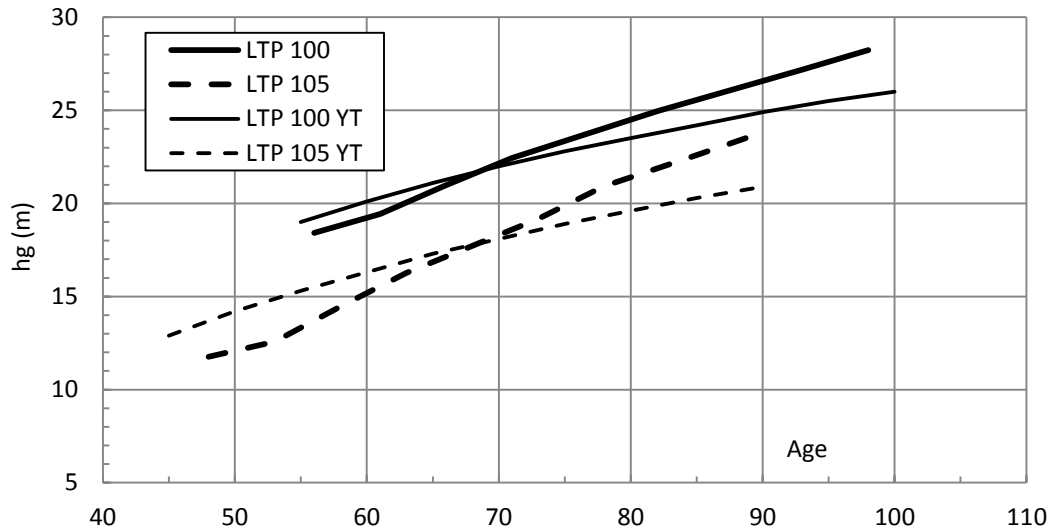


Figure 3. Comparison of mean height development  $hg$  on LTP 100 and 105 with the model one by yield tables (Halaj – Petráš 1998)

### 3.2 Changes in individual trees growth

Changes in tree growth were examined by tree ring series. Even when formation of tree rings is unambiguous in our climatic conditions, there are individual cases where tree rings vary. Two rings may form in some years, while no rings form or are scarcely recognizable in other years. Therefore, tree ring series were synchronized and dated to discover and eliminate these particularities. The percentage of parallelism in increment trends (Jačka 1989, Petráš et al. 1993) was calculated for all pairs of tree rings series apparent over the last 50 years. Results of this calculation delivered the wide range of 39–83%. Only 4 pairs of tree ring series exhibited less than 50% parallelism. Here, oaks with free crowns recorded the highest values up to 68–83%, while suppressed fir and beech ring series registered lower parallelism. This lower parallelism percentage confirmed that the tree ring series are correct.

In-depth analysis of tree ring series proceeded from our knowledge and experience. When growth conditions of examined trees change, the tree responds with changed increments. Thus, as conditions improve, both tree increments and variability increase, while their increments and variability are smaller in worsening conditions (Pollanschütz 1986, Petráš – Mecko 2011, Bošela et al. 2013, 2014). The timing of changes in growth conditions is also important. A sudden change in conditions induces radically larger or smaller increments in the increment curve. This knowledge was employed to investigate the actual influence of wild boars on radial increments in all 9 trees. The trees were divided into three groups for precise illustration. The first group comprised three oaks with long-term balanced increments and natural variability in the range of 1 to 4 mm (Figure 4). Despite considerable variability in increments, it was not possible to confirm marked depressions or increased increments in these trees during the 1965–2012 period. The second group included oak trees which experienced radial increments (Figure 5). The curves for trees number 2 and 4 indicate that more distinctive increment depressions occurred after 1980 or 1990, and they maintained this similar condition after the year 2000. From reasons stated above, we can conclude that neither depression was due to negative influences of wild boar activity near the trees, but rather was due to other causes. The most likely cause lies in the worsened position of the tree with respect to crown space. However, an exception to this is oak tree number 8 which grows on the lower side of the mud bath which has an approximate diameter of 4–5 m. This oak tree maintained its approximately balanced increments until 2005, and then a marked depression

occurred with increments decreasing from 1.3 mm to 0.4 mm. This was the minimum value recorded since 1930 in the entire study period. Since the crown of this tree is in the crown-level position, we can assume that this depression occurred because of the negative influence of the mud bath which is permanently filled with water and deep mud. In connection with this assumption, we stress the fact that oak tree number 9 (Figure 4), which has no increment depression, is located almost at the same distance from the mud bath, but on the upper aspect and thus negates a permanently wet root system. The third group consists of two firs and one beech tree (Figure 6). Fir tree number 5 is dominant; being the thickest of all trees. Since its increment variability is relatively broad, it is difficult to presume that the reason for its sudden increment depression over the last 3 years is due to wild boar activity. However, the situation with the beech tree is entirely different. This beech tree suffered distinct depression in 2005–2012, with following bigger increments between 1980 and 2004. Hence, the possibility certainly exists that negative influences emanated from wild boar activity.

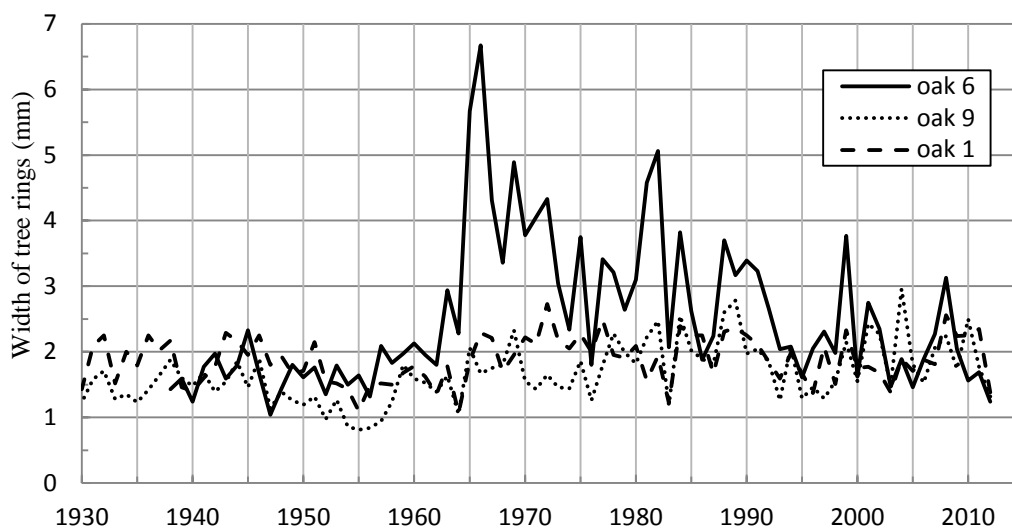


Figure 4. Radial increments of the oaks no. 1, 6 and 9 without any significant changes in the observed period

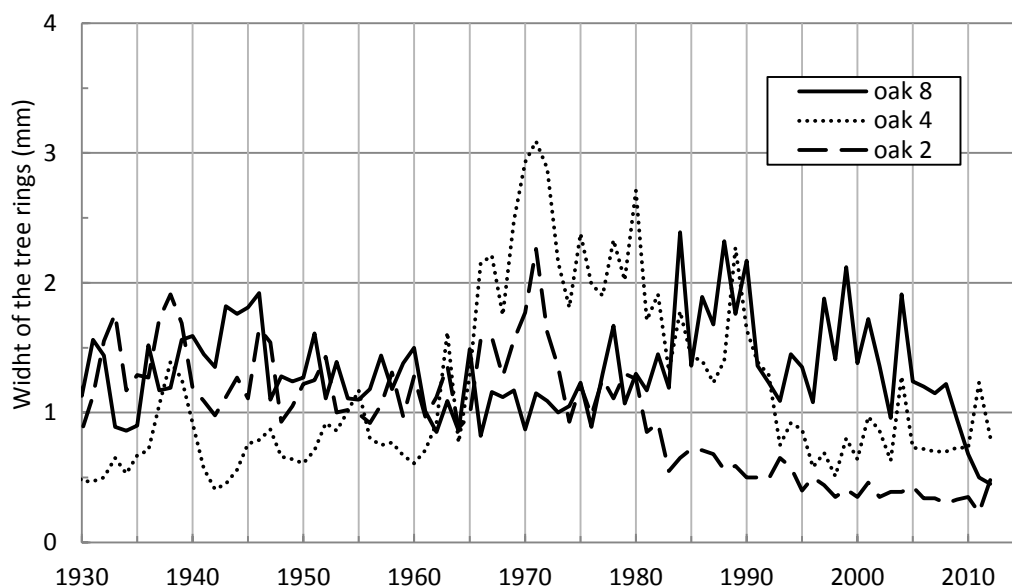
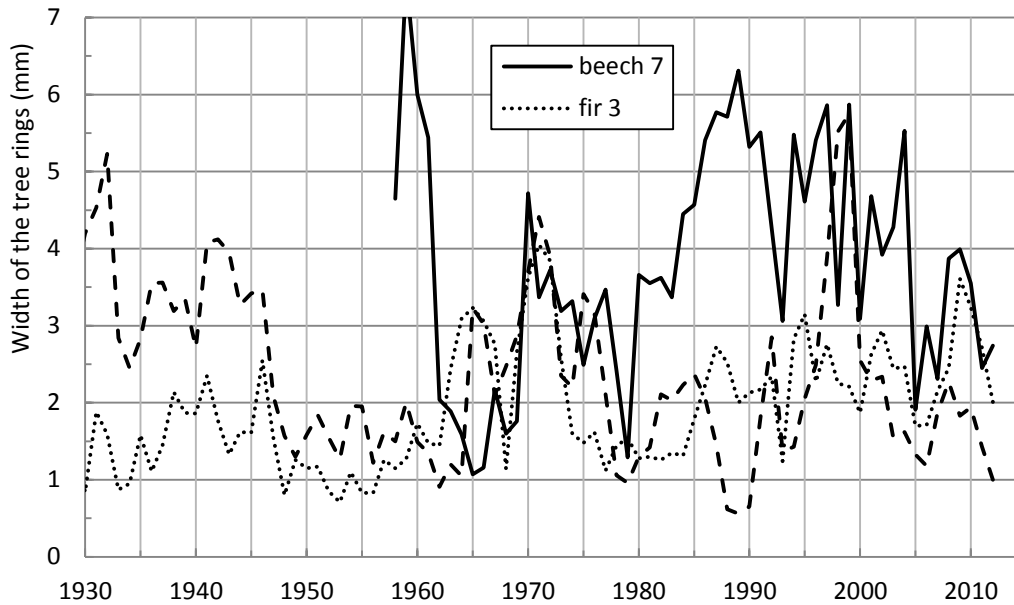


Figure 5. Radial increments of the oaks no. 2, 4 and 8 with more significant changes in the observed period



*Figure 6. Radial increments of firs no. 3 and 5 and the beech with no. 7 with more significant changes in the observed period*

On the basis of obtained results, we can state that soil surface rooting by wild boars cannot definitely be considered responsible for positive or negative influences. This certainly depends on scarification extent, depth, and intensity. In our two LTPs, there was whole-scale rooting, but only on the surface, with accompanying disruption of soil litter. The soil profile was not disturbed to a deep level in any area. In addition, although the approximate ten-year observation period of this study is relatively short, our intention is to continue monitoring these LTPs while the game preserve is maintained. This study has confirmed that analysis of radial increments on tree ring series is an appropriate and beneficial methodology designed for particular trees and their biotopes. Future research will enable increased precision in the study of these trees and their biotopes, and long-term monitoring of increased number of trees and tree species is pertinent.

#### 4 CONCLUSION

The objective of this work was to investigate methods which will determine and quantify the impact of wild boars on the increment and growth of forest trees and stands. Their influence on hectare and mean stand variables was observed in a game preserve with intensive wild boar management. Long-term measurements obtained from two long-term research plots of sessile oak trees served as a basic material for this observation. Tree increments of more tree species were observed on tree ring cores coming from differently damaged soil surface surrounding those trees.

Results showed that shallow rooting of the soil surface caused by wild boars has no influence on oak stand growth. The development of mean heights and diameters on both the permanent sample plots observed over 41 years is continuous, without significant changes after the year 2000 when the game preserve for wild boars was established in this locality.

A significant influence of wild boars was proven only on radial increments in some individual trees. Deeper rooting of the soil surface had no effect on eight of the nine examined trees; these were six oak and two fir trees. However, the remaining beech tree proved an exception, exhibiting increment depression over the last seven years. Mud baths installed for

the wild boars in this game reserve had different effects on the surrounding trees. Radial increments on an oak growing on the lower side of the larger mud bath which was permanently filled with water and deeper mud decreased quite significantly after 2005.

Our results indicated that it is possible to expect negative influence on forest tree increment resulting from wild boars activity, and that this is only a question of extent and intensity. We can state that it cannot be considered as damage a priori. With respect to the small amount of empirical material available herein, it is necessary to consider the achieved results as preliminary results. The method based on observation of whole-stands growth changes on the basis of long-term measurements obtained from long-term research plots is not sufficiently sensitive to the whole-area rooting of the soil surface by wild boars. It is hard to assume whether it would be suitable for comparison of growth changes with a control variant, i.e. without any rooting. The method of observation of radial increments on tree ring cores is more sensitive and persuasive. It is based on the ability of individual trees to respond to changed growth conditions.

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# Modelling of the 1D Convective Heat Exchange between Logs Subjected to Freezing and to Subsequent Defrosting and the Surrounding Environment

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**Abstract** – A 1D mathematical model for the computation of the temperature on the surface of cylindrical logs,  $t_{sr}$ , and the non-stationary temperature distribution along the radiuses of logs subjected to freezing and subsequent defrosting at convective exponentially changing boundary conditions has been suggested. The model includes mathematical descriptions of the thermal conductivity in radial direction,  $\lambda_r$ , the effective specific heat capacity,  $c_e$ , and the density,  $\rho$ , of the non-frozen and frozen wood, and also of the heat transfer coefficient between the surrounding air environment and the radial direction of horizontally situated logs,  $\alpha_r$ . With the help of the model, computations have been carried out for the determination of  $\alpha_r$ ,  $t_{sr}$ ,  $\lambda_{sr}$ , and 1D temperature distribution along the radiuses of beech logs with diameters of 0.24 m, initial temperature 20 °C, and moisture content 0.4 kg·kg<sup>-1</sup>, 0.8 kg·kg<sup>-1</sup>, and 1.2 kg·kg<sup>-1</sup>, during their freezing at –20 °C, and during subsequent thawing at 20 °C.

heat transfer coefficient / surface temperature / temperature distribution / beech logs / radius

**Kivonat** – Egydimenziós konvektív hővezetés modellezése fagyott és normál állapotú rönk és környezte között. Célunk egy 1D matematikai modell létrehozása volt, amely kiszámítja a hengeres farönk felületi hőmérsékletét,  $t_{sr}$ , és a rönk sugara menti hőmérséklet-eloszlást egy olyan hengeres farönkön, amelyet lefagyasztottak majd kiolvasztottak exponenciálisan változó hőátadási körülmények között. A modell magában foglalja a sugárirányú hővezetési tényező,  $\lambda_r$ , az effektív specifikus fajhő  $c_e$ , és a sűrűség  $\rho$  matematikai leírását nem-fagyott és fagyott állapotú faanyag esetében. Tartalmazza továbbá az  $\alpha_r$  radiális irányú hőátadási tényezőt a környező levegő és a vízszintesen fekvő rönk között. A modell segítségével számítások történtek az  $\alpha_r$ , a  $t_{sr}$ , és a  $\lambda_{sr}$ , valamint az 1D hőmérsékleteloszlás meghatározására 0,24 m átmérőjű bükk rönknél a sugár mentén a következő feltételek mellett: kezdeti hőmérséklet 20 °C, a nedvességtartalom értékei 0,4 kg·kg<sup>-1</sup>, 0,8 kg·kg<sup>-1</sup> és 1,2 kg·kg<sup>-1</sup>, a –20 °C-os fagyasztás során és az ezt követő felolvasztás folyamán 20°C-ig.

hővezetési együttható / felületi hőmérséklet / hőmérsékleteloszlás / bükk rönk / sugár

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## 1 INTRODUCTION

Logs prepared for veneer production are subjected to freezing and thawing in natural air conditions during the winter. The duration time and energy needed for the thermal treatment of frozen logs with the goal of plasticizing depends greatly on the level of freezing within the logs. (Sergovsky 1975, Shubin 1990, Trebula – Klement 2002, Videlov 2003, Deliiski 2004, 2009).

In the accessible specialized literature there are very few reports about the temperature distribution in frozen logs subjected to thawing (Steinhagen 1986, 1991, Steinhagen et al. 1987, Steinhagen – Lee 1988, Khattabi – Steinhagen 1992, 1993, 1995, Deliiski 2005, 2011, 2013b) and there is no information at all about the temperature distribution in logs during their natural or artificial freezing. That is why the modelling and the multi-parameter study of the processes of freezing and of the subsequent defrosting (thawing) of logs are of considerable scientific and practical interest.

The aim of the present work is to suggest a 1D mathematical model for the computation of the temperature on the surfaces of cylindrical logs,  $t_{st}$ , and the non-stationary temperature distribution along the radiuses of logs subjected to freezing and subsequent defrosting at convective exponentially changing boundary conditions. To achieve this goal, a base model of the heating and cooling processes of logs is used, one which has earlier been suggested and modified by the first co-author (Deliiski 2005, 2011, 2013b).

### Symbols:

$c$	= specific heat capacity ( $J \cdot kg^{-1} \cdot K^{-1}$ )
$D$	= diameter, m
exp	= exponent
$q$	= specific heat energy ( $kWh \cdot m^{-3}$ )
$r$	= radial coordinate: $0 \leq r \leq R$ , m;
$R$	= radius, m
$t$	= temperature ( $^{\circ}C$ ): $t = T - 273.15$
$T$	= temperature (K): $T = t + 273.15$
$u$	= moisture content ( $kg \cdot kg^{-1}$ ): $u = W/100$
$W$	= moisture content (%): $W = 100u$
$\alpha$	= heat transfer coefficient between the logs' surface and the air environment ( $W \cdot m^{-2} \cdot K^{-1}$ )
$\Delta$	= difference (for the temperature)
$\lambda$	= thermal conductivity ( $W \cdot m^{-1} \cdot K^{-1}$ )
$\rho$	= density ( $kg \cdot m^{-3}$ )
$\tau$	= time (s)
@	= at

### Subscripts and superscripts:

ad	= anatomical direction
b	= basic (for density, based on dry mass divided to green volume)
bw	= bound water
bwm	= maximum possible amount of bound water
c	= center (for the temperature or thermal conductivity on the logs' centers)
dfr	= defrosting (for the temperature of the defrosting medium)
e	= effective (for the specific heat capacity of the frozen and non-frozen wood)
exp	= exponent (for the time constant of the exponentially change in the air temperature)
fr	= freezing (for the temperature or for the duration of the freezing process or for the frozen state of the wood)

fsp	=	fiber saturation point of the wood
fw	=	free water
m	=	medium (for the temperature of the freezing or defrosting air environment)
m0	=	initial (for the medium temperature at the end of the logs freezing or defrosting)
m1	=	end (for the medium temperature at the end of the logs freezing or defrosting)
nfr	=	non-frozen (for the state of the wood)
r	=	radial direction
sr	=	surface on the radial direction
0	=	initial (for the radial or time coordinates or for the average mass temperature of the logs at the beginning of the freezing process)
1	=	final (for the average temperature of the logs at the end of the defrosting process)
273.15	=	at 273.15 K, i.e. at 0 °C (for the wood thermal conductivity)
293.15	=	at 293.15 K, i.e. at 20 °C (for the fiber saturation point of the wood)

## 2 MATERIAL AND METHODS

### 2.1 Mechanism of the 1D heat distribution in the logs

The mechanism of the heat distribution in logs during their heating or cooling can be described by the equation of heat conduction (also known as the equation of Fourier-Kirchhoff). When the length of the logs exceeds their diameter by at least 3 or 4 times, then the heat transfer through the frontal sides of the logs can be ignored, because it does not influence the change in temperature of their cross sections which are equally distant from the frontal sides (Chudinov 1968). In such cases, the following 1D model can be used for the calculation of the change in  $T$  only along the radius of the central cross sections during freezing and defrosting of the logs (i.e. along the coordinate  $r$  of these sections) (Deliiski 2011, 2013b):

$$c_e \rho \frac{\partial T(r, \tau)}{\partial \tau} = \lambda_r \left( \frac{\partial^2 T(r, \tau)}{\partial r^2} + \frac{1}{r} \frac{\partial T(r, \tau)}{\partial r} \right) + \frac{\partial \lambda_r(r, \tau)}{\partial T} \left( \frac{\partial T(r, \tau)}{\partial r} \right)^2 \quad (1)$$

with an initial condition

$$T(r, 0) = T_0 \quad (2)$$

and with one and the same boundary condition for the convective heat transfer valid for the entire surface of the logs:

- during the process of logs freezing:

$$\frac{\partial T(0, \tau)}{\partial r} = - \frac{\alpha_r^{\text{fr}}(\tau)}{\lambda_{\text{sr}}(\tau)} \left[ T_{\text{sr}}^{\text{fr}}(\tau) - T_{\text{m}}^{\text{fr}}(\tau) \right], \quad (3)$$

- during the process of logs defrosting:

$$\frac{\partial T(0, \tau)}{\partial r} = \frac{\alpha_r^{\text{dfr}}(\tau)}{\lambda_{\text{sr}}(\tau)} \left[ T_{\text{sr}}^{\text{dfr}}(\tau) - T_{\text{m}}^{\text{dfr}}(\tau) \right]. \quad (4)$$

It must be noted that the model presented by equations (1) ÷ (4) can also be solved with different mathematically described boundary conditions and heat transfer coefficients within them. The difference between the thermo-physical characteristics of the logs' bark and the logs' wood has not been taken into consideration in the model.

## 2.2 Change in the temperature of the freezing and the defrosting mediums

It is possible to have two cases for freezing of different materials in freezers. The first case is when the material is put into a working freezer with a constant unchanged temperature in it and, consequently, the freezing medium temperature  $T_m(\tau) = T_{m0} = \text{const}$ .

The mathematical model (1) ÷ (4) obtains more complicated boundary conditions in the second case; in this case the material is put into a freezer before it is switched on. Here, the temperature of the air environment in the freezer  $T_m^{\text{fr}}$  decreases exponentially (*Figure 1*) according to the equation

$$T_m^{\text{fr}} = T_{m1}^{\text{fr}} + (T_{m0}^{\text{fr}} - T_{m1}^{\text{fr}}) \exp\left(-\frac{\tau}{\tau_{\text{exp}}^{\text{fr}}}\right). \quad (5)$$

The defrosting of the frozen materials after the freezer's door is opened is realized at the exponential increase of the air temperature (*Figure 1*) according to the equation

$$T_m^{\text{dfr}} = T_{m1}^{\text{dfr}} - (T_{m1}^{\text{dfr}} - T_{m1}^{\text{fr}}) \exp\left(-\frac{\tau - \tau_{\text{fr}}}{\tau_{\text{exp}}^{\text{dfr}}}\right). \quad (6)$$

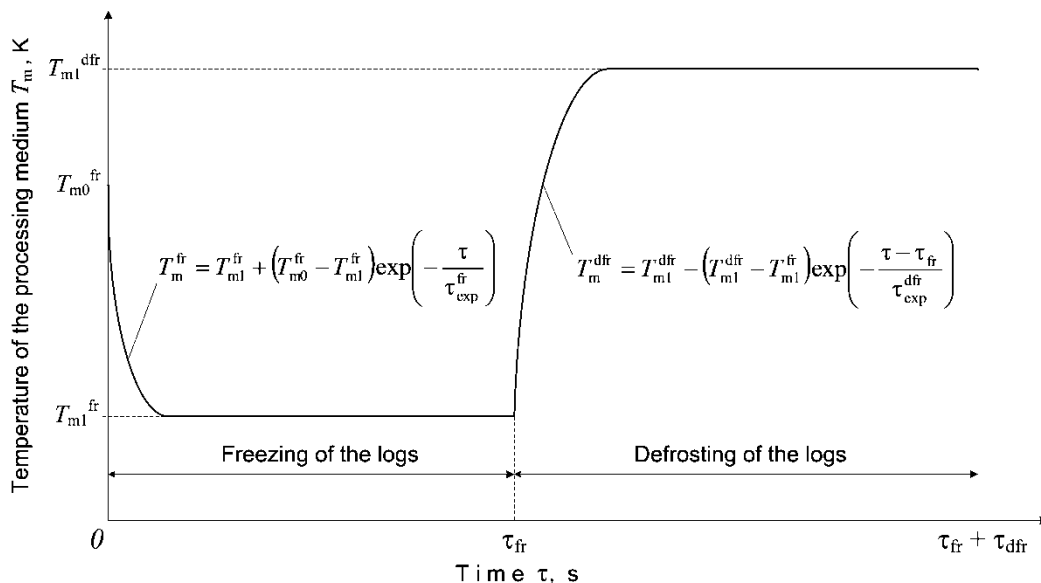


Figure 1. Exponential change in the air medium during the freezing and the defrosting of logs

## 2.3 Heat transfer coefficient between the air and the logs

It is known that the heating or cooling of logs in a gaseous (air) environment takes place through a convective heat exchange between the surfaces of the logs and the moving environment. If the movement is caused by differences in the density of the gas as a consequence of temperature differences in it, it is acceptable to call the convection free (not organized, natural).

As a rule, the freezing of wood materials at atmospheric conditions or in a freezer takes place in the conditions of free convection. For the calculation of the heat transfer coefficient in such conditions of heating or cooling of horizontally situated logs Chudinov (1968) suggests the following experimentally determined equation:

$$\alpha_r = 0.997 \sqrt[4]{\frac{\Delta T}{R}}. \quad (7)$$

It can be assumed that the heat transfer coefficients during freezing and defrosting of the materials are not equal to each other. Real free air convection is observed only during the freezing and defrosting of logs at natural atmospheric conditions or during the defrosting of frozen materials after the freezer's door is opened. That is why it can be written as:

$$\alpha_r^{\text{fr}} \approx 0.997 \sqrt[4]{\frac{\Delta T}{R}} = 0.997 \sqrt[4]{\frac{T(0, \tau) - T_m^{\text{fr}}(\tau)}{R}}, \quad (8)$$

$$\alpha_r^{\text{dfr}} = 0.997 \sqrt[4]{\frac{\Delta T}{R}} = 0.997 \sqrt[4]{\frac{T_m^{\text{dfr}}(\tau) - T(0, \tau)}{R}}. \quad (9)$$

More precise equations for the determination of the heat transfer coefficients between the surfaces of the logs in radial direction and the freezing or defrosting mediums can be obtained after suitable experiments have been carried out.

The model presented by equations (1) ÷ (4) can also be solved with different mathematically described heat transfer coefficients for separate parts of the logs' surfaces. The analysis and presentation of such cases are beyond the scope of this article.

## 2.4 Thermo-physical characteristics of the logs during their freezing and defrosting

The solution of the non-linear 1D mathematical model of the freezing and defrosting processes of logs, which is presented by the equations (1) ÷ (9), can be realized using the mathematical descriptions of the effective heat capacity of the frozen and non-frozen wood,  $c_e$ , the thermal conductivity of the wood in radial direction,  $\lambda_r$ , and the density of frozen and non-frozen wood,  $\rho$ , given in Deliiski (2011, 2013b). With the help of the mathematical description of  $\lambda_r$ , the current values of the thermal conductivity on the logs' surfaces in radial direction  $\lambda_{sr}(0, \tau)$ , which participates in eqs. (3) and (4), can also be calculated during the solution of the model.

The thermal conductivity of the wood can be calculated with the help of the following equations for  $\lambda(T, u, \rho_b, u_{\text{fsp}})$ :

$$\lambda = \lambda_{273.15} \gamma [1 + \beta(T - 273.15)], \quad (10)$$

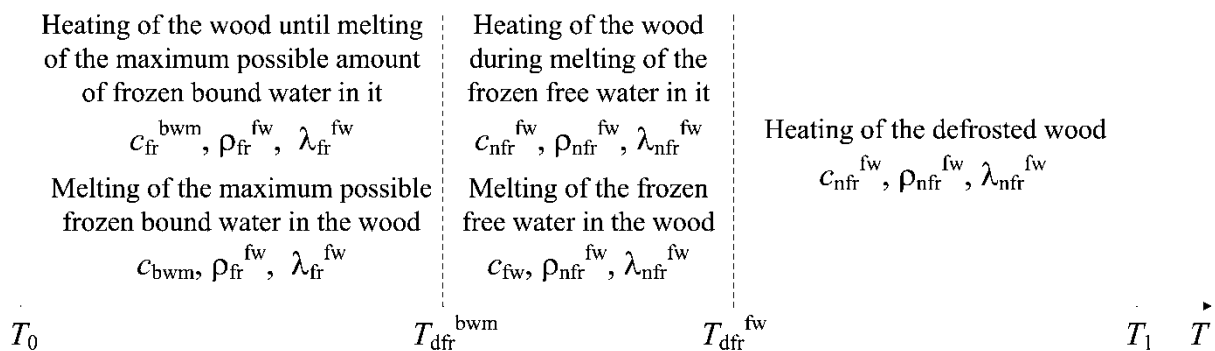
$$\lambda_{273.15} = K_{\text{ad}} \nu [0.165 + (1.39 + 3.8u)(3.3 \cdot 10^{-7} \rho_b^2 + 1.015 \cdot 10^{-3} \rho_b)], \quad (11)$$

$$\nu = 0.15 - 0.07u \quad @ \quad u \leq u_{\text{fsp}} + 0.1 \text{ kg} \cdot \text{kg}^{-1}, \quad (12)$$

$$\nu = 0.1284 - 0.013u \quad @ \quad u > u_{\text{fsp}} + 0.1 \text{ kg} \cdot \text{kg}^{-1}. \quad (13)$$

Equations for the calculation of the variables  $\gamma$  and  $\beta$  in eq. (10) for frozen and non-frozen wood from different wood species and an algorithm for usage of the mathematical description of  $\lambda$  during the solution of the model are given in Deliiski (2013a).

The ice in the wood can be formed from the freezing of hygroscopically bound water or from both the bound and the free water in the wood. It is widely accepted that the phase transition of water into ice and vice versa can be expressed with the help of the so-called “latent heat” in the ice of the frozen body. When solving problems connected to transient heat conduction in frozen wood, it makes sense to include the latent heat in the so-called effective specific heat capacity  $c_e$  (Chudinov 1966, 1968), which is equal to the sum of the wood’s own specific heat capacity  $c$  and the specific heat capacity of the wood which is frozen only with bound water or contains both bound and free water within it. As an example, in *Figure 1*, the symbols of the thermo-physical characteristics of the frozen and non-frozen wood are present, which are necessary for the computation of the temperature distribution in the wood during heating aimed at its defrosting in the respective temperature diapasons and for the computation of the energy consumption for the heating of the wood.



*Figure 2. Using different specific heat capacities –  $c$ , thermal conductivities –  $\lambda$  and densities –  $\rho$ , for the calculation of the temperature distribution in frozen and non-frozen wood when  $u > u_{fsp}$*

A mathematical description of these characteristics is given in Deliiski (2013b). When modelling processes connected with non-stationary temperature distribution in frozen wood with  $u > u_{fsp}$  during its defrosting, it is necessary to take into consideration that the effective specific heat capacity,  $c_e$ , for the separate temperature diapasons on *Figure 2* is equal to:

$$c_e = c_{fr}^{bwm} + c_{bwm} \quad @ \quad T_0 \leq T \leq T_{dfr}^{bwm}, \quad (14)$$

$$c_e = c_{nfr}^{fw} + c_{fw} \quad @ \quad T_{dfr}^{bwm} < T \leq T_{dfr}^{fw}, \quad (15)$$

$$c_e = c_{nfr}^{fw} \quad @ \quad T_{dfr}^{fw} < T \leq T_1. \quad (16)$$

During the computations of the freezing and defrosting processes of logs, effective heat capacities and densities of the wood subjected to thawing have been used and are shown in the mathematical descriptions of thermal conductivities below *Figure 2* (Deliiski 2011, 2013a, 2013b). The small difference (so named hysteresis) between these thermo-physical characteristics, and also between the temperatures  $T_{fr}^{bwm}$  and  $T_{dfr}^{bwm}$ , and between  $T_{fr}^{fw}$  and  $T_{dfr}^{fw}$  during the freezing and defrosting of the wood (Chudinov 1966, 1968) needs to be additionally studied and mathematically described.

### 3 RESULTS AND DISCUSSION

The abovementioned mathematical descriptions of  $T_m^{\text{fr}}$ ,  $T_m^{\text{dfr}}$ ,  $\alpha_r^{\text{fr}}$  and  $\alpha_r^{\text{dfr}}$  have been introduced before by the first co-author who earlier created and later modified a non-stationary model of the heating and cooling of cylindrical wood materials (Deliiski 2005, 2011, 2013b). This model is presented in common form by the eqs. (1) ÷ (4).

The updated model with the descriptions of  $T_m^{\text{fr}}$ ,  $T_m^{\text{dfr}}$ ,  $\alpha_r^{\text{fr}}$ , and  $\alpha_r^{\text{dfr}}$  has been solved with the help of explicit schemes of the finite difference method which, in a way, is analogous to the one used and described in (Deliiski 2009, 2011) for the solution of a model of the heating and cooling process of cylindrical wood materials. During the computations, these schemes allow for the determination of the temperature at each node of the calculation mesh using the current values of the thermo-physical characteristics of the frozen or non-frozen wood depending on the momentous aggregate state of the bound and free water in the wood in separate nodes.

To help with the solutions of the updated model, a software program has been prepared in the calculation environment of Visual FORTRAN Professional, which is a part of the office-package of Windows. The program helped carry out computations for the determination of the 1D change of temperature in beech logs (*Fagus Silvatica L.*) subjected to 50 hours freezing at  $t_{m1}^{\text{fr}} = -20$  °C and a following 50 h defrosting at  $t_{m2}^{\text{dfr}} = -20$  °C .

The freezing and subsequent defrosting of logs with a diameter of  $D = 0.24$  m (i.e. with a radius of  $R = 0.12$  m), initial wood temperature  $t_0 = 20$  °C and three values of the wood moisture content:  $u = 0.4$  kg · kg<sup>-1</sup>,  $u = 0.8$  kg · kg<sup>-1</sup>, and  $u = 1.2$  kg · kg<sup>-1</sup> have been studied. The moisture content of the beech logs is usually situated in this range of  $u$ , which are used for the production of veneer. All logs with such  $u$  contain a maximum possible quantity of bound water. Besides this, the logs with  $u = 0.4$  kg · kg<sup>-1</sup> contain a relatively little amount of free water, the log with  $u = 0.8$  kg · kg<sup>-1</sup> contains a significant quantity of free water and the log with  $u = 1.2$  kg · kg<sup>-1</sup> contains almost a maximum possible quantity of free water.

The decreasing of  $t_m^{\text{fr}}$  from the value of  $t_{m0}^{\text{fr}} = t_0 = 20$  °C to  $t_{m1}^{\text{fr}} = -20$  °C = const and the following increasing of  $t_m^{\text{dfr}}$  from  $t_{m1}^{\text{fr}} = -20$  °C = const to  $t_{m1}^{\text{dfr}} = 20$  °C = const go exponentially with time constants  $\tau_{\text{exp}}^{\text{fr}} = \tau_{\text{exp}}^{\text{dfr}} = 3600$  s. The calculated values according to eqs. (5) and (6) exponential change of  $t_m^{\text{fr}}$  and  $t_m^{\text{dfr}}$  can be seen in the *Figure 2* for the curve of  $t_m$ .

The duration of 50 h of the logs freezing at  $t_{m1}^{\text{fr}} = -20$  °C has been proven as being enough for the reaching of a complete freezing of the free water in the all of the studied logs. The calculations were done with average values of basic density of beech wood  $\rho_b = 560$  kg · m<sup>-3</sup> and fiber saturation point at 293.15 K (i.e. at 20 °C) of this wood  $u_{\text{fsp}}^{293.15} = 0.31$  kg · kg<sup>-1</sup> (Nikolov – Videlov 1987, Pozgaj et al. 1997). A coefficient  $K_r = K_{\text{ad}} = 1.35$  (Deliiski 2003, 2013a) in eq. (11) for the beech wood was used.

*Table 1* shows the computed distribution of the temperature in 4 equally distant from each other nodes of the calculation mesh in the central cross-section of the beech log with  $u = 0.8$  kg · kg<sup>-1</sup> at every 2 h of the freezing and of the following defrosting processes. The corresponding input data, which is used for the solution of the 1D model, is underlined in *Table 1*. The remaining input data, which is not underlined in this table, relates mainly to the

parameters of the equipment with which the thermal treatment of the wood materials with the aim of their freezing or defrosting is carried out. Using this input data, the energy parameters of the freezing or defrosting process and the efficiency from the usage of the equipment can be calculated. The fourth column from right to left on *Table 1* shows the calculated values of  $\alpha_r$  according to eq. (8) and the last two columns of *Table 1* show the calculated values of the wood thermal conductivities on the log's surface,  $\lambda_{sr}$ , and in the center of the log,  $\lambda_c$ , during the freezing and defrosting processes.

In *Figure 3* the computed change in the freezing and in the defrosting medium temperatures,  $t_m^{fr}$  and  $t_m^{dfr}$  respectively (both temperatures are shown as  $t_m$  on the figure), in the surface temperature of the log,  $t_{sr}^{fr}$  and  $t_{sr}^{dfr}$  (both of them are shown as  $t_{sr}$  on the figure) and also in the temperature in the central points of the log,  $t_c$ , during the freezing and subsequent defrosting, depending on the wood moisture content,  $u$ , is shown.

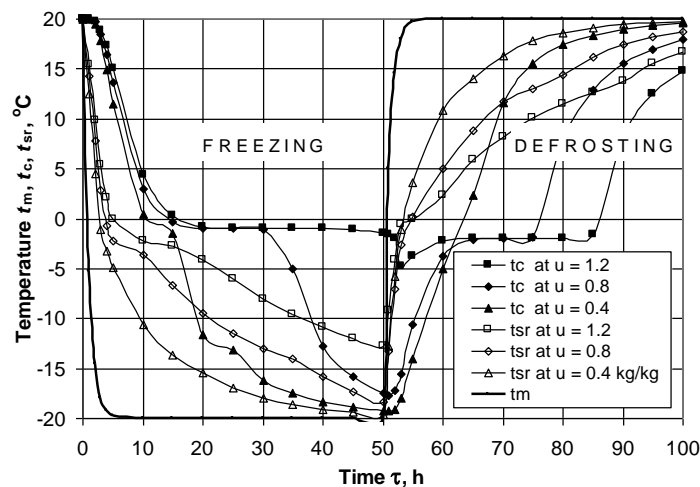


Figure 3. Change in  $t_m$ ,  $t_{sr}$ , and  $t_c$  of beech logs with  $D = 0.24$  m and  $t_0 = 20$  °C during their 50 h freezing at  $-20$  °C and following 50 h defrosting at  $20$  °C, depending on  $u$

In *Figure 4* the computed change in the heat transfer coefficient between the surfaces of the logs in radial direction,  $\alpha_r$ , and the freezing and defrosting mediums during the studied processes, depending on  $u$  is shown.

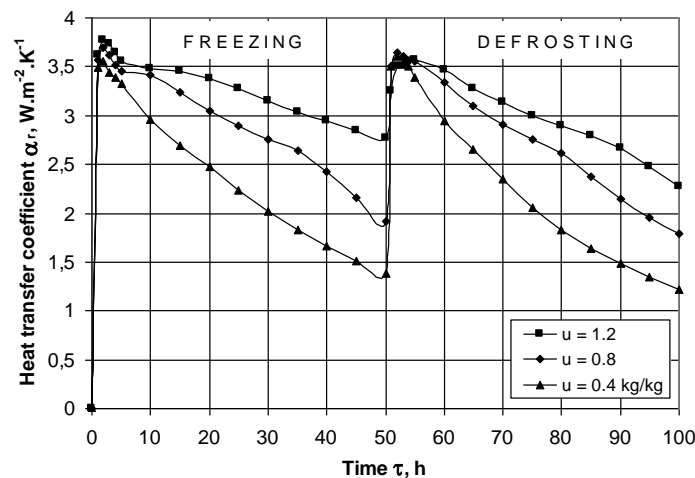


Figure 4. Change in  $\alpha_r$  of beech logs with  $D = 0.24$  m and  $t_0 = 20$  °C during their 50 h freezing at  $-20$  °C and following 50 h defrosting at  $20$  °C, depending on  $u$

Table 1. Change in  $T_m^{fr}$  and  $T_m^{dfr}$  (third column),  $T_{sr}^{fr}$  and  $T_{sr}^{dfr}$  (fourth column),  $\alpha_r$  (fourth column from right), in  $\lambda_{sr}$  and  $\lambda_c$  (second and first column from right), and in  $t$  in 4 equally distant from each other characteristic points of the central cross section of a beech log with  $D = 0.24$  m,  $t_0 = 20^\circ\text{C}$ , and  $u = 0.8$  kg·kg<sup>-1</sup> during every 2 h of its freezing at  $t_{ml}^{fr} = -20^\circ\text{C}$  and during the subsequent defrosting at  $t_{ml}^{dfr} = 20^\circ\text{C}$

I N P U T D A T A

FREEZING AND DEFROSTING OF A BEECH LOG WITH DIAMETER OF 0,24 m

Kq=11 M= 11 N= 0 KD= 1 Ry=560. Kwr=1.35 Kwpr=1.78 U=0.800 Ufsp293=0.31 D=2.4 L= 9.0  
 to= 20.0 tmo= 20.0 tmfr=-20.0 tmdfr= 20.0 t3= 0.0 t4= 0.0 dtm=.001 t01= 0. dTAU= 80  
 Tfr=3600. Tdfr=3600. T3= 0. dT3= 0. dtm3= 0. T4= 0. T5= 0. TAUpoc.=360000 INT= 7200  
 ds=.008 Si=.10 Roi=120. Ai=.00000022 dFa=0.05 KK=.2 tcenter=-17.45 dtwc= 0.1 Ts= 0  
 Pw=.30 Vw=14.39 Va=47.95 tbi= 0. Sim=0.200 Xp=1.00 L-log=0.00 D-log=.24 dx=.01200

R E S U L T S

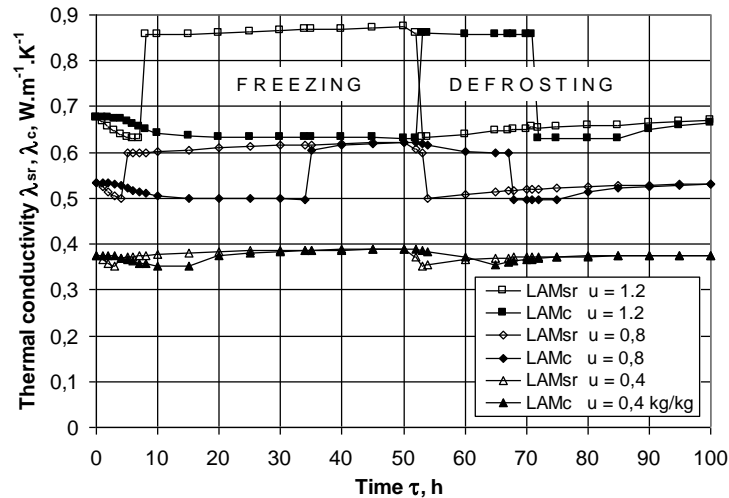
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Time	Energy	Temperature		Temp.in charact.points				on R,mm	tav	dtav/	ALFAr		LAMsr	LAMc
TAU	q	tm	tsr	30	60	90	120	Simpson	dtau	Chud.	Videl.	W/m.K	W/m.K	
h	kWh/m3	oC	oC	oC	oC	oC	oC	oC	oC/h	W/m2.K	W/m2.K			
0	0.00	20.0	20.00	20.0	20.0	20.0	20.0	20.00	0.00	0.00	0.00	0.534	0.534	
2	3.28	-14.6	7.79	11.9	17.3	19.0	19.7	15.91	-3.18	3.69	4.36	0.514	0.533	
4	8.49	-19.3	-0.76	3.0	10.4	14.1	16.4	9.37	-3.16	3.52	4.15	0.499	0.528	
6	12.35	-19.9	-2.41	-0.1	4.9	8.4	10.8	4.75	-1.63	3.46	4.09	0.600	0.518	
8	14.41	-20.0	-2.79	-0.9	2.2	4.4	6.1	2.14	-1.05	3.45	4.07	0.600	0.511	
10	16.49	-20.0	-3.59	-1.1	0.7	2.0	3.0	0.49	-1.10	3.41	4.03	0.602	0.505	
12	17.36	-20.0	-4.74	-1.8	-0.3	0.5	1.2	-0.67	-0.39	3.35	3.95	0.603	0.502	
14	18.44	-20.0	-6.26	-3.8	-0.8	-0.3	0.1	-1.63	-0.37	3.26	3.85	0.606	0.501	
16	19.75	-20.0	-7.14	-5.0	-1.0	-0.8	-0.6	-2.39	-0.52	3.21	3.79	0.607	0.499	
18	20.14	-20.0	-8.41	-6.4	-1.1	-1.0	-0.9	-3.01	-0.19	3.13	3.69	0.609	0.499	
20	21.00	-20.0	-9.50	-7.8	-1.7	-1.0	-1.0	-3.78	-0.27	3.05	3.60	0.610	0.499	
22	22.27	-20.0	-10.33	-8.8	-3.7	-1.1	-1.0	-4.57	-0.37	2.99	3.53	0.612	0.499	
24	23.03	-20.0	-11.14	-9.7	-4.6	-1.2	-1.0	-5.18	-0.73	2.92	3.45	0.613	0.499	
26	23.49	-20.0	-11.87	-10.6	-6.2	-1.8	-1.0	-5.99	-0.28	2.86	3.38	0.614	0.499	
28	24.78	-20.0	-12.44	-11.3	-7.3	-3.6	-1.0	-6.83	-0.37	2.81	3.32	0.615	0.499	
30	25.12	-20.0	-13.00	-11.9	-8.1	-4.3	-1.1	-7.42	-0.24	2.76	3.25	0.616	0.499	
32	26.01	-20.0	-13.47	-12.5	-9.1	-5.8	-1.2	-8.28	-0.31	2.71	3.20	0.616	0.498	
34	27.30	-20.0	-13.89	-13.0	-9.8	-6.8	-1.7	-9.12	-0.48	2.66	3.15	0.617	0.498	
36	28.52	-20.0	-14.32	-13.5	-11.0	-9.1	-7.5	-10.94	-1.01	2.62	3.09	0.617	0.607	
38	29.46	-20.0	-15.03	-14.4	-12.7	-11.5	-10.6	-12.72	-0.79	2.53	2.99	0.619	0.612	
40	30.19	-20.0	-15.82	-15.3	-14.1	-13.2	-12.7	-14.12	-0.62	2.42	2.86	0.620	0.615	
42	30.76	-20.0	-16.53	-16.1	-15.2	-14.6	-14.1	-15.24	-0.49	2.31	2.73	0.621	0.617	
44	31.21	-20.0	-17.12	-16.8	-16.1	-15.6	-15.3	-16.12	-0.40	2.21	2.61	0.622	0.619	
46	31.56	-20.0	-17.61	-17.4	-16.8	-16.4	-16.2	-16.83	-0.32	2.11	2.49	0.622	0.620	
48	31.85	-20.0	-18.01	-17.8	-17.4	-17.1	-16.9	-17.40	-0.25	2.01	2.38	0.623	0.621	
50	32.08	-20.0	-18.34	-18.2	-17.8	-17.6	-17.5	-17.85	-0.20	1.92	2.27	0.623	0.622	
52	30.00	14.5	-7.07	-10.3	-14.9	-16.5	-17.2	-13.80	3.22	3.65	4.32	0.607	0.622	
54	25.73	19.3	-0.15	-3.1	-8.4	-11.3	-13.1	-7.79	2.05	3.56	4.20	0.500	0.616	
56	23.67	19.9	0.48	-2.0	-5.0	-7.0	-8.5	-4.81	1.04	3.56	4.20	0.501	0.609	
58	21.64	20.0	2.78	-1.3	-3.3	-4.6	-5.5	-2.84	0.65	3.45	4.07	0.505	0.605	
60	20.23	20.0	4.96	1.6	-2.4	-3.2	-3.8	-1.36	0.55	3.34	3.94	0.509	0.602	
62	18.49	20.0	6.61	3.9	-2.0	-2.4	-2.8	-0.05	0.54	3.24	3.83	0.511	0.600	
64	17.27	20.0	8.01	5.7	-1.6	-2.1	-2.2	1.03	0.55	3.15	3.72	0.514	0.600	
66	15.64	20.0	9.27	7.2	0.8	-2.0	-2.0	2.10	0.69	3.07	3.62	0.516	0.599	
68	13.56	20.0	10.41	8.6	2.1	-1.8	-2.0	2.96	1.08	2.98	3.52	0.518	0.497	
70	12.71	20.0	11.32	9.7	4.2	-1.2	-2.0	4.05	0.37	2.91	3.43	0.519	0.497	
72	11.81	20.0	12.06	10.6	5.8	1.3	-2.0	5.18	0.42	2.84	3.36	0.520	0.497	
74	11.03	20.0	12.71	11.4	7.0	2.9	-1.9	6.18	0.54	2.78	3.29	0.522	0.497	
76	10.36	20.0	13.28	12.1	8.0	4.0	-1.6	7.03	1.61	2.73	3.22	0.522	0.498	
78	8.89	20.0	13.76	12.7	9.1	6.2	3.7	8.89	1.24	2.68	3.16	0.523	0.506	
80	7.21	20.0	14.38	13.5	11.0	9.2	7.9	11.01	0.91	2.61	3.08	0.524	0.514	
82	5.93	20.0	15.14	14.4	12.5	11.3	10.4	12.62	0.71	2.52	2.97	0.526	0.518	
84	4.92	20.0	15.86	15.3	13.8	12.9	12.2	13.88	0.56	2.42	2.85	0.527	0.521	
86	4.11	20.0	16.47	16.0	14.8	14.1	13.6	14.89	0.46	2.32	2.74	0.528	0.523	
88	3.46	20.0	16.99	16.6	15.7	15.1	14.6	15.71	0.37	2.23	2.64	0.529	0.525	
90	2.92	20.0	17.42	17.1	16.3	15.8	15.5	16.38	0.30	2.15	2.54	0.529	0.526	
92	2.48	20.0	17.78	17.5	16.9	16.5	16.2	16.93	0.25	2.07	2.44	0.530	0.527	
94	2.11	20.0	18.09	17.9	17.3	17.0	16.8	17.38	0.21	1.99	2.35	0.530	0.528	
96	1.81	20.0	18.34	18.2	17.7	17.5	17.3	17.76	0.17	1.92	2.27	0.531	0.529	
98	1.56	20.0	18.56	18.4	18.0	17.8	17.7	18.07	0.14	1.86	2.19	0.531	0.530	
100	1.35	20.0	18.74	18.6	18.3	18.1	18.0	18.34	0.12	1.79	2.12	0.532	0.530	

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E N D

The computed change in the thermal conductivity of the surfaces of the logs in radial direction,  $\lambda_{sr}$ , and of the centres of the logs  $\lambda_c$ , depending on  $u$  during the freezing and defrosting processes is shown in *Figure 5*.



*Figure 5.* Change in  $\lambda_{sr}$  and in  $\lambda_c$  of beech logs with  $D = 0.24$  m and  $t_0 = 20$  °C during their 50 h freezing at  $-20$  °C and following 50 h defrosting at  $20$  °C, depending on  $u$

In Deliiski (2009) curves showing the percentage of water being in ice-phase on the diameter of beech logs subjected to defrosting with  $t_0 = -20$  °C as a function of  $u > u_{fsp}$  and the relationship  $\tau / D$  can be seen.

The obtained results lead to the following conclusions:

1. On the curves of characteristic points situated on the logs' centers in *Figure 3*. the specific almost horizontal sections of retention of the temperature  $t_c$  for a long period of time in the range from  $-1$  °C to  $-2$  °C can be seen, while in these points a complete freezing of the free water and after that a complete melting of the ice created by it occurs in the wood (Chudinov 1966, 1968).

It can be noted that such retention of the temperature on the logs' axes has been observed in wide experimental studies during the defrosting process of pine logs containing ice from the free water (Steinhagen 1986, Khattabi – Steinhagen 1992, 1993).

2. The character of the change in the heat transfer coefficient,  $\alpha_r$ , is almost identical during the studied freezing and defrosting processes of logs with given value of the wood moisture content (*Figure 4*). The reason for this is the equality of the difference  $t_0 - t_m^{fr}$  during the freezing with the difference  $t_m^{dfr} - t_m^{fr}$  during the defrosting for one and the same moments from the beginning of these processes at  $t_{m0}^{fr} = 20$  °C,  $t_{m1}^{fr} = -20$  °C,  $t_{m1}^{dfr} = 20$  °C, and  $\tau_{exp}^{fr} = \tau_{exp}^{dfr} = 3600$  s. At other values of these variables, the change in  $\alpha_r$  during the freezing and the defrosting would be different.

3. With the increase of the duration of the freezing and the defrosting processes, the coefficient  $\alpha_r$  decreases because of the decreasing of the difference  $\Delta T$  between the processing medium temperature and the surface temperature of the logs (see eqs. (8) and (9)).

4. The character of the change in the wood thermal conductivity on the logs' surfaces,  $\lambda_{sr}$ , and in the logs' centers,  $\lambda_c$ , is very complex (*Figure 5*). The current values of  $\lambda_{sr}$  and of  $\lambda_c$  depend not only on the wood moisture content and on the current temperature in the respective points on the logs' radiuses, but also on the momentous aggregate condition of the water in these points (Deliiski, 2013a). The larger values of  $\lambda_{sr}$  and  $\lambda_c$  in *Figure 5* related to

the central points or surfaces of the frozen logs, and the lower values of  $\lambda_{sr}$  and  $\lambda_c$  related to the logs' points with frozen free water in them at respective moments.

Table 1, Figure 3, and Figure 5 show that the complete freezing of the free water at the surface and at the center of the beech log with  $u = 0.8 \text{ kg}\cdot\text{kg}^{-1}$  occurs between 4 and 6 h and between 34 and 36 h respectively, when the temperature in these points becomes lower than  $-2 \text{ }^\circ\text{C}$ . Analogously, the melting of the frozen free water at the surface and at the center of this log starts between 52 and 54 h and between 66 and 68 h respectively. The complete melting of the frozen free water at the center of the log occurs between 76 and 78 h, when the temperature in this point becomes higher than  $-1 \text{ }^\circ\text{C}$ .

## 4 CONCLUSIONS

The present paper describes the 1D mathematical model for the computation of the temperature on the surfaces of logs suggested by the authors,  $t_{sr}$ , and the non-stationary temperature distribution along the radiuses of logs subjected to freezing and to subsequent defrosting at convective exponentially changing boundary conditions. As a base model, the heating and cooling processes of logs is used, which was created and modified earlier by the first co-author. The mechanism of the heat distribution along the radiuses of the logs during their freezing and subsequent defrosting is described by the 1D partial differential equation of heat conduction. For the solution of the model, an explicit form of the finite-difference method is used, which allows for the exclusion of any simplifications in the model.

For the numerical solution of the model, a software program has been prepared in FORTRAN, which has been input in the Visual Fortran Professional calculation environment developed by Microsoft. With the help of the program an example computation has been carried out for the determination of the 1D change in the temperature along the radiuses of beech logs with diameter 0.24 m, initial temperature  $20 \text{ }^\circ\text{C}$  and moisture content  $0.4 \text{ kg}\cdot\text{kg}^{-1}$ ,  $0.8 \text{ kg}\cdot\text{kg}^{-1}$  and  $1.2 \text{ kg}\cdot\text{kg}^{-1}$ , during 50 hours freezing at exponentially decreasing air temperature until reaching of  $-20 \text{ }^\circ\text{C}$  and during the following 50 h defrosting at exponentially increasing air temperature until reaching of  $20 \text{ }^\circ\text{C}$ .

The results presented in the figures in this paper show that the procedures for calculation of the non-stationary 1D temperature change in the prepared software program function well for the mutually connected processes of the freezing and the defrosting of the logs at convective boundary conditions.

The obtained results show the complex character of the change in the temperature on the logs' surfaces and along the logs' radiuses, and also of the heat transfer coefficient between the logs' surfaces and the processing freezing or defrosting air environment. Also the change in the wood thermal conductivity on the logs' surfaces and in the separate points along the logs' radiuses, especially strong depending on the aggregate condition of the water in each point at every moment of the studied processes, has a very complex character.

The presented model, after its update with new experimentally obtained data about the heat transfer coefficient and with mathematically described hysteresis between the thermo-physical characteristics of the wood during its freezing and defrosting, can be used for a science-based determination of the duration of the freezing and defrosting processes of logs at different initial and boundary conditions.

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# ACTA SILVATICA ET LIGNARIA HUNGARICA

## Vol. 11, Nr. 2

### Contents

SZŐCS, Levente – MELIKA, George – THURÓCZY, Csaba – CSÓKA, György: Parasitoid complex of leafminer <i>Pyllonorycter comparella</i> (Lepidoptera, Gracillariidae) in Hungary .....	91
NOWINSZKY, László – PUSKÁS, János: Sex Ratio Analysis of Some Macrolepidoptera Species Collected by Hungarian Forestry Light-Traps .....	99
FOLCZ, Ádám – BÖRCsök, Zoltán: Macrofungi in the Botanical Garden of the University of West Hungary, Sopron .....	111
KUČERA, Peter: On the Occurrence of Natural Norway spruce Woodland in the Pieniny Mts (Western Carpathians) .....	123
KESERŰ, Zsolt – BALLA, Ildikó – ANTAL, Borbála – RÉDEI, Károly: Micropropagation of Leuce-poplars and evaluation of their development under sandy site conditions in Hungary .....	139
VIZVÁRI, Zoltán – KISS, Tibor – MÁTHÉ, Kálmán – ODRY, Péter – VÉR, Csaba – DIVOS, Ferenc: Multi-Frequency Electrical Impedance Measurement on a Wooden Disc Sample .....	153
<b>Guide for Authors</b> .....	163
<b>Contents and Abstracts of Bulletin of Forestry Science, Vol. 5, 2014</b> The full papers can be found and downloaded in pdf format from the journal's webpage ( <a href="http://www.erdtudkoz.hu">www.erdtudkoz.hu</a> ) .....	165

# ACTA SILVATICA ET LIGNARIA HUNGARICA

## Vol. 11, Nr. 2

### Tartalomjegyzék

SZŐCS Levente – MELIKA, George – THURÓCZY Csaba – CSÓKA György: A <i>Populus alba</i> -n élő <i>Phyllonorycter comparella</i> (Lepidoptera: Gracillariidae) levélaknázó magyarországi parazitoid együttese	91
NOWINSZKY László – PUSKÁS János: A magyar erdészeti fénycsapdák által gyűjtött néhány macrolepidoptera faj ivararányának elemzése	99
FOLCZ Ádám – BÖRCsök Zoltán: Nagygombák a Nyugat-magyarországi Egyetem Soproni Botanikus Kertjében	111
KUČERA, Peter: A közönséges luc természetes állománya a Pieninekben (Nyugati-Kárpátok)	123
KESERŰ Zsolt – BALLA Ildikó – ANTAL Borbála – RÉDEI Károly: Leuce-nyárok mikroszaporítása és homokterületen történő termesztésük kiértékelése Magyarországon	139
VIZVÁRI Zoltán – KISS Tibor – MÁTHÉ Kálmán – ODRY Péter – VÉR Csaba – DIVOS, Ferenc: Fakorongok multi-frekvenciás elektromos impedancia mérése	153
<b>Szerzői útmutató</b>	163
<b>Erdészettudományi Közlemények 2015. évi kötetének tartalma és a tudományos cikkek angol nyelvű kivonata</b> A tanulmányok teljes terjedelemben letölthetők pdf formátumban a kiadvány honlapjáról ( <a href="http://www.erdtudkoz.hu">www.erdtudkoz.hu</a> )	165

## Parasitoid complex of leaf miner *Phyllonorycter comparella* (Lepidoptera, Gracillariidae) in Hungary

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**Abstract** – Between 2011 and 2014, 1,154 mines of *Phyllonorycter comparella* (Duponchel) were collected at 12 locations in Hungary and were put into single-mine rearing containers. A total of 574 parasitoid specimens belonging to 29 parasitoid species (26 Chalcididae, 2 Encyrtidae and 1 Braconidae) emerged. Of these species, 13 have not yet been mentioned in either international or in Hungarian literature as a parasitoid of the *P. comparella*. The species assemblages of the parasitoid complexes varied greatly among the sample sites. The primary dominant species of the total samples was found to be *Sympiesis sericeicornis* (Nees), an abundant idiobiont solitary ectoparasitoid. Among the species reared, we have found specialist parasitoids such as *Achrysocharoides scaposa* (Erdős) and even species never recorded from *Populus* (*Zagrammosoma variegatum* (Masi)) according to the Universal Chalcidoidea Database.

### *Phyllonorycter comparella* / *Populus alba* / parasitoid assemblages

**Kivonat** – A *Populus alba*-n él *Phyllonorycter comparella* (Lepidoptera: Gracillariidae) levélaknázó magyarországi parazitoid együttese. 1154 *Phyllonorycter comparella* (Duponchel) levélaknát gyjtöttünk és tettünk egyedi nevelésbe 2011–2014 között, 12 Magyarországi helyszínr. 1. A nevelésb 1 kikelt 574 parazitoid egyed alapján 29 fajt (26 Chalcididae, 2 Encyrtidae és 1 Braconidae) sikerült azonosítani. Ezek közül 13 fajt sem a nemzetközi sem a hazai szakirodalom korábbról nem említi, mint a *P. comparella* parazitoidjait. Különböz mintagyjtési helyszínek parazitoid komplexum fajegyüttesében eltérések mutatkoztak. A teljes minta els dleges domináns faja a *Sympiesis sericeicornis* (Nees), egy gyakori idiobiont szoliter ektoparazitoid volt. A nevelésb 1 kikelt fajok között olyan specialista fajokat is azonosítottunk, mint az *Achrysocharoides scaposa* (Erdős) valamint *Populus*ról eddig, a Nemzetközi Chalcidoidea Adatbázis (Universal Chalcidoidea Database) által még nem regisztrált *Zagrammosoma variegatum* (Masi) parazitoid fajt is.

## 1 INTRODUCTION

*Phyllonorycter comparella* (Duponchel) is a palaeartic species occurring all over the Eurasian continent (De Prins – De Prins 2014). Its main host plant is *Populus alba* (Gozmány 1956), but it can also be found on *Populus tremula* (Linné) and *Populus × canescens* (Ation) (De Prins – De Prins 2014). It is a bivoltine species; the adults fly from June till August and

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also in October (Sz cs 1977). The species overwinters in the adult stage. The blotch mines are located on the underside of the leaf between the leaf veins (Csóka 2003).

Based on our observations, *Ph. comparella* can locally occur at a very high density with several mines on a single leaf. Similar observations have been made by Yedfremova et al. in Russia (Ul'yanovsk Province) in 2007 (Yefremova et al. 2009). A number of papers predict a change in insect behaviour because of the effects of climate change (Cannon 1998, Csóka 1996, Csóka 1997, Dukes et al. 2009, Fleming – Volney 1995, Kirilenko – Sedjo 2007, Walther et al. 2002), and common and even rare species may become severe pests. Therefore, we believe that this species has the potential ability to cause severe outbreaks and possibly heavy damage on *Populus alba* and *Populus × canescens*.

It is always important to study the regulating potential and the species composition of the natural enemy complex of such a species.

The parasitoid complex of *Ph. comparella* has been inadequately studied thus far. Two of the main parasitoid databases publish a rather small number of parasitoids (De Prins – De Prins 2014, Noyes 2015). In Hungary only two researchers (Erd s 1956, Sz cs 1965, Sz cs 1979) reared parasitoids from *Ph. comparella*. The aim of this work is to summarize the knowledge of the parasitoid species developing in *Ph. comparella* in Hungary in comparison with the data available in European literature.

## 2 MATERIAL AND METHODS

We studied the parasitoid complexes of various leaf miners over a period of four years (2011–2014). A total of 1,154 leaf mines were collected from *P. alba* at 12 locations across Hungary (Table 1).

Single mine rearings were used. The mines were cut out from the leaf in order to avoid contamination from other parasitoids (i.e. egg or aphid parasitoids) which parasitize other insects on leaves. After a short drying period, the samples were placed individually into air ventilated plastic rearing tubes.

The parasitoids that emerged were preserved in 95% ethanol, and later identified by Csaba Thuróczy and George Melika. The emerging leaf miner adults were also identified to make sure that the identification of the leaf mines was correct. After the identification process, the adult insects were kept in alcohol for further molecular analysis.

## 3 RESULTS AND DISCUSSION

From the samples collected during the four consequent years (2011–2014), a total of 574 parasitoid specimens emerged from 255 parasitized samples. These specimens belong to 29 parasitoid species (26, 2, and 1 species to the families Chalcididae, Encyrtidae and Braconidae, respectively). The total parasitization rate of our samples was 22.1% (n=1154 samples collected), and at different sample locations (with regard to the sample size) varied between 7.3% (n=55) and 75% (n=4) (Table 1).

Table 1. A summary of the sampling locations, the sample sizes and the parasitism rates of our rearings.

WGS84: the coordinates of the sample plots;

TMC: total leaf mines collected; TPM: total parasitized leaf mines;

TSE: total specimen reared; P%: percentage of parasitism.

Localities	WGS84 (GPS) coordinates		Sampling date	TMC	TPM	TSE	P%
	Lat. (N)	Long. (E)					
Ásotthalom	46°11'26.90"	19°46'25.00"	07.10.2013	147	21	25	14.3
			07.15.2014	238	73	99	30.7
Gödöll	47°36'06.70"	19°24'17.91"	07.27.2012	3	0	0	0
Gyöngyösoroszi	47°50'40.45"	19°54'03.54"	08.13.2012	5	0	0	0
Hort	47°40'52.51"	19°45'20.60"	09.08.2013	4	3	3	75
Jánd	48°06'56.28"	22°21'24.69"	06.19.2013	192	44	256	22.9
Kisk rös	46°35'12.72"	19°15'18.75"	07.08.2013	17	9	19	52.9
Kiskunhalas	46°22'13.47"	19°34'03.04"	07.13.2011	45	17	39	37.8
			07.15.2014	72	12	11	16.7
Mátrafüred	47°49'31.36"	19°58'28.60"	06.07.2011	39	18	22	46.2
			06.28.2011	47	13	28	27.7
			06.26.2012	108	1	5	0.9
			06.25.2013	56	8	8	14.3
Poroszló	47°39'02.11"	20°40'25.28"	07.11.2013	43	8	16	18.6
Püspökladány	47°20'04.55"	21°05'22.83"	07.11.2013	55	12	19	21.8
Tatabánya	47°31'48.96"	18°25'42.11"	10.19.2013	55	4	7	7.3
Törtel	47°05'15.56"	19°53'24.33"	07.08.2013	28	14	17	50
<b>TOTAL</b>				<b>1,154</b>	<b>257</b>	<b>574</b>	<b>22.3</b>

The identified parasitoid species of the complex is listed in Table 2.

We compared our species list with two of the most important on-line databases, the Universal Chalcidoidea Database, hosted by the British Natural History Museum (Noyes 2015) and the Global Taxonomic Database of Gracillariidae (Lepidoptera), hosted by the Belgian Biodiversity Platform (De Prins – De Prins 2014) and discovered that 15 species found during this research were not listed in these databases as parasitoids of the *Ph. comparella*.

From the samples collected at Ásotthalom, a total of 124 specimens belonging to 14 species have been identified. The parasitization rate at this place was 24.2% with 93 parasitized samples. The parasitoid complex was dominated by the synovigenic idiobiont solitary *Sympiesis sericeicornis* (Nees, 1834) (43%, n = 93 parasitised samples) and *Zagrammosoma talitzkii* (Boucek, 1961) (12.9%, n=93). Subdominant species were *Pediobius saulius* (Walker, 1839) (8.6%), *Minotetrastichus frontalis* (Nees, 1834) (8.6%), *Closterocerus trifasciatus* (Westwood, 1883) (7.5%) and a species belonging to the family Braconidae (7.5%). *S. sericeicornis* was also dominant at two other locations with small sample sizes (Kisk rös and Törtel) and present in nearly all samples from the 12 sampling locations. *Ageniaspis testaceipes* (Ratzeburg, 1848), was the dominant species of the samples collected at Jánd (n = 192 total samples collected) causing 43.2% (from n = 44 parasitized samples) of the leaf miner mortality.

Table 2. The checklist and the number of parasitoid species reared from *Phyllonorycter comparella* in Hungary. Species marked with "\*" have been recorded for the first time from *P. comparella*.

Parasitoids reared	Ásotthalom	Hort	Jánd	Kisk rös	Kiskunhalas	Mátrafüred	Poroszló	Püspökladány	Tatabánya	Törtel	TOTAL
<b>Encyrtidae</b>											
<i>Ageniaspis testaceipes</i> *	23		230	11				2	4		270
<i>Encyrtidae</i> sp.*					23	24					47
<b>Eulophidae</b>											
<i>Achrysocharoides attilis</i> *						1					1
<i>Achrysocharoides cilla</i> *					1	15					16
<i>Achrysocharoides scaposa</i>				1		3	12	14		4	34
<i>Aprostocetus</i> sp.					1						1
<i>Baryscapus nigroviolaceus</i>	2		2								4
<i>Baryscapus</i> sp.					2	4					6
<i>Chrysocharis</i> sp.	2										2
<i>Chrysocharis laomedon</i>			3								3
<i>Chrysocharis nephereus</i>	1	1			1						3
<i>Chrysocharis pentheus</i> *	1					1					2
<i>Chrysocharis</i> sp. "A"*									1		1
<i>Cirrospilus elegantissimus</i>			4			2					6
<i>Cirrospilus lyncus</i>			1		1		1				3
<i>Closterocerus trifasciatus</i> *	8		5	1	9	1			1		25
<i>Minotetrastichus frontalis</i>	8		2				2				12
<i>Neochrysocharis formosus</i>	5	1									6
<i>Pediobius pyrgo</i> *					2						2
<i>Pediobius saulius</i>	9		2		2	1				1	15
<i>Pnigalio agraulis</i>			2								2
<i>Pnigalio pectinicornis</i>			1								1
<i>Pnigalio soemius</i> *	1										1
<i>Sympiesis acalle</i> *	1										1
<i>Sympiesis dolichogaster</i> *						1					1
<i>Sympiesis sericeicornis</i>	42		3	6	2	9	1	3	1	8	75
<i>Zagrammosoma talitzkii</i> *	13				6					4	23
<i>Zagrammosoma variegatum</i> *						1					1
<b>Braconidae</b>											
<i>Braconidae</i> sp.	8	1	1								10
<b>TOTAL</b>	<b>124</b>	<b>3</b>	<b>256</b>	<b>19</b>	<b>50</b>	<b>63</b>	<b>16</b>	<b>19</b>	<b>7</b>	<b>17</b>	<b>574</b>

From the 12 species that were present in these samples, we identified only one parasitoid species belonging to the Braconidae family. At this location, the sub dominant species were *C. trifasciatus* (11.4%, n=44 parasitized samples) and *Cirrospilus elegantissimus* (9.1%, n = 44 parasitized samples). *Zagrammosoma talitzkii* was dominant in the samples collected at Kiskunhalas, representing 21.4% of the 28 parasitized samples. The species occurred at only 3 locations. The secondary dominant species of the complex was *C. trifasciatus* (17.9%, 28 parasitized samples) and an unidentified species from the Encyrtidae family. *Achrysocharoides cilla* (Walker, 1839) was only dominant in the samples collected at Mátrafüred, being present in 32.5% of the 40 parasitised samples.

The species also occurred in the samples from Kiskunhalas with an insignificant parasitization rate. The secondary dominant species was *S. sericeicornis* (22.5%, n = 40 parasitized samples). *Achrysocharoides scaposa* (Erdős, 1961) was the dominant species at Poroszló (12.5%, n = 8 parasitized samples and 43 total samples collected) and Püspökladány (17.5%, n = 12 parasitized samples and 55 samples collected) but reared from a relatively small sample size. The species has been observed in samples from five different collecting locations.

The samples from Hort and Tatabánya contained few parasitized samples with only 3 parasitoid species emerged with one specimen each. *S. sericeicornis* is a solitary idiobiont species and was the most frequent species with a high capacity for controlling the miners. As with *C. trifasciatus*, it has a wide range of hosts and both species can be also be secondary parasitoids (Askew 1979, Stojankovic and Markovic 2005, Noyes 2015).

A more specialized species with a narrow host range, the koinobiont gregarious *A. testaceipes*, was also abundant, but dominant only at one sample place. According to the Universal Chalcidoidea Database (Noyes 2015) *A. scaposa* has been reared only from *Ph. comparella*. We identified the species in 5 of our samples and it was dominant in 2 (Püspökladány and Poroszló).

The gregarious koinobiont *A. cilla* was identified from only 2 sample locations (Kiskunhalas and Mátrafüred). Despite its low abundance within the samples, at Mátrafüred it seems to be the major controlling agent of its host. *Zagrammosoma variegatum* (Masi, 1907), an idiobiont solitary ectoparasitoid, has never been described from any species living on *Populus* sp (De Prins and De Prins 2014, Noyes 2015). In our research, we only observed the species at Mátrafüred. At this location we identified another rare and semi-specialist species, *Achrysocharoides altilis* (Delucchi, 1954).

In 3.5% of the 257 parasitized mines we found more than one species of coexisting parasitoids. On these rare occasions, both parasitoid species/specimens are able to complete the larval development. Such events/phenomena are termed the coexistence of multiple parasitoids, a phenomenon among parasitoids of various insects that is well known and has been documented for a long time (Askew – Shaw 1979, Amarasekare 2000a, Amarasekare 2000b, Klapwijk – Owent 2011, Borer et al. 2004, Hackett-Jones et al. 2009, László – Tóthmérész 2013). The coexistence of two parasitoids foraging on a single host occurs when the distribution of the parasitoids is sufficiently aggregated (May and Hüssel 1981, Klopfer and Ives 1997), which triggers intra and interspecific competition (MacArthur and Levins 1967). The successful development of the parasitoids on a single host requires the partitioning or sharing of the food resource (Harvey et al. 2013) by partial host consumption (Miller 1982) and/or the separation of the temporal niche (Hackett-Jones et al. 2009, Amarasekare 2000a) In this case, the competition between the parasitoid species is delimited by the earlier mentioned requirements.

In our samples we registered 9 cases of multiple parasitism. In most cases (6 of 9) the parasitoid couple was composed from a koinobiont and an idiobiont parasitoid, where the koinobiont is a primary parasitoid and the idiobiont is a secondary generalist parasitoid. Therefore, we might say that it is likely that the first (primary) encounter could be the

koinobiont endoparasitoid. On one occasion, we registered two idiobiont ectoparasitoids that had completed their stages.

The most surprising finding was the presence of three koinobiont species from a single mine, which is a rather rare event (Askew 1979, Miller 1982).

Next follows a brief overview on the biology of the parasitoid pairs emerged from single mines (abbreviations: En = endoparasitoid; Ec = ectoparasitoid; G = gregarious; S = solitary; K = koinobiont; I = idiobiont. Read as follows: En,G,K = Endoparasitoid, Gregarious, Koinobiont):

- 1 female specimen of *Achrysocharoides attilis* (En,G,K) +  
1 male specimen of *Baryscapus* sp. (Ec,G,I)
- 1 female specimen of *Braconidae* (En,S,K) +  
1 male specimen of *Pnigalio agraulis* (Ec,S,I)
- 2 female specimens of *Achrysocharoides scaposa* (En,G,K) +  
1 female and 1 male specimen of *Minotetrastichus frontalis* (Ec,G,I)
- 1 male specimen of *Baryscapus nigroviolaceus* (En,S,K) +  
1 female specimen of *Minotetrastichus frontalis* (Ec,G,I)
- 1 female specimen of *Chrysocharis nephereus* (En,S,K) +  
1 female specimen of *Pnigalio soemius* (Ec,S,I)
- 1 male specimen of *Pediobius saulius* (En,S,K) +  
1 female specimen of *Zagrammosoma talitzkii* (Ec,S,I)
- 1 female specimen of *Braconidae* (En,S,K) +  
1 female specimen of *Minotetrastichus frontalis* (Ec,G,I)
- 1 female specimen of *Zagrammosoma talitzkii* (Ec,S,I) +  
1 male specimen of *Sympiesis sericeicornis* (Ec,S,I)
- 1 female specimen of *Chrysocharis nephereus* (En,S,K) +  
1 female specimen of *Neochrysocharis formosus* (En,G,K) +  
1 male specimen of *Braconidae* (En,S,K).

The two relevant online databases (Noyes 2015, De prins and De Prins 2014) list only a small number (19) of parasitoid species associated with the *P. comparella*, reflecting a small amount of studies regarding species load of the leaf miners parasitoid complexes. In this work we contribute 13 species of parasitoids as first records for the host *Phyllnorycter comparella*. Overall, we summarize that 36 parasitoid species have been recorded from *Ph. comparella* so far, including 29 species listed in the present publication. The species composition of the parasitoid complexes of sample sites varied greatly. It is acceptable to say that the parasitoid species of the leaf miner presented a habitat-specific occurrence as found by Hawkins (1994) and Gibbson et al. (1997). Common species, with a generally wide host range, being abundant in nearly all places represented the highest number of species of the entire complex (18 species). The endoparasitoid species, like the *A. testasceipes*, had the biggest impact on the leaf miner *P. comparella*.

As found at the sample sites, the *Ph. comparella* leaf miner can sometimes be very abundant (Yefremova 2009, personal observation). Climate change might trigger outbreaks of this species (as seen with other insects), causing heavy damage to its host plant (Kyle et al. 2014).

Nowadays many forests are composed from a low diversity of trees due to economic forest use; this low diversity of trees may help to intensify the outbreaks of many species (Jactel and Brockerhoff 2007). A number of papers show that diverse habitats contain more complex food-webs with a number of natural enemy species (Paine 1966, Polis and Strong 1996). It is always important to study the species composition of both native (Sz cs et al. 2013) and invasive (Sz cs et al. 2014) species with outbreak ability.

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## Sex Ratio Analysis of Some Macrolepidoptera Species Collected by Hungarian Forestry Light Traps

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**Abstract** – We analysed the sex ratio of 32 macrolepidopteran species caught by Hungarian forestry light traps. That the ratio of males and females collected by light trap varies by species has been known for decades; however, the sex ratio found in the natural population is not known. All 32 species were processed separately, but by the same method. Both males and females were counted throughout the whole swarming. We calculated these figures and inspected the difference in the level of significance with the  $\chi^2$  test. For each swarming we calculated the percentage of males and females. We also calculated the values of the variation coefficients, which express the deviations in average percentages.

Males make up the majority of the moths captured in the trap; this result was mirrored by 29 of the 32 species investigated. One of the exceptions was the *Pelosia muscerda* Hfn. where we observed a male to female ratio that was equal. In addition to that *Watsoniana cultraria* Fabr. is the only one species captured by light traps that showed a significant female majority.

Our results confirm that the majority of moths captured in traps are males. However, the proportion of males and females of each species, and even within the same species, tended to differ greatly with each swarming. Yet, it must be noted that these results speak only for those specimens captured by light traps and cannot be related directly to the actual sex ratio of populations living in the natural environment.

**Macrolepidoptera / sex ratio / forestry light-trap / Hungary**

**Kivonat** – A magyar erdészeti fénycsapdák által gy jtött néhány macrolepidoptera faj ivar-arányának elemzése. Évtizedek óta ismert, hogy fénycsapdával gy jtött hímek és n stények aránya fajonként változik. Nem ismert azonban, hogy milyen a nemek aránya a természetes populációban. 32 fénycsapdával gy jtött Macrolepidoptera faj hím és n stény arányát elemeztük. Minden fajt külön-külön dolgoztunk fel, de azonos módszerrel. Mind a hímeket, mind n stényeket megszámloltuk egész rajzás során. Ezeket összeadtuk és az eltérések szignifikancia szintjét  $\chi^2$  próbával ellen rítettük. Kiszámítottuk a variációs koefficiens értékeket is, amelyek az eltéréseket az átlag százalékában fejezi ki.

Megállapítottuk, hogy a vizsgált 32 fajból 29 rajzásban magasabb volt a befogott hímek száma, mint a n stényeké. Kivétel volt a hamvas zuzmószöv (*Pelosia muscerda* Hfn.), amelynél a hím és n stény arány egyenl és egyetlen faj bükkfa sarlósszöv (*Watsoniana cultraria* Fabr.), amelynél a n stények voltak többségben.

Eredményeink szerint a legtöbb faj esetében több a fénycsapdával befogott hím mint n stény, azonban ez az arány az egyes fajoknál, de még ugyanazon faj, más és más rajzásában is nagyon különböz . Ezek az eredmények azonban nem vonatkoznak közvetlenül a természetben él populációk valós hím-n stény arányaira, hanem csak a fénycsapdás fogásokra.

**Macrolepidoptera / hím n stény arány / erdészeti fénycsapda / Magyarország**

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## 1 INTRODUCTION

That the proportion of males and females captured by light trap varies by species has been known for decades; however the sex ratio within the natural population is unknown.

Within the various orders of insects, Williams (1939) studied the sex ratio by species of the specimens caught. Of the 51 species of the Noctuidae family, the females of 2 species were not attracted to light at all. The females of 27 species represented 1–20% of the total number of specimens: this ratio was 20–46% with 16 species: the number of males and females was identical in the case of 3 species and there were only 3 species where more females than males were attracted to light. There is also a behavioural dimorphism: in 2 samples of 37 individuals of ghost moth (*Hepialus humuli* L.) only 22% males flew to light traps even though sweep net samples gave an approximately 50:50 sex-ratio (Williams 1939). According to Mallet (1984), in the case of *Hepialus humuli* L. more females than males are attracted to light, which suggests that the females are the more mobile gender.

Nanu König (1968) examined 13 Lepidoptera families to establish the ratio of males and females attracted by light. The females of the various families represented 0.2–30% of the catch. Schurr (1971) captured mostly male specimens of the vine moth (*Eupoecilia ambiguella* Hbn.) with his trap running on white light (of 510–610 nm). Járfás et al. (1974) used different sources of light to study the sex ratio of the turnip moth (*Agrotis segetum* Den. et Schiff.). Whatever the light source, the proportion of males was between 48–66%. Czencz (1973) holds that the males of the diamondback moth (*Plutella xylostella* L.) are more drawn to light than the females (75–25%). She does not provide any explanation for this, however.

Bürgés Gál (1980), with a 125W mercury light trap, found that with the nut fruit tortrix (*Cydia splendana* Hbn.), males appear 3–4 days earlier than females. The male to female ratio is 1.9:1. This was almost the same as the male to female ratio of the European chestnut weevil (*Curculio elephas* Gyllenhal), which was 1.83:1.

Egyptian Bollworm (*Earias insulana* Boisduval) moths were collected by Yathom (1981) from mercury vapour light traps operating in Israel between 1974 and 1980. He concludes that the sex ratio generally favoured males.

El-Abdullah et al. (1984) report on a mere 10% of the trapped specimens of the Asiatic rice borer (*Cilo suppressalis* Walker) being males. Skuhřavý et al. (1993) caught male specimens of the saddle gall midge (*Haplodiplosis marginata* von Roser) almost exclusively, both with a Minnesota-type and a UV light trap.

Itämies et al. (1986) employed a light trap to examine the flight pattern of grey mountain carpet (*Entephria caesiata* Den. et Schiff.) between 1978 and 1982 in the Finnish forest areas of Lapland. With the exception of 1978, males dominated the catch, although this dominance was not overwhelmingly high.

Some researchers have published statements of general validity. Novák (1974) has published sex index data relating to 96 species of moths trapped. Depending on the ratio of females, he arranged the species examined into five classes and, within certain limits, he regarded his results as constant. Malicky (1974) also believes that the sex ratio of a captured species is the same over many years through the use of the same source of light at different places of observation; in other words, sex ratio is value specific to a given species. Szarukán (1975) claims that the average proportion of female specimens of the dog's tooth (*Lacanobia suasa* Schiff.) is 33%. He found minimal difference between the first and second generations. A study in Egypt (Sadek 2001), found that 8.04% of the female mediterranean brocade (*Spodoptera littoralis* Fabr.) specimens that had been trapped had not been fertilized, while the ratios of those fertilized once or more than once were 37.25% and 54.76%. The sex ratio of the sulphur knapweed moth (*Agapeta zoegana* L.) was equal when examined at daytime, but at night the overwhelming majority of the specimens caught by light trap were male. The

difference may be explained by the dissimilar reaction of the two sexes to light (Story et al. 2001). In Switzerland Cordillot Duelli (1986) noticed that the ratio of females captured by light trap in one generation of European corn-borer (*Ostrinia nubilalis* Hb.) was 47.3%. The above mentioned authors regard sex ratio as a static value specific to a species, despite the fact that the annual differences in sex ratio are valuable results related to population changes and not statistical deviation data without a biological value (Szeke Szarukán 1982). According to Mohai Herczig (1979) and also to Lesznyák et al. (1993), the proportion in percentage of females rises and falls in harmony with the rise and fall of the amount of insects caught in light traps. The latter of the abovementioned authors have found that the ratio of females is affected first of all by the minimum values of temperature. El-Deeb (1992) established that the number of European corn-borer (*Ostrinia nubilalis* Hbn.) females captured by light trap surpassed that of the males. However, the ratio was affected positively by maximum and minimum temperature, and negatively by relative vapour content.

Some authors have found that both males and females have to be in a certain physiological state to be attracted to sources of light. Terskov Kolomic (1966) report that females of the white satin moth (*Leucoma salicis* L.) can be trapped before egg laying, while those of the gypsy moth (*Lymantria dispar* L.) in Siberia are more prone to be trapped after egg-laying. This latter bit of information is of special interest because there are no data in literature about how gypsy moth females of the European and American populations fly to light considering that the females there are incapable of flight. In contrast to that females of the gypsy moth (*Lymantria dispar* L.) population in East Asia do fly (Wallner et al. 1995; Reineke Zebitz 1998; Charlton et al. 1999). Females of the European corn-borer (*Ostrinia nubilalis* Hbn.), on the other hand, may fly to light any time following their emergence from the pupal state (Showers et al. 1974). Then again, Elliott Dirks (1979) claim they are trappable in the largest numbers 3.2 – 4.4 days after mating.

Sathiyandam Baskaran (1999) observed that the ratio of females of the groundnut leaf miner (*Aproaerema modicella* Deventer) in India changed during different periods in the night. The ratio of mated to unmated females also changed. The number of males and females of some caddis fly (Trichoptera) species caught by light traps in New Zealand displayed a definite change in swarming. Males were the majority at the start of the swarming, while females made up the majority at the end (Ward et al. 1996). Dickler Steuerwald (1997) found significant differences from one year to the next in the number of specimens as well as in the sex ratio of noctuids (Noctuidae) captured by light traps in apple orchards in Germany.

In the study of Myers et al. (1998) the sex ratio of gypsy moth (*Lymantria dispar* L.) pupae varied strikingly between low-density and high-density populations.

Altermatt et al. (2009) experimentally studied the flight to light behaviour of two moth species – the small ermine moth, *Yponomeuta cagnagella* (Hbn.), and the scorched carpet moth, *Ligdia adustata* (Den. et Schiff.). They found that male moths were significantly more (about 1.6 times more frequently) attracted to light than female moths. It was established that there is a sexual dimorphism in the flight to light behaviour of moths.

Garris Snyder (2010) investigated and recorded the sex ratio of 28 southern species in the USA. They tested the well-known view that UV light-trap collections of moths are considerably skewed toward males. Twelve species demonstrated a statistically notable male preponderance, but a wide range of sex ratios was found. Two of the 28 species demonstrated significant bias for both males and females during different observation periods, illustrating the need to collect over the entire flight period. Since the sex ratio of collected organisms varies by species and by time, this knowledge must be taken into consideration when using light trap collection to make population estimates and to gather information for conservation or control of any particular species.

According to Tabadkani et al. (2013) appreciation of the proportion of genders of arthropods is a pertinent issue not only in ecological studies and in biological programs, but also in plant protection. In this study, they continued to examine the factors leading to erroneous estimates of sex ratios of insect species. They examined the predatory gall midge, *Aphidoletes aphidimyza* Rondani (Diptera: Cecidomyiidae), and the results explicitly suggest that direct estimation of the sex ratio in natural populations may be affected by some secondary factors such as differential mortality of sexes, protandry, and differential distribution of males and females over time and/or across habitat.

While they could provide us with important, useful information for everyday practice, there are very few publications reporting on research of this kind. Unfortunately, this kind of research is both time consuming and energy consuming; thus, in the foreseeable future, we cannot expect any major breakthrough in this area.

Researchers have been studying the sex ratio of the insects trapped for decades, but their observations had little practical value for plant protection prognostics. The authors of most studies confine themselves to releasing the figures recording the number of males and females, perhaps even their ratio in terms of percentage of the captured specimens, but they refrain from drawing any conclusions. Admittedly, any such attempt would be a vain endeavour, especially in the case of the data provided by light traps operating for short periods. Whereas if the sex ratio of the populations in the environment shifts in the direction of a preponderance of females and that change is reflected in the catch, a growth in the number of the females trapped might indicate the start of gradation. Therefore, awareness of the regularity of sex ratio transformation may also be used for the purposes of prognosis in the case of species where both sexes fly well to light. Some researchers have published general statements concerning this.

When the sex ratio of an observed species at different observation sites and in different years produces decidedly different values, it would be useful to examine regularities in changes. From the point of view of prognosis, the differences in time have main importance, because they might be related to hypercyclic movement. Our own research revealed a rise of the number of turnip moth (*Agrotis segetum* Den. et Schiff.) females in the years which were followed by gradation (in years 1962 and 1968) (Nowinszky Kiss 1981). It is assumed that the proportion of females affects the number of individuals of future generations. In the year prior to the years of gradation, the number of females increased.

We examined the changes of the number of the females in the turnip moth (*Agrotis segetum* Den. et Schiff.) population between 1957 and 1990. The light-trap catches of 65 observing stations were used (Kiss et al., 2003).

It is concluded that over 99 specimens in each generation the ratio of females is 0.38 from all individual number. If the number of individuals is between 5 and 99, the proportion is 0.44. The proportion of females is 0.46 if the number is between 100 and 499; it is 0.38 between 500 and 999. Between 100 and 999 this value is 0.40. For all observing stations, we calculated the female individual proportions and the 95% confidence intervals for them. We established that higher female proportion belongs to the lower individual numbers, but this proportion is close to the feature if individual numbers are high. It was observed that in the year before gradations begin, the rate of females was extremely high in many cases.

## 2 MATERIAL

The development of a light trap network began in 1952 in Hungary. The traps were used in research institutes, for plant protection, and for forestry purposes. The three-type light trap network is still working and works with uniformly Jermy-type traps. Over the past decades,

the national light-trap network has provided an enormous and inestimable amount of scientific insect material for entomological research and plant protection practice (Nowinszky 2003).

The Jermy-type light trap (Jermy 1961) consists of a frame, a truss, a cover, a light source, a funnel, and a killing device. All the components are painted black, except for the funnel, which is white. A metal ring holding the funnel and a zinc-plated tin joins the steel frame. The cover is 100 cm in diameter. The distance between the lower edge of the cover and the higher edge of the funnel is 20–30 cm. The light source is a 100W normal electric bulb with a colour temperature of 2900°K. The lamp is in the middle of the trussing, 200 cm above ground. The upper diameter of the funnel is 32 cm, while the lower one is 5 cm, and its height is 25 cm. In each case chloroform was used as a killing agent.

The forestry light traps are operational from 6 p.m. (UT) to 4 a.m. every night of the year, regardless of weather, or the time of sunrise and sunset. The operation is suspended only on days when the temperature is below 0 C° and the ground is covered by an unbroken layer of snow. All the insects trapped during the course of a night go into the same collecting jar and so a single set of data will represent the nightly catch result at the given observation site.

In this study we used the catch data of the Hungarian Forestry light trap network of the Forest Research Institute. The light traps were operating in 16 light trap stations across the whole territory of Hungary. The light trap stations, geographic coordinates and years of operation are presented in *Table 1*.

*Table 1. The light trap stations, geographic coordinates and years of operation*

<i>Light-trap stations</i>	<i>Geographic coordinates</i>	<i>Years of operation</i>
Budakeszi	47°30 83 N 18°56 03 E	1962–1970
Erd smecske	46°10 51 N 18°30 80 E	1970
Fels tárkány	47°58 44 N 20°25 07 E	1961–1970
Gerla	46°42 01 N 21°11 07 E	1962–1970
Gyulaj	46°30 51 N 18°17 76 E	1970
Makkoshotyka	48°21 52 N 21°31 17 E	1961–1970
Mátraháza	47°46 87 N 19°55 69 E	1961–1970
Répáshuta	48°02 90 N 20°31 70 E	1962–1970
Sopron	47°41 01 N 16°34 79 E	1962–1970
Szakonyfalu	46°55 45 N 16°13 71 E	1967–1970
Szentpéterföldre	46°37 02 N 16°45 64 E	1968–1970
Szombathely	47°14 01 N 16°37 22 E	1962–1970
Tolna	46°25 60 N 18°46 95 E	1962–1970
Tompa	46°12 28 N 19°38 08 E	1962–1970
Várgesztes	47°28 52 N 18°23 91 E	1962–1970
Zalaerd d	47°03 44 N 17°03 30 E	1970

For our study, 32 forest phytophagous Macrolepidoptera species were selected from the national forestry light trap network material dating back to the years between 1961 and 1970.

The species were selected based on data available from several light traps over many years.

Table 2. Catching data of caught species

Families and species	Light-traps	Years	Number of		
					<sup>2</sup> P<
<b>Lasiocampidae</b>					
December Moth <i>Poecilocampa populi</i> (Linnaeus, 1758)	2	9	2,817	317	0.01
Autumn Eggar <i>Eurigaster rimicola</i> (Denis et Schiffermüller, 1775)	1	9	2,027	40	0.01
Barred Hook-tip <i>Watsonalla cultraria</i> (Fabricius, 1775)	3	5	818	1,484	0.01
Scarce Hook-tip <i>Sabra harpagula</i> (Esper, 1786)	2	5	888	134	0.01
<b>Thyatiridae</b>					
Popular Lutestring <i>Tethea or</i> (Denis et Schiffermüller, 1775)	3	8	2,353	929	0.01
<b>Geometridae</b>					
Maiden's Blush <i>Cyclophora punctaria</i> (Linnaeus, 1758)	1	4	1,197	1,331	0.01
Clay Triple-lines <i>Cyclophora linearia</i> (Hübner, 1799)	4	7	4,005	4,864	0.01
November Moth <i>Epirrita dilutata</i> (Denis et Schiffermüller, 1775)	3	2	1,216	114	0.01
Dingy Shell <i>Euchoeca nebulata</i> (Scopoli, 1763)	8	8	2,656	427	0.01
Sharp-angled Peacock <i>Macaria alternata</i> (Denis et Schiffermüller, 1775)	5	9	3,018	1,990	0.01
Featheres Thorn <i>Colotois pennaria</i> (Linnaeus, 1761)	9	7	4,561	567	0.01
Peppered Moth <i>Biston betularia</i> (Linnaeus, 1758)	5	8	4,359	34	0.01
Pale Oak Beauty <i>Hypomecis punctinalis</i> (Scopoli, 1763)	9	9	12,715	2,938	0.01
The Engrailed <i>Ectropis bistortata</i> (Goeze, 1781)	12	9	17,815	1,517	0.01
<b>Notodontidae</b>					
Buff-tip <i>Phalera bucephala</i> (Linnaeus, 1758)	3	4	495	7	0.01
Plumed Prominent <i>Ptilophora plumigera</i> (Denis et Schiffermüller, 1775)	1	4	2,118	275	0.01
Small Chocolate-tip <i>Clostera pigra</i> (Hufnagel, 1766)	1	10	886	12	0.01
Chocolate-tip <i>Clostera curtula</i> (Linnaeus, 1758)	1	10	699	7	0.01

Table 2. Catching data of caught species (continuation)

Families and species	Light-traps	Years	Number of		
					<sup>2</sup> P<
<b>Lymantriidae</b>					
Pale Tussock <i>Calliteara pudibunda</i> (Linnaeus, 1758)	4	7	1,411	72	0.01
Yellow-tail <i>Euproctis similis</i> (Fuessly, 1775)	2	9	540	38	0.01
White Satin Moth <i>Leucoma salicis</i> (Linnaeus, 1758)	1	5	157	18	0.01
<b>Arctiidae</b>					
Dotted Footman <i>Pelosia muscerda</i> (Hufnagel, 1766)	3	10	1,791	1,762	NS
Scarce Footman <i>Eilema complana</i> (Linnaeus, 1758)	6	10	8,244	6,156	0.01
Common Footman <i>Eilema lurideola</i> (Zincken, 1817)	5	9	15,980	12,252	0.01
<b>Noctuidae</b>					
Small Quaker <i>Orthosia cruda</i> (Denis et Schiffermüller 1775)	5	7	6,820	3,201	0.01
Hebrew Character <i>Orthosia gothica</i> (Linnaeus, 1758)	8	9	4,565	727	0.01
The Satellite <i>Eupsilia transversa</i> (Hufnagel, 1766)	7	10	2,974	2,911	NS
The Chestnut <i>Conistra vaccinii</i> (Linnaeus, 1761)	11	10	11,479	5,428	0.01
Pale-lemon Sallow <i>Xanthia ocellaris</i> (Borkhausen, 1792)	3	8	542	886	0.01
The Dun-bar <i>Cosmia trapezina</i> (Linnaeus, 1758)	6	8	2,097	1,483	0.01
Lesser Belle <i>Colobochyla salicalis</i> (Denis et Schiffermüller, 1775)	2	9	2,056	614	0.01
Jubilee Fan-foot <i>Zanclognatha lunalis</i> (Scopoli, 1763)	4	10	12,077	8,234	0.01

The flying period and primary food plants of caught moths are shown in Table 3.

### 3 METHODS

All species were processed separately, but by the same method. The number of captured males and females was counted for the entire swarming. These were summarized and the difference in level of significance was calculated with <sup>2</sup> test.

For each swarming we calculated the percentage of males and females. We also calculated the values of the coefficients of variation, which express the deviations in average percentages.

Table 3. The flying period and primary food plants of caught moths

<i>Families and species</i>	<i>Flying period of moths</i>	<i>Primary food plants</i>
<b>Lasiocampidae</b>		
<i>P. populi</i> L.	October-November	Quercus, Betula, Populus, Tilia
<i>E. rimicola</i> Den. et Schiff.	September-October	Quercus
<i>W. cultraria</i> Fabr.	May-June; July-August	Fagus
<i>S. harpagula</i> Esp.	May-June; July-August	Tilia, Betula, Quercus
<b>Thyatiridae</b>		
<i>T. or</i> Den. et Schiff.	April-May; August	Populus
<b>Geometridae</b>		
<i>C. punctaria</i> L.	April-May; July-August	Quercus, Betula
<i>C. linearia</i> Hbn.	May-June; July-August	Fagus, Quercus, Betula
<i>E. dilutata</i> Den. et Schiff.	September-November	Quercus, Acer, Betula, Ulmus
<i>E. nebulata</i> Scop.	June-August	Alnus Quercus, Acer, Betula
<i>M. alternata</i> Den. et Schiff.	April-May; July-August	Salix, Alnus, Prunus
<i>C. pennaria</i> L.	September-November	Carpinus, Quercus, Tilia, Salix
<i>B. betularia</i> L.	May-June; July-August	Betula, Ulmus, Salix, Fraxinus
<i>H. punctinalis</i> Scop.	May-July	Quercus, Betula
<i>E. bistortata</i> Gze.	April-May	Polyphagous (Acer, Alnus)
<b>Notodontidae</b>		
<i>Ph. bucephala</i> L.	May-June ; July-August	Quercus, Tilia, Salix
<i>P. plumigera</i> Den. et Schiff.	October-December	Acer, Fagus, Prunus
<i>C. pigra</i> Hfn.	April-June; July-August	Salix
<i>C. curtula</i> L.	April-May; June-August	Populus, Salix
<b>Lymantriidae</b>		
<i>C. pudibunda</i> L.	May-June	Fagus, Carpinus, Ulmus, Tilia,
<i>E. similis</i> Fuesl.	June-July	Quercus-, Ulmus-, Tilia, Salix
<i>L. salicis</i> L.	June-July	Populus
<b>Arctiidae</b>		
<i>P. muscerda</i> Hfn.	July-August	Lichenes
<i>E. complana</i> L.	July-August	Lichenes
<i>E. lurideola</i> Znck.	July-August	Lichenes
<b>Noctuidae</b>		
<i>O. cruda</i> Den. et Schiff.	March-April	Quercus, Betula, Acer, Carpinus
<i>O. gothica</i> L.	March-April	Quercus, Tilia, Ulmus, Betula
<i>E. transversa</i> Hfn.	September-April	Quercus, Betula
<i>C. vaccinii</i> L.	September-April	Quercus, Betula
<i>X. ocellaris</i> Brkh.	September-April	Quercus, Tilia, Acer, Ulmus,
<i>C. trapezina</i> L.	June-September	Quercus, Fagus, Betula, Acer
<i>C. salicalis</i> Den. et Schiff.	May-August	Populus, Salix
<i>Z. lunalis</i> Scop.	May-September	Fagus

#### 4 RESULTS AND DISCUSSION

The results are shown in *Table 4*.

*Table 4. The percentage of males and females, deviations and coefficients of variation.*

Families and species	Moths	Mean %	s	CV	Mean %	s	CV
<b>Lasiocampidae</b>							
<i>Poecilocampa populi</i> L.	2,134	0.83	0.177	0.21	0.17	0.177	1.04
<i>Eriogaster rimicola</i> Den. et Schiff.	2,067	0.98	0.018	0.02	0.02	0.018	0.90
<b>Drepanidae</b>							
<i>Watsonalla cultraria</i> Fabr.	2,308	0.37	0.111	0.30	0.63	0.106	0.17
<i>Sabra harpagula</i> Esp.	1,022	0.88	0.042	0.05	0.12	0.042	0.37
<b>Thyatiridae</b>							
<i>Tethea or</i> Den. et Schiff.	3,292	0.75	0.092	0.12	0.25	0.106	0.42
<b>Geometridae</b>							
<i>Cyclophora punctaria</i> L.	2,528	0.61	0.179	0.29	0.39	0.179	0.46
<i>Cyclophora linearia</i> Hbn.	8,862	0.48	0.102	0.21	0.52	0.092	0.18
<i>Epirrita dilutata</i> Den. et Schiff.	1,340	0.89	0.040	0.04	0.11	0.034	0.31
<i>Euchoeca nebulata</i> Scop.	3,083	0.86	0.141	0.16	0.12	0.141	1.17
<i>Macaria alternata</i> Den. et Schiff.	5,004	0.75	0.114	0.15	0.25	0.115	0.46
<i>Colotois pennaria</i> L.	5,128	0.91	0.086	0.09	0.09	0.086	0.96
<i>Biston betularia</i> L.	4,393	0.99	0.011	0.01	0.01	0.011	1.10
<i>Hypomecis punctinalis</i> Scop.	15,553	0.88	0.076	0.09	0.12	0.076	0.63
<i>Ectropis bistortata</i> Goeze	19,443	0.97	0.048	0.05	0.03	0.045	1.50
<b>Notodontidae</b>							
<i>Phalera bucephala</i> L.	502	0.98	0.019	0.02	0.02	0.019	0.95
<i>Ptilophora plumigera</i> Den. et Schiff.	2,393	0.90	0.054	0.06	0.10	0.054	0.54
<i>Clostera pigra</i> Hfn.	886	0.99	0.013	0.01	0.01	0.008	0.80
<i>Clostera curtula</i> L.	699	0.99	0.009	0.01	0.01	0.009	0.90
<b>Lymantriidae</b>							
<i>Calliteara pudibunda</i> L.	1,183	0.95	0.039	0.04	0.05	0.039	0.78
<i>Euproctis similis</i> Fuesl.	578	0.94	0.037	0.04	0.06	0.036	0.60
<i>Leucoma salicis</i> L.	175	0.89	0.073	0.08	0.11	0.074	0.67
<b>Arctiidae</b>							
<i>Pelosia muscerda</i> Hfn.	3,553	0.53	0.108	0.20	0.47	0.108	0.23
<i>Eilema complana</i> L.	13,400	0.56	0.129	0.23	0.44	0.129	0.29
<i>Eilema lurideola</i> Zinck.	28,232	0.68	0.174	0.26	0.32	0.174	0.54
<b>Noctuidae</b>							
<i>Orthosia cruda</i> Den. et Schiff.	10,021	0.66	0.105	0.16	0.34	0.105	0.31
<i>Orthosia gothica</i> L.	5,292	0.88	0.071	0.08	0.12	0.071	0.59
<i>Eupsilia transversa</i> Hfn.	5,885	0.51	0.083	0.16	0.49	0.083	0.17
<i>Conistra vaccinii</i> L.	16,907	0.69	0.123	0.18	0.31	0.123	0.40
<i>Xanthia ocellaris</i> Borkh.	1,428	0.39	0.194	0.50	0.61	0.194	0.32
<i>Cosmia trapezina</i> L.	3,580	0.58	0.082	0.14	0.42	0.082	0.19
<i>Colobochoyla salicalis</i> Denis et Schiff.	2,670	0.81	0.085	0.10	0.19	0.085	0.45
<i>Zanclognatha lunalis</i> Scop.	20,311	0.62	0.130	0.21	0.38	0.130	0.34

Notes: Mean % and Mean % = averaged percentage of males and females,  
s and s = deviations, CV and CV = coefficient of variations.

We found the majority of moths collected in light traps are males. This result is true of 29 species from the investigated 32 species. However, the proportion of males and females of each species, and even within the same species, differed greatly during each swarming.

One probable explanation may be the protandry for greater number of males. Cordillot (1989) established that occurrence of the male of the European corn-borer (*Ostrinia nubilalis* Hbn.) in the light trap preceded the females' occurrence by  $3.8 \pm 1.5$  days on average. This phenomenon was named "protandry" by Stockel and Peyelut (1984). That the females of some species are attracted to light in greater number after mating may be the cause of this.

Yathom (1981) found the most frequently light trapped Egyptian bollworm (*Earias insulana* Boisduval) females mated once. Lopez et al. (2000) found that the overwhelming majority of the females of *Mythimna unipuncta* (Haworth) flew to light after egg-laying. We also observed the "protandry" phenomenon in the case of the dotted footman *Pelosia muscerda* Hfn. in Tompa 1970; the male and female ratio is equal with this species. Males and females of both species (*Pelosia muscerda* Hfn. and *Eupsilia transverse* Hfn.), are captured nearly in similar number. Cordillot (1989) found that the overall ratio of European corn-borer (*Ostrinia nubilalis* Hbn.) was found almost the same; of the captured moths, 52.7% were males and 47.3% were females.

*Watsoniana cultraria* Fabr. is the only one species captured by light traps that showed a significant female majority. We examined 8 swarmings; females outnumbered males in 7 of them. The males of this moth also fly in the sunlight and they can usually be seen among the higher branches of beech trees. This may be a reason why fewer males of this species are captured at night.

For decades it has been known that the proportion of male and female individuals of various insect species caught by light trap tends to differ. This fact proves that the ratio of the various species represented the catch are not the same as the ratio that appears in nature (Kiss et al. 2003). The reasons for this fact may be many. Flight is difficult for females because of their increased weight due to developing eggs. The males may be more active as they visit the females with the aim of the mating. It is also possible that the males have a greater affinity to light (Waringer 2003).

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# Macrofungi in the Botanical Garden of the University of West Hungary, Sopron

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**Abstract** – Botanical gardens have diverse habitats and floristic conditions. The aim of this study was to examine whether these specific environmental conditions have a positive impact on the appearance of mushrooms. Between 2011 and 2013, mycological observations were performed in the Botanical Garden of the University of West Hungary, Sopron. A total of 171 mushrooms species were identified. Several rare species and two protected species were found. The identification and classification of the species reveal how botanical gardens provide a special habitat for mushrooms. These features of botanical gardens are beneficial for fungal dissemination and preservation.

**botanical garden/special habitats/fruit-body monitoring/ex situ conservation**

**Kivonat** – Nagygombák a Nyugat-magyarországi Egyetem Soproni Botanikus Kertjében.

A botanikus kertekben, sajátosságaikból adódóan igen változatos term helyi és florisztikai viszonyok vannak. Tanulmányunk célja vizsgálni, hogy ezek a speciális környezeti feltételek milyen kedvező hatással vannak a nagygombák megjelenésére. A 2011-13 években mikológiai megfigyeléseket végeztünk a Nyugat-magyarországi Egyetem soproni botanikus kertjében. Vizsgálataink során összesen 171 nagygomba fajt sikerült kimutatnunk, melyek között számos ritka és két Magyarországon védett faj is el került. A fajok meghatározása és csoportosítása rávilágított arra, hogy milyen speciális élőhelyet nyújtanak a botanikus kertek a nagygombáknak. A botanikus kert adottságai lehetőséget nyújtanak a gombák széleskörű megismerésére és megőrzésére.

**botanikus kert/speciális élőhely/term test monitoring/ex situ megőrzés**

## 1 INTRODUCTION

The history of European botanical gardens dates back to the Middle Ages. They evolved from early herbalist gardens and were already widespread in the 16th century. Their focus shifted to educational and scientific purposes in the 18th century and reached their present form and role in the 18th-19th centuries (Hill 1915). The main objectives of botanical gardens today are to collect, preserve, and exhibit plants native to a given location as well as more colorful or rare non-native species (Brickell 1991, Dongyan – Zuoshuang 2008). These types of gardens have become increasingly important today. Their artificial habitats, diverse flora, and habitat conditions are favorable for mushroom species as well.

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The aim of this study is to illustrate the important role such gardens (e.g. arboretums, botanical gardens, parks) play not only in the preservation of flora, but also in the equally important preservation of mushrooms. We demonstrate this with the example of the Botanical Garden of the University of West Hungary in Sopron.

There are several studies that examine mushrooms occurring in urban environments. Most of these studies focus on the many rare and protected species that appear in urban environments and explore how these urban environments are especially favorable for parasitic and saprotrophic species (Luszczynski 1997, Pál-Fám 2001, Pál-Fám – Boros 2006). The mycological importance of urban parks and gardens emphasizes the differences that exist among artificially created environments and disturbed environments. These green urban areas are an important habitat for mushrooms (Lisiewska – Strakulska 2002, Kasprowicz et al 2011, Skorupski et al 2011, Bujakiewicz – Kujawa 2000). For example, during a five-year study of the People's Garden of Miskolc, a total of 124 species were identified. Of these, only 33 species were mycorrhizal, but several rare and protected species were also found (Kaposvári 2013). Fungi observation in the botanical garden of Soroksár began 40 years ago (Konecni et al. 1973); since then 274 fungi species have been identified, including a number of rare and protected species (Rimóczy 1993, 1998). A list of 58 species of macromycetes was recorded in the Warsaw Botanical Garden between 1961 and 1963 and is discussed in a paper by Szober (1965). The Central Botanical Garden of Belarus in Minsk recorded 12 species of macromycetes, which is the topic of a paper by Dischuk (2001). In one of his studies, Szczepkowski (2007) identified 79 species of macromycetes in the Dendrological Park of the Warsaw Agricultural University in Warsaw between 1996 and 2005. Arboretums also favor the diversity of the lichen fungi species (Ladd et al 2009). A more preferred topic within the study of fungi in urban environments is the examination of mushrooms nurtured in glasshouses and greenhouses where tropical species can often be found (Pidlich-Aigner et al. 2002, Gubitz 2012, Lukács et al. 2010, 2011). Szczepkowski et al. (2014) reported a total of 206 species that existed in greenhouses in five large European cities. Arboretums also favor the diversity of lichen fungi species (Ladd et al. 2009).

## 2 MATERIALS AND METHODS

### 2.1 Description of the study area

The Botanical Garden of the University of West Hungary is located in Sopron near the north-western border of Hungary and Austria. Geographically it lies in the eastern foothills of the Sopron hills. From a geobotanical point of view, it is considered a border of the floristic region of the Eastern Alps (Alpicum, subdivision "Ceticum") and the Pannonian basin (Pannonicum, subdivision "Castriferreicum"). Afforestation of the area the Botanical Garden of Sopron occupies today began in 1897; before that, it was home to oak forests, orchards, and kitchen gardens. The development of the Botanical Garden began after 1923 and evolved on 17.2 hectares; the area was primarily used for teaching and practicing forestry. Later, new plantations in plant-geographical clusters were created. Thus, groups of East Asian and North American woody flora evolved (Nemky – Vancsura 1970, Kocsó 1996, 2008). Typically the garden contains a variety of temperate zone plants in its collection and international seed exchanges are largely responsible for their expansion (Kocsó, 1996). Two of the most important missions of the Botanical Garden are to collect a great many and a great variety of taxa, and also to conserve many species (also protected and not threatened) in situ and ex situ. In 2007 the garden contained 2691 woody plant taxa, and 3691 herbaceous taxa and, due to the varied plant cover, there were 57 moss species as well (Kocsó 2008, Szücs 2008). Among the plant taxa were 16 in situ protected species and 135 ex situ protected species (Kocsó 2008).

The soils of the Botanical Garden were developed on loess, marl and marl-like sediments, and they belong to the reference soil groups of Cambisols and Luvisols. The soil texture is loam, clayey-loam, and the structure is well-formed subangular and angular blocky. The nutrient and water supply statuses of the upper soil horizons are good for weak acidic soil pH. Rain water flows here from the eastern foothills of the Sopron hills providing the area with good water conditions. Due to its geographical location, the garden has a sub-Alpine climate. The average annual rainfall is 690 mm; the 12-hour humidity in July is about 56%, which is a good climate for hornbeam and oak (Cserpes – Kocsó 1996).

## 2.2 Macrofungi investigation

The mycological observations in the Botanical Garden began in 2004, but our field surveys were completed between 2011 and 2013. The number of surveys varied from six to eight times per year and were weather dependent: there were 1-2 surveys in the spring and 3-4 in the summer and autumn. As the university is located in the garden, informal observations were made frequently.

*Table 1.: Date of macrofungi observation*

Years	Spring	Summer	Autumn
2011	22.05.	05.06.;	25.09.; 04.10., 21.10.; 07.11.
2012	25.05.	05.06.; 14.06.; 17.07.; 24-27.07.;	20.09.; 28.09.; 01-05.10.; 17-19.10.; 24.10.; 08.11.
2013	07.05.; 17.05.;	05-07.06.; 25.06.; 26.08.;	05-06.09.; 01-12.09.; 18-19.09.; 01-03.10.; 08-10.10.; 16-18.10.

Observations of the mushrooms were regularly carried out by exploring the entire Botanical Garden utilizing the garden paths after major rainfalls (*Figure 1.*). The Botanical Garden has an extremely diverse flora which made it impossible to prepare individual sampling areas.

During the surveys, the number of locations where fruiting bodies of different species were found was recorded (*Table 2.*). If this occurred in the same location over several years, it is not calculated as a different position. The mushrooms were identified in the field, photo documented and, in some cases, fungarium were also completed. The species were identified by their macroscopical and ecological specifics, as well as their microscopical specifics (cystidium and spore measurements). The spore and cystidia observations were made using a Nikon H600L and a Zeiss Axio Imager light microscope at 600× and 1000× magnification. Images were analyzed with Image Pro Plus 7.0 software. If needed, chemicals reagents were used to identify the fungus (e.g. 10% KOH, Meltzer-reagent, or by the cystidium observation kongo-red dye stuff was used). The following literature was used during the identification: Kundes – Vesterholt (2012), Bohus et al. (1951), Igmándy (1991), Galli (1996), Krigelsteiner (2000 a, b.), Aronsen (2012), Assoyov – Miksik (2013), Froslev – Stjernegaard (2013), Tulloss – Yang (2014), Bandini (2014).

Nomenclature followed the Mycobank method (Robert et al. 2014). The species found are listed in *Table 2* with the name of the taxon and the frequency index (rA = relative abundance, from 1 to 5; 1 = very rare, 5 = very common) [how frequently the species was observed during survey time, how abundantly it was found], number of location (Lo). In summary, the Lo value points to a spatial frequency, and the value of rA is the temporal frequency of the appearances.

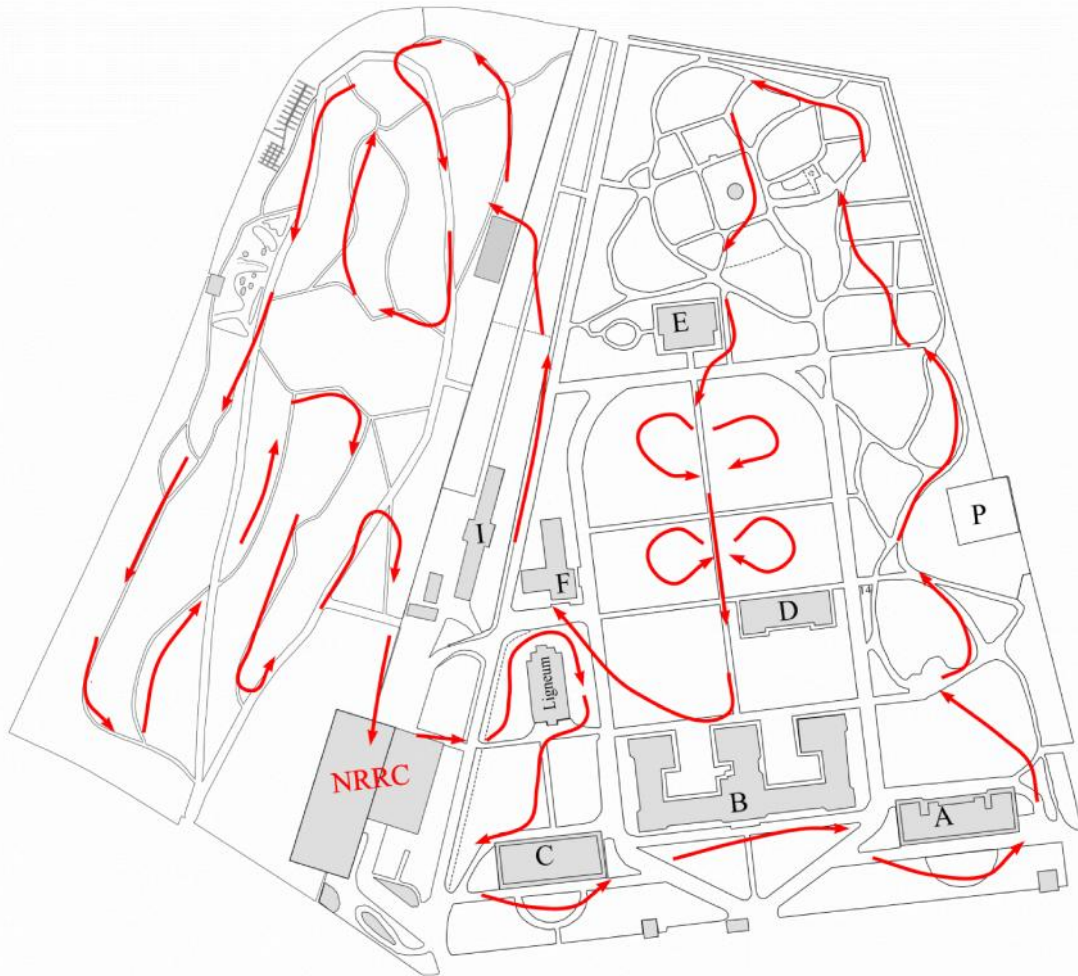


Figure 1: Map of the botanical garden paths used during the observations

The species found were classified on the basis of Arnolds et al. (1995) and our observations (Ls; m = mycorrhizal, p = parasite (also necrotorf and biotrof), sl = saprotrophic lignicole, st = saprotrophic terricole, sc = saprotrophic on crop residues). In questionable cases (e.g. *Clitopilus prunulus*) the work of Rinaldi et al. (2008) was consulted. A conservation assessment was also made based on the works of Rimóczi et al. (1999) and Siller et al. (2005, 2006), taking into account the changes in Hungarian law (Magyar Közlöny 2013). The conservation assessment is in the rightmost column (P) (0 = Extinct 1 = Critically Endangered; 2 = Endangered; 3 = Vulnerable; 4 = Lower Risk; (Rimóczi et al. 1999). The two species highlighted with bold letters are protected by law in Hungary (Magyar Közlöny 2013)

### 3 RESULTS, CONCLUSIONS

Previously Kocsó (2008) mentioned the existence of 75 species in the Botanical Garden, but he does not name them. In the period between 2011 and 2013, 171 mushroom taxa were identified in the Botanical Garden of the University of West Hungary, Sopron (see Table 2 in Appendix).

Categorized by life form, 33% of the species are mycorrhizal, 5% are parasitical, 22% are saprotrophic lignicole, and 40% are saprotrophic terricole or grow on plant debris. The overall species composition does not differ notably from the composition found in the Sopron hills (Folcz et al. 2013). Among the species are calciphilous and acidophilous species as well.

When the set of species is compared with data from the Sopron region, we see that the identified species partially overlap in the Dudlesz forest (which has more calcareous substrates) (Frank 1997) and also with the fungi in the Sopron mountains (which is more acidic) (Folcz et al. 2013). In other words, a sorting of blending area exists between the two mushroom worlds which is in line with the growing conditions of the landscape site. Among the species, there are a total of 15 that are yet unpublished for the Sopron area: *Amanita lividopallescens* (Secr. ex Boud.) Kühner & Romagn., *Bolbitius titubans* (Bull.) Fr., *Clitocybe costata* Kühner & Romagn., *Galerina laevis* Singer, *Hygrocybe subglobispora* f. *aurantiorubra* Arnolds, *Hygrophorus mesotephros* Berk. & Broome, *Hohenbuehelia petaloides* (Bull.) Schulzer, *Hemimycena cucullata* (Pers.) Singer s.l., *Lactarius sanguifluus* (Paulet) Fr., *Leucoagaricus americanus* (Peck) Vellinga, *Macrotyphula fistulosa* (Holmsk.) R.H. Petersen, *Melanogaster variegatus* (Vittad.) Tul. & C. Tul., *Pluteus ephebeus* (Fr.) Gillet, *Stropharia coronilla* (Bull. ex DC.) Quéf., *Tephrocybe rancida* (Fr.) Donk. Two protected: *Agaricus bohusii* Bon, *Polyporus tuberaster* (Jacq. ex Pers.) Fr, ten endangered species, and many rare species occur among the species. Of the species, 42% (71 species) are on the recommended red list of Hungarian fungi, thus the Botanical Garden plays a significant role in the protection of fungi.

#### 4 DISCUSSION

The diversity of vegetation and the special care and treatment inherent to botanical gardens help create exceptional conditions which are highly favorable to the colonization of fungi. The artificial and disturbed soil conditions are favorable for a variety of terrestrial fungi and the diverse flora have a positive effect on mycorrhizal colonization. The garden is cultivated in such a manner that dead wood and wood debris are allowed to accumulate on the forest floor, so the rate of the lignicole mushrooms is similar to that found in natural forests which are also rich in dead wood. Many of the species appear regularly, others appear rarely, which can be traced back to meteorological conditions during the period. Hawksworth (1991) called attention the importance of international protection of fungi. He underlined the importance of both ex-situ and in-situ methods. Moore et al. (2001) recently highlighted the importance of in situ fungi conservation which stresses habitat protection as a key step in fungi protection. The safeguarding of mushrooms in Italy can also be highlighted as an example. One of the cornerstones of this habitat preservation (Venturelli et al. 2011), is the defense of the arbuscular mycorrhizal fungi (Turrini – Giovanetti 2012). In the case of botanical gardens, exsitu mushroom protection is also possible because the gardens are artificial habitats. The rare and protected species that were found show that botanical gardens may be suitable for this task of habitat support if we keep in mind the ex situ (or sometimes the in situ) conservation of the forming and sustaining fungi. Based on our results, botanical gardens are very important habitats for fungi and this provides many opportunities both for science and for active fungi protection (Szober 1965, Pál-Fám et al 2004, Szczepowski 2007). The (ex situ) conservation in such places creates opportunities for mycological research and educational activities and for mycological biodiversity preservation (Hu – Zhang 2008, Varese et al. 2011).

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APPENDIX

Table 2. The observed macrofungi species in the botanical garden of the University of West Hungary, Sopron

Nr.	Species	rA	Lo	Ls	P
Ascomycota					
1	<i>Ascocoryne sarcoides</i> (Jacq.) J.W. Groves & D.E.	1	1	sl	3
3	<i>Helvella crispa</i> (Scop.) Fr.	4	6	st	3
4	<i>Helvella elastica</i> Bull.	1	2	st	3
5	<i>Humaria hemisphaerica</i> (F.H. Wigg.) Fuckel	1	2	st	4
6	<i>Kretzschmaria deusta</i> (Hoffm.) P.M.D. Martin	1	1	p	–
7	<i>Tarzetta cupularis</i> (L.) Svr ek	1	1	st	4
8	<i>Morchella esculenta</i> (L.) Pers.	1	1	st	3
9	<i>Peziza varia</i> (Hedw.) Alb. & Schwein.	3	2	sl	4
10	<i>Sarcoscypha austriaca</i> (Beck ex Sacc.) Boud.	2	3	sl	4
11	<i>Xylaria hypoxylon</i> (L.) Grev.	1	2	sl	–
12	<i>Xylaria polymorpha</i> (Pers.) Grev	1	3	sl	–
Bazidiomycota					
13	<i>Agaricus augustus</i> Fr.	1	2	st	2
14	<i>Agaricus bitorquis</i> (Quél.) Sacc.	2	1	st	–
15	<b><i>Agaricus bohusii</i> Bon</b>	1	1	st	2
16	<i>Agaricus campestris</i> L.	1	1	st	–
17	<i>Agaricus sylvaticus</i> Schaeff.	2	3	st	–
18	<i>Agaricus xanthodermus</i> Genev.	5	10	st	–
19	<i>Agrocybe erebia</i> (Fr.) Kühn.	1	1	st	2
20	<i>Agrocybe praecox</i> (Pers.) Fayod	2	2	st	–
21	<i>Amanita battarrae</i> (Boud.) Bon	1	1	m	3
22	<i>Amanita citrina</i> (Schaeff.) Pers.	1	1	m	3
23	<i>Amanita lividopallescens</i> (Secr. ex Boud.) Kühner & Romagn.	2	1	m	2
24	<i>Amanita muscaria</i> (L.) Lam.	2	2	m	3
25	<i>Amanita phalloides</i> var. <i>phalloides</i> (Vaill. ex Fr.) Link	3	3	m	–
26	<i>Amanita rubescens</i> Pers.	1	1	m	–
27	<i>Amanita solitaria</i> (Bull.) Mérat	2	3	m	2
28	<i>Amanita strobiliformis</i> (Paul.: Vitt.) Bertil.	2	4	m	3
29	<i>Armillaria mellea</i> (Vahl.) P. Kumm.	1	1	p	–
30	<i>Armillaria ostoyae</i> (Romagn.) Herink	1	2	p	–
31	<i>Armillaria tabescens</i> (Vahl.) P. Kumm.	2	1	p	–
32	<i>Auricularia auricula-judae</i> (Bull.) Quél.	1	1	sl	–
33	<i>Auriscalpium vulgare</i> Gray	3	2	sl	–
34	<i>Bolbitius titubans</i> (Bull.) Fr.	3	5	sc	3
35	<i>Boletus impolitus</i> Fr.	1	1	m	4
36	<i>Boletus luridus</i> Sowerby	2	3	m	4
37	<i>Boletus radicans</i> Pers.	3	2	m	3
38	<i>Calocybe gambosa</i> (Fr.) Donk	1	1	st	–
39	<i>Chlorophyllum olivieri</i> (Barla) Vellinga	3	7	st	–
40	<i>Chroogomphus rutilus</i> (Schaeff.) O.K. Mill.	2	2	m	–
41	<i>Clitocybe costata</i> Kühner & Romagn	1	1	st	3
42	<i>Clitocybe gibba</i> (Pers.) P. Kumm.	2	2	st	3

Nr.	Species	rA	Lo	Ls	P
43	<i>Clitocybe nebularis</i> (Batsch) P. Kumm.	1	2	st	3
44	<i>Clitocybe odora</i> (Bull.) P. Kumm.	1	1	st	3
45	<i>Clitocybe rivulosa</i> (Pers.) P. Kumm.	1	2	st	3
46	<i>Clitocybula platyphylla</i> (Pers.) Malençon & Bertault	1	2	st	–
47	<i>Clitopilus prunulus</i> (Scop.) P. Kumm.	2	4	st	–
48	<i>Coprinellus disseminatus</i> (Pers.) J. E. Lange	4	7	sl	–
49	<i>Coprinellus micaceus</i> (Bull.) Vilgalys, Hopple & Jacq. Johnson	5	6	sl	–
50	<i>Coprinopsis atramentaria</i> (Bull.) Redhead, Vilgalys & Moncalvo	1	1	sl	–
51	<i>Coprinus comatus</i> (O. F. Müll.) Pers.	4	5	st	–
52	<i>Coprinus leiocephalus</i> P.D. Orton	1	1	st	–
53	<i>Coprinus cinereus</i> S.F. Gray	1	1	sc	–
54	<i>Cortinarius infractus</i> Berk. s. l.	1	1	m	–
55	<i>Cortinarius largus</i> Fr.	1	1	m	3
56	<i>Cortinarius subpurpurascens</i> (Batsch) Fr.	1	2	m	3
57	<i>Cortinarius trivialis</i> J. E. Lange s. l.	2	1	m	–
58	<i>Crepidotus mollis</i> (Schaeff.) Staude	1	1	sl	–
59	<i>Cyathus olla</i> (Batsch) Pers.	2	3	sl	–
60	<i>Cyathus striatus</i> (Huds.) Willd.	3	4	sl	–
61	<i>Dacrymyces chrysospermus</i> Berk. & M.A. Curtis	1	2	sl	–
62	<i>Daedalea quercina</i> (L.) Pers.	1	1	sl	–
63	<i>Entoloma rhodopolium</i> (Fr.) P. Kumm.	1	1	m	–
64	<i>Exidia nigricans</i> (With.) P. Roberts	1	2	sl	–
65	<i>Flammulina velutipes</i> (Curtis) Singer	1	2	p	–
66	<i>Fomitopsis pinicola</i> (Sw.) P. Karst	1	1	p	–
67	<i>Galerina laevis</i> (Pers.) Singer	1	3	st	3
68	<i>Geastrum fimbriatum</i> Fr.	1	1	st	3
69	<i>Geastrum triplex</i> Jungh.	3	5	st	3
70	<i>Gomphidius glutinosus</i> (Schaeff.) Fr.	2	2	m	–
71	<i>Gymnopus dryophilus</i> (Bull.) Murrill	5	9	st	–
72	<i>Gymnopus fusipes</i> (Bull.) Gray	1	1	sl	–
73	<i>Hebeloma crustuliniforme</i> (Bull.) Quéf.	1	2	m	–
74	<i>Hemimycena cucullata</i> (Pers.) Singer s. l.	2	3	st	2
75	<i>Hemipholiota populnea</i> (Pers.) Bon	1	1	sl	–
76	<i>Hohenbuehelia petaloides</i> (Bull.) Schulzer	1	1	st	3
77	<i>Hygrocybe ceracea</i> (Wulf.) P. Kummer.	1	1	st	3
78	<i>Hygrocybe subglobispora</i> f. <i>aurantiorubra</i> Arnolds	1	1	st	2
79	<i>Hygrocybe virginea</i> (Wulfen) P.D. Orton & Watling	1	1	st	2
80	<i>Hygrophoropsis aurantiaca</i> (Wulfen) Maire	1	2	st	–
81	<i>Hygrophorus hypothejus</i> (Fr.:Fr.)Fr.	1	1	m	3
82	<i>Hygrophorus mesotephrus</i> Berk. & Broome	1	1	m	3
83	<i>Hypholoma fasciculare</i> (Huds.) P. Kumm.	2	5	sl	–
84	<i>Hypholoma lateritium</i> (Schaeff.) P. Kumm.	1	1	sl	–
85	<i>Infundibulicybe geotropa</i> (Bull.) Harmaja	1	1	st	–
86	<i>Inocybe asterospora</i> Quéf. s. l.	1	1	m	3
87	<i>Inocybe flocculosa</i> Sacc. s. l.	1	1	m	3
88	<i>Inocybe geophylla</i> (Bull.) P. Kumm.	1	3	m	3
89	<i>Inocybe rimosa</i> (Bull.) P. Kumm.	1	3	m	–
90	<i>Laccaria amethystina</i> Cooke	1	1	m	3

Nr.	Species	rA	Lo	Ls	P
91	<i>Laccaria bicolor</i> (Maire) P.D. Orton	1	1	m	3
92	<i>Lacrymaria lacrymabunda</i> (Bull.) Pat.	4	7	st	–
93	<i>Lactarius aurantiacus</i> (Pers.) Gray	1	2	m	–
94	<i>Lactarius blennius</i> (Fr.) Fr.	1	1	m	–
95	<i>Lactarius chrysorrheus</i> Fr.	1	2	m	–
96	<i>Lactarius circellatus</i> Fr.	1	1	m	–
97	<i>Lactarius decipiens</i> Quél.	1	1	m	–
98	<i>Lactarius deliciosus</i> (L.) Gray	1	1	m	4
99	<i>Lactarius deterrimus</i> Gröger	1	2	m	–
100	<i>Lactarius quietus</i> (Fr.) Fr.	3	7	m	–
101	<i>Lactarius sanguifluus</i> (Paulet) Fr.	2	1	m	–
102	<i>Lactarius torminosus</i> (Schaeff.) Gray	1	1	m	4
103	<i>Lactarius turpis</i> (Weinm.) Fr.	1	2	m	3
104	<i>Leccinum pseudoscabrum</i> (Kallenb.) Šutara	1	1	m	4
105	<i>Lentinus tigrinus</i> (Bull.) Fr.	1	1	sl	–
106	<i>Lepiota cristata</i> P. Kumm.	2	6	st	–
107	<i>Lepista inversa</i> (Scop.) Pat.	4	5	st	–
108	<i>Lepista luscina</i> (Fr.) Singer	1	1	st	–
109	<i>Lepista nuda</i> (Bull.) Cooke	2	5	st	–
110	<i>Lepista saeva</i> (Fr.) P. D. Orton	1	2	st	–
111	<i>Lepista sordida</i> (Schumach.) Singer	1	2	st	–
112	<i>Leucoagaricus americanus</i> (Peck) Vellinga	1	1	st	2
113	<i>Leucoagaricus leucothites</i> (Vittad.) Wasser	2	3	st	–
114	<i>Lycoperdon perlatum</i> Pers.	1	1	st	–
115	<i>Lycoperdon pratense</i> Pers.	1	1	st	2
116	<i>Lyophyllum decastes</i> (Fr.) Singer	2	4	st	–
117	<i>Macrolepiota mastoidea</i> (Fr.) Singer	1	1	st	–
118	<i>Macrotyphula fistulosa</i> (Holmsk.) R.H. Petersen	1	1	sl	3
119	<i>Marasmius oreades</i> (Bolton) Fr.	1	1	sc	–
120	<i>Melanogaster variegatus</i> (Vittad.) Tul. & C. Tul.	1	2	m	3
121	<i>Melanoleuca melaleuca</i> (Pers.) Murrill	1	1	st	–
122	<i>Mycena galericulata</i> (Scop.) Gray	1	1	st	–
123	<i>Mycena polygramma</i> (Bull.) Gray	1	1	sl	–
124	<i>Mycena pura</i> (Pers.) P. Kumm.	3	7	st	–
125	<i>Mycena rosea</i> Gramberg	1	2	st	–
126	<i>Panellus stipticus</i> (Bull.) P. Karst.	1	1	sl	–
127	<i>Paxillus involutus</i> (Batsch) Fr.	1	2	m	–
128	<i>Pholiota squarrosa</i> (Vahl) P. Kumm.	1	1	sl	3
129	<i>Placodes betulinus</i> (Bull.) Quél.	1	2	p	–
130	<i>Pleurotus ostreatus</i> (Jacq.) P. Kumm.	1	2	p	–
131	<i>Pluteus atromarginatus</i> (Konrad) Kühner	1	1	sl	4
132	<i>Pluteus cervinus</i> (Schaeff.) P. Kumm.	3	8	sl	–
133	<i>Pluteus ephebeus</i> (Fr.) Gillet	1	1	sl	4
134	<i>Polyporus arcularius</i> (Batsch) Fr.	1	1	sl	–
135	<i>Polyporus squamosus</i> (Huds.) Fr.	1	2	p	–
136	<b><i>Polyporus tuberaster</i> (Jacq. ex Pers.) Fr</b>	1	1	sl	3
137	<i>Psathyrella artemisiae</i> (Pass.) Konrad & Maubl. s. l.	1	1	sl	3
138	<i>Psathyrella candolleana</i> (Fr.) Maire	1	1	sl	–

Nr.	Species	rA	Lo	Ls	P
139	<i>Psathyrella marcescibilis</i> (Britzelm.) Singer	3	8	st	3
140	<i>Psathyrella multipedata</i> (Peck) A. H. Sm.	1	1	sl	3
141	<i>Psathyrella piluliformis</i> (Bull.) P. D. Orton	1	2	sl	–
142	<i>Ramaria stricta</i> (Pers.) Quél.	1	2	sl	–
143	<i>Rhodocollybia butyracea</i> (Bull.) Lennox	3	6	st	–
144	<i>Rhodocybe gemina</i> (Paulet) Kuyper & Noordel.	1	2	st	3
145	<i>Russula cyanoxantha</i> (Schaeff.) Fr.	1	2	m	–
146	<i>Russula delica</i> Fr.	1	1	m	–
147	<i>Russula emetica</i> (Schaeff.) Pers. <i>s. l.</i>	1	1	m	3
148	<i>Russula foetens</i> Pers.	2	4	m	–
149	<i>Russula lepida</i> Fr.	1	1	m	3
150	<i>Russula undulata</i> Velen.	1	2	m	3
151	<i>Schizophyllum commune</i> Fr.	4	12	sl	–
152	<i>Scleroderma citrinum</i> Pers.	2	4	m	4
153	<i>Scleroderma verrucosum</i> (Bull.) Pers.	2	3	m	4
154	<i>Stereum hirsutum</i> (Willd.) Pers.	1	2	sl	–
155	<i>Strobilurus stephanocystis</i> (Kühner & Romagn. ex Hora) Singer	1	1	sc	–
156	<i>Stropharia aeruginosa</i> (Curtis) Quél.	3	6	st	–
157	<i>Stropharia coronilla</i> (Bull. ex DC.) Quél.	1	2	st	–
158	<i>Stropharia rugosoannulata</i> Farl. ex Murrill	1	1	st	3
159	<i>Suillus granulatus</i> (L.) Roussel	5	15	m	–
160	<i>Suillus grevillei</i> (Klotzsch) Singer	2	4	m	–
161	<i>Suillus viscidus</i> (L.) Roussel	2	6	m	3
162	<i>Tephrocybe rancida</i> (Fr.) Donk	1	1	st	3
163	<i>Trametes versicolor</i> (L.) Lloyd	2	3	sl	–
164	<i>Tricholoma batschii</i> Gulden	1	4	m	3
165	<i>Tricholoma scalpturatum</i> (Fr.) Quél.	2	2	m	3
166	<i>Tricholoma terreum</i> (Schaeff.) P. Kumm.	3	5	m	–
167	<i>Tubaria furfuracea</i> (Pers.) Gillet	1	2	sl	–
168	<i>Volvopluteus gloiocephalus</i> (DC.) Vizzini, Contu & Justo	1	3	sl	–
169	<i>Xerocomus cisalpinus</i> Simonini, H. Ladurner & Peintner	1	1	m	4
170	<i>Xerocomus porosporus</i> (Imler ex Bon & G. Moreno) Contu	1	1	m	4
171	<i>Xerula radicata</i> (Relhan) Dörfelt.	4	10	sl	–

# On the Occurrence of Natural Norway Spruce Woodland in the Pieniny Mts (Western Carpathians)

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**Abstract** – According to Kulczyński (1928), a natural *Picea abies* forest occurs in the mountain range of the Pieniny Mts on the slopes of the mountain Vysoké skalky. Later, various data on the altitudinal range of this stand (and thus on the locality itself) were published: (1) 890 (900)–950 m, (2) 1 000–1 050 m or (3) around 1 000 m. The aim of the field research was to verify published data on the occurrence of an Upper Mountain Norway spruce woodland (association *Polysticho-Piceetum*) in the territory of Vysoké skalky. A field study in the area has revealed that most of published data on the occurrence of the Kulczyński's *Picea* woodland are incorrect, as its real upper altitudinal limit reaches  $\pm$  940 m. The main result is the confirmation of the real existence of a *Vaccinium myrtillus-Homogyne alpina-Picea abies* phytocoenosis in the territory of Vysoké skalky. However, tree species other than *Picea abies* (e.g. *Fagus sylvatica*, *Abies alba*, *Acer pseudoplatanus*) could persist in the special habitat of Kulczyński's 'Picea woodland'. The natural vegetation of this place was formed by mixed forest stand.

**Malé Pieniny Mts / *Picea abies* woodland / phytosociology / *Polysticho-Piceetum* / Vysoké skalky**

**Kivonat** – A közönséges luc természetes állománya a Pieninekben (Nyugati-Kárpátok). Kulczyński (1928) természetes lucos állomány előfordulását jelezte a Pieninek montán régiójában (a Vysoké skalky csúcs térségében). A későbbiekben ezen állomány tengerszintfeletti magasságáról (s egyáltalán elhelyezkedéséről) különböző adatokat közöltek: (1) 890 (900)–950 m, (2) 1 000–1 050 m vagy (3) 1 000 m körül. A terepi kutatások célja a montán lucfenyves társulás (*Polysticho-Piceetum*) előfordulásának felülvizsgálata volt a Vysoké skalky térségében. A vizsgálatok rávilágítottak arra, hogy a Kulczyński-féle állományra vonatkozó adatok többsége téves. A felmérés megerősítette egy *Vaccinium myrtillus-Homogyne alpina-Picea abies* növényközösség előfordulását a területen (ennek felső elterjedési határa 940 m tszf. m. körül található), azonban súlypontos más lombos fafajok (pl. *Fagus sylvatica*, *Abies alba*, *Acer pseudoplatanus*) előfordulása is a térségben, azaz a természetes erdőtársulás fétételezhetően egy montán, lombelegyes fenyves lehetett.

**Kis-Pieninek / *Picea abies* állományok / növénytársulástan / *Polysticho-Piceetum* / Vysoké skalky**

## 1 INTRODUCTION

A distinct feature of the landscape of higher mountain ranges of the Western Carpathians is formation of a separate altitudinal vegetational zone dominated by native *Picea abies* forests (class *Piceetea excelsae* Klika 1948). According to the different ecological conditions and corresponding floristical variance, several associations were described that are divided into

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two major groups (Kučera 2012a): (A) *Piceetalia excelsae* Pawłowski ex Pawłowski et al. 1928 on generally nutrient-poor acidic substrates and (B) *Athyrio filicis-feminae-Piceetalia* Hadač ex Hadač et al. 1969 on calcareous substrates. In the Slovak part of the Western Carpathians, seven associations among the two major groups of *Picea abies*-dominated plant communities are currently recognised in the present (Kučera 2012a): (A) *Vaccinio myrtilli-Piceetum*, *Athyrio alpestris-Piceetum*, and *Chrysanthemo rotundifolii-Piceetum* and (B) *Seslerio variae-Piceetum*, *Corthuso matthioli-Piceetum*, *Adenostylo alliariae-Piceetum excelsae*, and *Oxalido acetosellae-Piceetum*. In the Polish phytocoenological literature there are two traditionally differentiated units of supramontane *Picea abies*-dominated communities (see J. M. Matuszkiewicz 2002): *Plagiothecio-Piceetum tatricum* (name is validly published only with the epithet ‘tatricum’; a syntaxonomic synonym to *Vaccinio myrtilli-Piceetum* Šoltés 1976) and *Polysticho-Piceetum* (a syntaxonomic (heterotypical) synonym to *Oxalido acetosellae-Piceetum* Březina et Hadač in Hadač et al. 1969 (Kučera 2012a)).

The first evidence of the existence of a native Norway spruce stand (*Picea abies*) in the Pieniny Mts was published by Kulczyński (1928) in the famous work *The plant associations of the Pieniny*. This author gave a short introduction to the differentiation of Carpathians’ forests into altitudinal vegetation zones and pointed out that the typical form of *Picea abies* forests could also exist out of the elevation range 1 200–1 550 m (i.e. altitudinal range usually considered as a territory of native supramontane *Picea abies* forests) as a result of natural conditions.

In the Pieniny Mts, reaching up to only 1 049.8 m (Vysoké skalky/Wysokie Skalky/Wysoka in the eastern part of the Pieniny Mts – the so-called Malé Pieniny/Male Pieniny), he found an example of such ‘extrazonal’ occurrence, which he regarded as a natural phytocoenosis:

“Auf dem Gebiete des Pieniny-Zuges tritt der natürliche Fichtenwald nur an einer einzigen Stelle, unterhalb des Gipfels »Wysokie Skalki«, in einer Höhe von ca. 900 ü. M. auf. Er ist als ein kleines Assoziationsindividuum von einigen Hektaren Oberfläche ausgebildet.” (Kulczyński 1928, p. 119).

A very brief description and evaluation of this stand situated at the Polish slopes of Vysoké skalky is followed by a complete listing of species found in the whole stand, with cover-abundance values (Kulczyński 1928, p. 119–120).

To date, I have not found published phytosociological relevés from the area of Kulczyński’s *Picea* stand of the Malé Pieniny in the available literature sources. Later authors have only mentioned occurrence of a special vegetation type of *Picea abies* in the Pieniny Mts, mostly without reference to Kulczyński (1928). J. Matuszkiewicz (1977) evaluated Kulczyński’s stand within the association *Polysticho lonchitidis-Piceetum* W. Matuszkiewicz ex J. Matuszkiewicz 1977 and utilized the data of Kulczyński (1928) as a ‘phytosociological relevé’ (see Results).

Reference to the occurrence of the association *Polysticho-Piceetum* in the Pieniny Mts was repeated by W. Matuszkiewicz (1982, 2014), W. Matuszkiewicz – J. M. Matuszkiewicz (1996) and J. M. Matuszkiewicz (2002). In contrast, Medwecka-Kornaś (1959, 1972) did not include the Pieniny Mts in the list of mountain ranges with occurrence of a vegetation type of Norway spruce in her overview of woodland and scrub communities of Poland.

Localization of the occurrence of the *Picea abies* stand under consideration (Malé Pieniny, on the slope of Vysoké skalky) varies in the publications. According to the first author, Kulczyński (1928), his hectares of *Piceetum excelsae* occur in the elevation ca. 900 m. In the chapter of Zarzycki (1982, Tab. I), the altitudinal range of *Piceetum excelsae* occurrence is limited to elevations of 890–950 m. The same values are given in Tab. I by Zarzycki (1981, p. 14), although in the text (p. 32) the indicated values are 900–950 m.

A somewhat different place is given by W. Matuszkiewicz (1982, p. 216–217, *Polysticho-Piceetum*; 2014): in the elevation ca. 1 000–1 050 m below the summit of

Wysokie Skalki. This location was apparently followed by Szelağ (1995) as he wrote that *Polysticho-Piceetum* occurs only in the summit part of the Wysokie Skalki, at the elevation 1 000–1 050 m. In the same book as Zarzycki (see above), Pancer-Kotejowa et al. (1982, p. 319) mentioned an elevation ‘around 1 000 m’.

In the third group of works, there is no exact elevation range in metres given, e.g. J. Matuszkiewicz (1977): on the highest summit of the Malé Pieniny; Pancer-Kotejowa et al. (1982) on page 325 did not give exact data on location (see above).

In respect to the overall distribution of natural non-mixed *Picea abies* forests in the Western Carpathians (Matuszkiewicz 2002, Kučera 2012a, cf. Kulczyński 1928, p. 118), published data on the occurrence of a natural *Picea abies* community in the Pieniny Mts are very interesting. Such a unique existence was probably one of the reasons why this area is under protection in the form of a nature reserve, declared several decades ago. As the original data was published only by Kulczyński (1928), I was drawn to understand the nature of such special *Piceetum* occurrence to supplement my knowledge of Western Carpathians’ *Picea abies* woodlands and to gain field experience and data by myself. Verification of published data on the occurrence of a natural Norway spruce woodland in the territory of Vysoké skalky was the aim of the undertaken research: its results are presented in this paper.

## 2 MATERIAL AND METHODS

Phytosociological relevés were carried out in the year 2011 in cover-abundance scale in terms of Braun-Blanquet (1951) and extended with degrees 2a and 2b according to Barkman et al. (1964) (cf. Westhoff – van der Maarel 1973). Examples of the recorded phytocoenoses (7 phytosociological relevés) are given in the Appendix. Coordinates (WGS-84) were obtained with the device GPSMAP® 60CSx in addition to a hiking map of Pieninský národný park (2001). The age and height of trees was estimated (cf. notes in Kučera 2013). Nomenclature of the vascular plants and bryophytes follows the lists of Marhold et al. (1998) and Kubinská – Janovicová (1998). Collected specimens of bryophytes (herbarium BBZ) were determined by Mgr A. Petrášová and chosen vascular plants by RNDr D. Bernátová, CSc. The programs Turboveg for Windows (Hennekens 2012) and JUICE (Tichý 2012) [cf. Hennekens – Schaminée 2001, Tichý 2002] were used for the processing of phytosociological relevés.

## 3 RESULTS

### 3.1 Actual vegetation conditions of forests at Vysoké skalky

In fact, the uppermost part of the slopes of Vysoké skalky is not covered by stands of *Picea abies*. A substantial part of the cited elevations 1 000–1 050 m (see Introduction) is occupied by a compact summit rock, available without climbing only from the northwest side from a very short ridge with the lowest elevation 1 032 m. Above that only spurs of *Fagus sylvatica* forest reach, and the highest situated trees are of *Sorbus aucuparia*, *Fagus sylvatica* and *Acer pseudoplatanus* (with *Coryllus avellana*, *Sambucus racemosa*, *Salix caprea*, *Cotoneaster integerrimus*, *Ribes alpinum*).

Adjacent slopes on the Slovak part of the state border (running along the main ridge of the mountain range) are covered by climax deciduous forests dominated by *Fagus sylvatica*; *Acer pseudoplatanus* is admixed, I noticed saplings of *Abies alba* and also several *Picea abies* trees. Cultures of *Picea abies* spread below the summit of Vysoké skalky in lower altitudes only.

A similar picture appears on the Polish slopes of the area around the summit of the Vysoké skalky. Notable is a treeless gap to the northeast between summit rock walls and the upper forest line reaching around 1 029 m a.s.l. (49°22,833' N, 20°33,333' E, ± 6 m). Forest stands are dominated by *Fagus sylvatica* as demonstrated by phytosociological sample No. 1 (see Appendix).

To the east of the relevé plot No. 1, the field layer has a more grassy character (*Hordelymus europaeus*) and onward is without *Lunaria* (with *Aconitum moldavicum* [cf. Kučera 2011], *Cimifuga europaea*). Above the upper edge of the relevé plot No. 1 no stand of *Picea abies* grew in the past: located there is a group of three dead *Picea abies* trunks only, and several other *Picea* trees are scattered in the beech forest in the vicinity of the relevé No. 1.

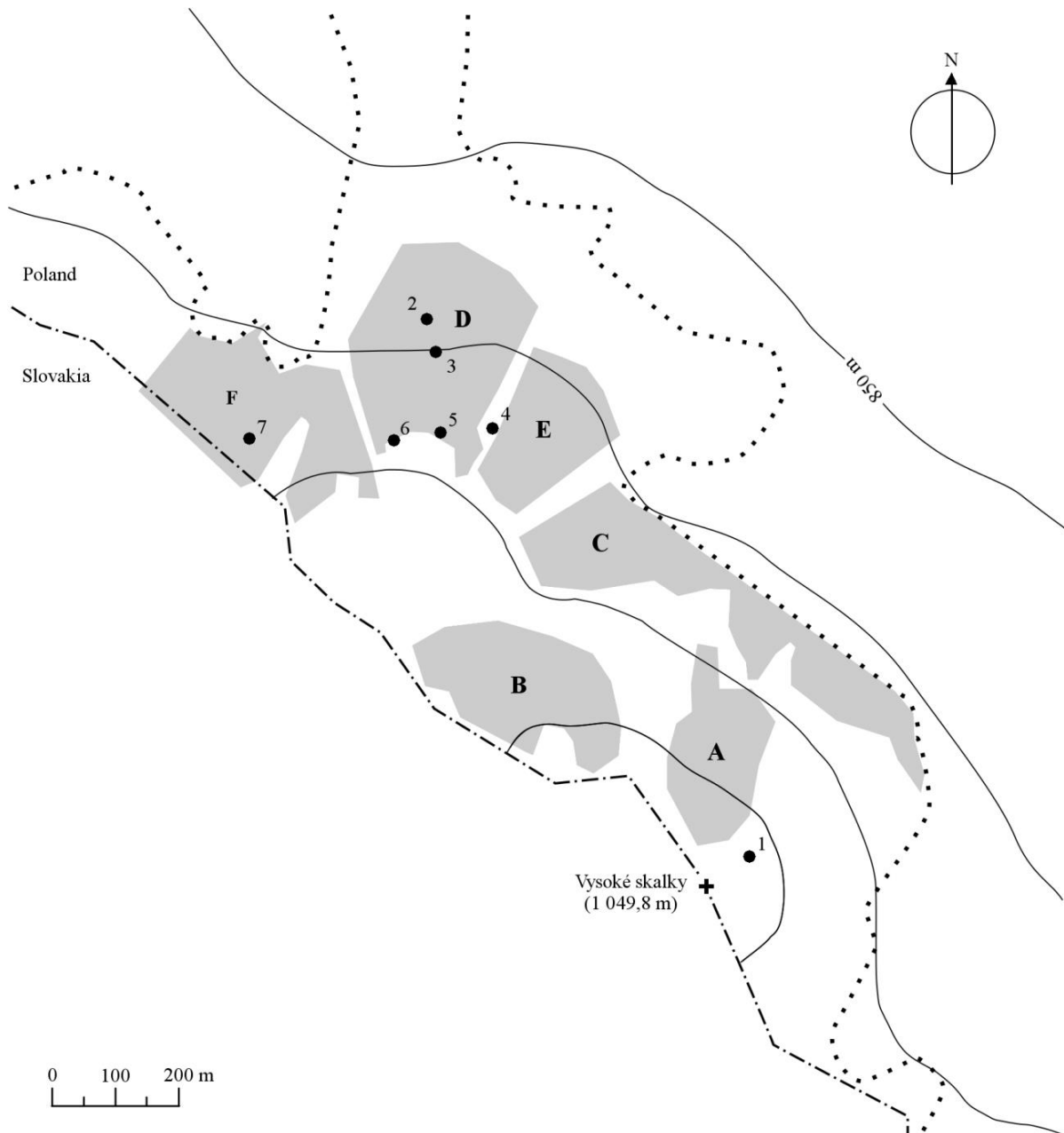
Nevertheless, there is one stand of *Picea abies* close to the summit of Vysoké skalky (Figure 1: A). It is formed as an island inside of a beech forest and its upper limit reaches to elevation ca. 1 020 m (below the northwest ridge of the Vysoké skalky) and its lower limit to ca. 950 m. This stand was severely disturbed some years ago (before 2010) and today contains only a small portion of living *Picea abies* trees.

The herb layer and the entire spread of this former *Picea abies* island is marked by dominant *Rubus idaeus*, while a field layer of the adjacent beech areas under the summit ridge is made up of various vegetation types (*Galium odoratum*, *Asarum europaeum*, *Oxalis acetosella* [*Polygonatum verticillatum*, *Senecio ovatus*, sparsely *Rubus idaeus* of lower growth etc., also saplings of *Acer platanoides*] or *Dryopteris filix-mas*, *Galium odoratum*, *Lunaria rediviva* or below the summit *Lunaria rediviva*, *Dryopteris filix-mas* [*Lonicera nigra*, *Sambucus racemosa*, *Dentaria glandulosa*]). The most remarkable feature of the canopy of this *Picea abies* island (Figure 1: A) is the equal age of the trees (age class ca. 60–80 years): the stand is a planted monoculture. Also, the overall natural conditions (e.g. soil, climate) of mentioned *Picea*- vs. *Fagus*-dominated adjacent stands are more or less the same: the most important factor that differentiated the development of the undergrowth of the respective stands was the presence or absence of an anthropic influence = the plantation of *Picea abies*.

Similarly differentiated is another *Picea abies* island (Fig. 1: B) lying west of coordinates 49°22,859' N, 20°33,277' N (± 6 m, 1 027 m), with a lower elevation limit of 964 m (49°22,916' N, 20°33,197' E, ± 12 m). In the same manner as the previous one, it has artificial borders to the adjacent *Fagus* stands. This *Picea abies* stand-island was also planted by man less than ten decades ago (up to ca. 60–80 years). Part of the *Picea* trees was also wind-blown and another part died after a recent parasite infestation, but the rest *Picea* trees have survived. The area of the windthrow is overgrown by *Rubus idaeus* and *Senecio nemorensis* agg. Today there is admixed *Fagus sylvatica*, which rejuvenates (I noticed also a sapling of *Abies alba* in the stand's border). A potential rejuvenation of *Picea abies* is blocked here by rich secondary undergrowth: this subpopulation (as well as the previous one) will not survive a natural development of the vegetation cover.

Also the last of the *Picea abies* stands below the summit of Vysoké skalky on the Polish side of the state border slopes, neighbouring upon recent pastures (Figure 1: C, prevailing area between elevations 910–940 m), is a plantation, nowadays mostly destroyed by wind (*Rubus idaeus* is the dominant plant species recently).

Further apart from the summit of Vysoké skalky to the northwest, there are other *Picea abies* stands (Figure 1: D–E, F). Finally, in the area of the stand marked as 'D', I could confirm the data of Kulczyński (1928, p. 119) regarding the existence of *Picea abies* woodland with an herb layer dominated by *Vaccinium myrtillus*, with occurrence of *Homogyne alpina* and also *Vaccinium vitis-idaea*. Its elevation range is ca. 870–940 m. This unique vegetation is induced by thin soil cover developing on special bedrock – carbonate hill debris with boulders frequently reaching dimensions of 40 × 40 cm. Debris boulders are not weathering much so the corresponding habitat is very conservative. Uprooting of trees reveals bare boulders of hill debris.



Legend:

- Individual areas of Norway spruce growth (A–F) in the territory of Vysoké skalky
- Border of current woodland
- State border between Slovakia and Poland
- Contour lines, interval 50 m
- Sites of phytosociological relevés No. 1–7 (see Appendix)

Figure 1. Sketch map of the locality

However, also here a great part of *Picea abies* population was attacked by parasites (before 2010) and died. Insect attack was most probably preceded by wind disturbance of this and other stands in the vicinity. However, a sufficient amount of *Picea* trees resisted and survival of this population is ensured by rich natural rejuvenation of Norway spruce with variable age spans, and forming thickets here and there. An example of the vegetation type is given by the phytosociological relevé No. 2. The lateral border of *Vaccinium myrtillus*-*Homogyne alpina*-

*Picea abies* woodland to the former original beech forest is indicated by typical herbaceous facies of beech woodlands (*Galium odoratum* [also dominant], *Asarum europaeum*, *Symphytum cordatum*, or *Astrantia major*, *Petasites albus*; also *Cicerbita alpina* and *Streptopus amplexifolius* grow here (site of the last species: 49°23,001' N, 20°33,097' E, ± 7 m, 930 m)) and the absence of *Vaccinium myrtillus*. The background here at the hill debris border could also be formed by carbonate debris, but the size of the rocks is considerably smaller and the soil profile is deeper than in habitat of *Vaccinium myrtillus*-*Homogyne alpina*-*Picea abies* woodland.

Above the upper right corner of the relevé plot No. 2, vegetation with *Hypericum maculatum*, *Deschampsia cespitosa*, *Agrostis capillaris*, *Taraxacum* sp. is growing. This occurrence indicates an influence of the former pasture.

According to Kulczyński (1928, p. 118–119), this area is home of a native *Picea abies* woodland. However, despite the special habitat conditions, trees of *Fagus sylvatica* could also grow and prosper here and could withstand windstorms. In the vicinity of the relevé plot No. 2, an older *Fagus* tree (more than 60–70 years) is growing and it is a source for (microlocally considerably rich) the rejuvenation of *Fagus*. Such a vegetation sample is given in relevé No. 3, spatially only 10 m from the relevé plot No. 2. The prevailing part of the *Picea* trees on the plot was windblown, other parts died after parasite attack; *Fagus* grow successfully.

However, even *Fagus* can dominate in the canopy in this debris territory. The next relevé (No. 4) was recorded on the lateral debris ridge, with rocks on the surface. A different vegetation type is developed here.

To the east of the relevé plot at the slope beginning down from this lateral ridge, hill debris and communities with *Vaccinium myrtillus* cover the slope in the same manner as west of the ridge (Figure 1: D). *Lonicera nigra* is abundant, rejuvenation of *Picea abies* is rich under the parasite-attacked and wind-blown *Picea* trees (Figure 1: E). Also, an old *Abies alba* tree was recorded (below coordinates 49°22,986' N, 20°33,224' E, ± 9 m, 930 m a.s.l., here also an example of secondary, temporary occurrence of *Cotoneaster integerrimus*, see in Discussion): this tree species should be included among species that once co-determined the original woodland type. Secondary succession is here in a more advanced phase as in the stand marked in the Figure 1 as 'D'.

The original course of the upper limit of the local natural occurrence of woodland with abundant *Picea abies* is hard to determine today. *Picea abies* was propagated by human intervention directly (plantation) or indirectly (spreading by secondary succession). Relevé plot No. 5 is probably one of the highest points mixed woodland with *Picea abies* could reach originally. Today, a great part of the former *Picea* stand has been destroyed – however, its population will persist as the species has rejuvenated long ago. An older *Fagus* tree uphill above the relevé plot is the origin of the numerous beech saplings (with cover-abundance value 4 [–5]), a different habitat is indicated there by *Sanicula europaea* (richly), *Primula elatior*, *Symphytum cordatum*, *Ranunculus platanifolius*, *Mercurialis perennis*, *Galium odoratum* (*Pulmonaria obscura*). Originally there was already abeech woodland developed here.

The recent vegetation cover in the western uppermost part of the *Picea* stand 'D' is interesting. Under dead *Picea* trunks, a rich undergrowth of *Lonicera nigra* and *Vaccinium myrtillus* grows. The slope relief indicates that hill debris is still 'living'. In the geological history, hill debris of 'stand D' probably originated through the collapse of solid rock. In the uppermost, rocky part of the hill debris, with formation of small rock walls and falling rocks and boulders (in the other place, we have seen also a small vertical cavern: 49°22.953' N, 20°33.167' E, ± 7 m), wind and parasite disturbance caused all mature *Picea* specimens to die out and only young ones are living. However, the *Fagus* population here (relevé plot No. 6)

could also grow successfully, supplemented with rejuvenation from trees growing uphill. After *Picea* died out, treeless scrub vegetation partly developed. The uppermost edge of the hill debris begins slightly higher. There, under tall *Fagus* trees, a *Vaccinium myrtillus* population is still growing, but only of a dwarfed growth. Further uphill there is distributed typical beech vegetation of deeper calcareous soils.

The last *Picea abies* stand in the Polish part of the territory of Vysoké skalky, marked on *Figure 1* by the letter 'F', is again a plantation. It continues directly to a younger spruce plantation (age 40–60 years) on the Slovak part of the state border. Relevé No. 7 shows clearly striking differences in the herb layer in comparison to relevés 2, 3 and 5. Also, non-indigenous species *Larix decidua* was planted here.

### 3.2 Formal comments on the association *Polysticho-Piceetum* and data on its occurrence at Vysoké skalky

The correct association name completed according to Recomm. 10C of the International Code of Phytosociological Nomenclature (ICPN: Weber et al. 2000) is *Polysticho lonchitidis-Piceetum* W. Matuszkiewicz ex J. Matuszkiewicz 1977 (see Kučera 2010, who also designated a lectotype to the association). Yet, phytocoenoses of Kulczyński's *Vaccinium myrtillus-Homogyne alpina-Picea abies* woodland (see above) do not correspond to this syntaxon as well as to its probable syntaxonomical synonym *Oxalido acetosellae-Piceetum* Březina et Hadač in Hadač et al. 1969.

Also questionable is the utilisation of Kulczyński's (1928) phytosociological data on '*Piceetum excelsae*' as a phytosociological relevé; for example, in the synthesis of J. Matuszkiewicz (1977). In contrast to other species lists in the work of Kulczyński, the species listing on pages 119–120 (Kulczyński 1928) should not be accepted as a constancy table nor as a single relevé by reason that it is a list of all species found in the whole area of the occurrence of the *Picea abies* stand extending up to 'several hectares' (see Kulczyński 1928, p. 119), only accompanied with cover-abundance values. Therefore, the name proposed by Kulczyński – i.e. *Piceetum excelsae* Kulczyński 1928 – is to be nomenclaturally treated not only as a nomen illegitimum (ICPN Art. 31), but also primarily as a nomen invalidum (Art. 2b → Art. 7).

## 4 DISCUSSION

As shown above, there exist more than one *Picea abies* stand in the territory of Vysoké skalky (*Figure 1*: A–F). The elevation range of both *Picea abies* stands 'A' and 'B', reaching continuously down to 950 m/964 m, considerably exceed the altitudinal limits of 1 000–1 050 m given by Matuszkiewicz (1982, 2014) and Szelağ (1995) for natural occurrence of a *Polysticho-Piceetum* stand. As no such *Picea abies* stands on the slopes of Vysoké skalky exist, these published data should be taken as incorrect. In addition, occurrence of several *Picea abies* trees in the vicinity of the relevé No. 1 could not be the reason for the publishing of such data on occurrence of a native Norway spruce woodland in the summit area of Vysoké skalky.

In respect to elevation range of the stands 'A' and 'B', more closely fitting would be the information of Pancer-Kotejowa et al. (1982, p. 319: around 1 000 m), which could be applied to both *Picea abies* stands under consideration (*Figure 1*: A, B). Nonetheless, their species composition is *completely different* from the phytosociological data of Kulczyński (1928, see p. 119–120). The same stands for the species composition of *Picea* stands are labeled in *Figure 1* as 'C' and 'F'.

On the other hand, the real existence of a *Vaccinium myrtillus*-*Homogyne alpina*-*Picea abies* community in the territory of Vysoké skalky (Kulczyński 1928) was indeed confirmed (see relevé No. 2, within the *Picea abies* stand ‘D’). Field occurrence of this vegetation type corresponded exactly to the place of *Piceetum excelsae* indicated by Kulczyński (1928) in his phytosociological map of the Pieniny. Kulczyński (1928, p. 119) is at the same time almost the only one who published correct information on its distribution; the second person was Zarzycki (1981, 1982) with elevation data 900–950 m resp. 890–950 m.



Figure 2. Plant cover in the site of the relevé No. 2.

Among all other *Picea* stands of Vysoké skalky, the area of the stand ‘D’ is the one that provides a sustainable existence of the *Picea abies* population, including interspecific competition. This is due to a special conservative debris habitat. In my opinion, the stand marked in the map (Figure 1) as ‘E’ is also an example of the refugium habitat of the *Picea abies* population: the total area of the natural occurrence of *Picea abies* in the territory of Vysoké skalky is thus larger than Kulczyński (1928) suggested in his phytosociological map.

However, the phytosociological sample No. 3 confirms that the locality described by Kulczyński (1928, p. 119) is not an example of the authentic *Picea abies* forest. The situation of species composition described by Kulczyński (1928) has its origin in long-term historic land use, deforestation of the prevalent territory of slopes of Malé Pieniny mountain ridge, and changes in species composition of woodland rests. Also, the species list of Kulczyński (1928) confirms the appearance of species with human-influenced distribution, ‘aliens’ in the originally common woodlands of this area: *Antennaria dioica*, *Hypericum maculatum*, *Potentilla erecta*, *Cruciata glabra* (*Veronica chamaedrys*; *Cotoneaster integerrimus* from secondary, temporary lighting of the stand).

Recent development of vegetation cover point to more-balanced mixed stands of *Picea abies* and *Fagus sylvatica* in the future. Thus, in contrast to Kulczyński (1928) and data of other authors on the natural existence of an autochthonous *Picea abies* plant community (see Introduction) in the territory of Vysoké skalky, I am of the opinion that the existence of *Picea abies* in this locality should be evaluated more likely as an isolated local relict of the *Picea abies* population persisting within forest of mixed tree species composition. Constant special habitat conditions are confirmed by remarkable co-occurrence of dominant *Vaccinium*

*myrtilus* and *V. vitis-idaea* at the first place, *Homogyne alpina* and *Calamagrostis villosa*. In contrast to all other *Picea* stands (plantations) in the territory of Vysoké skalky, a gradation of *Rubus idaeus* does not occur.

## 5 SIGNIFICANCE OF *PICEA ABIES* OCCURRENCE AT VYSOKÉ SKALKY

If the population of *Picea abies* of the stands 'D' and 'E' (Figure 1) is really a native (thus relict) population, then it could have developed most likely a unique, isolated gene pool. Genetic studies of possible differences compared to the closest native stands (Tatry Mts) should be considered. *Picea abies* forests predominated in the landscape of northern Slovakia in early Holocene (cf. Jankovská 1991, 1998). In the lower mountain ranges adjacent to the Tatry Mts there is reconstructed a potential natural vegetation consisting of *Abies-Fagus* forests (cf. Kučera 2012a, b; see also Kučera 2015): the *Picea abies* woodland of Vysoké skalky in the Malé Pieniny Mts was separated from the closest native *Picea abies* populations in the Tatry Mts during the Holocene (cf. Krippel 1963, 1986) by natural beech and fir-beech forests of the territory of Pieniny and the Spišská Magura Mts. Current Norway spruce stands covering large areas of both mountain ranges are results of long-term anthropic influence (see below).

Long-term isolation of the *Picea abies* population of Vysoké skalky could also lead to a particular adaptation to ecological conditions of the lower montane vegetation zone, which has special importance for forestry. However, the existence of Norway spruce cultures of uncertain provenience in the vicinity of *Picea abies* population of the stands 'D' and 'E' is a threat to preservation of the supposed original genotypic and phenotypic features.

*Picea abies* individuals and stands originated by plantation and saplings/young trees from their rejuvenation should be strictly regulated above all in the area of the Nature Reserve Wysokie Skalki. Also, spruce plantations west of this nature reserve should not directly influence the genetic composition of the above-mentioned relict population.

## 6 REMARKS TO *PICEA ABIES* OCCURRENCE IN THE SURROUNDINGS OF VYSOKÉ SKALKY

In the wider surroundings, we have also noticed a recent plantation of Norway spruce (cf. also Pawłowski 1925). However, as we can see in the territory of Vysoké skalky, plantations of Norway spruce are very vulnerable cultures in these low altitudes of the Malé Pieniny. Economic profit here is insecure because of the lability of tree stands and strong predisposition of *Picea abies* stands to disturbance (even total elimination) by wind and insect attacks.

Besides the plantation of Norway spruce, on the recent borders of pastures to forest in the Malé Pieniny, easy rejuvenation of *Picea abies* is in progress here and there. This is a typical form of a secondary succession, a spontaneous process of the spreading of forest back to its original place. In the lower altitudes, in a lower montane altitudinal vegetation belt, the first and long-lasting stage (in several phases) is formed by *Picea abies* (cf. Myczkowski – Grabski 1962) if a 'parent' *Picea abies* stand (plantation) is in the surroundings. A secondary succession of *Fagus sylvatica* in such secondary, substitutional *Picea* stands could only be seen much later (cf. Kučera 2012a, chapter 4). However, long-term natural competition between *Picea abies* and other plant species generally results in the elimination of *Picea abies* in the lower montane belt. It seems that this rule has the one and only exception in the

whole region of the Malé Pieniny – a population of *Picea abies* within the Kulczyński's (1928) stand of '*Piceetum excelsae*' is maintained by special habitat conditions.

Except for such special cases, anthropogenic changes of the vegetation cover (direct or indirect) are the reason of the occurrence or even dominance of *Picea abies* in forest stands of the Pieniny Mts. According to my evaluation (cf. Kučera 2007, 2012b), stands evaluated as *Piceo-Abietetum carpaticum* by Kulczyński (1928) also originated by a spontaneous secondary succession, related to the planting of *Picea abies* (or direct plantation establishment) and the historical land use in the centuries before 1900 (see distribution of this plant community on his phytosociological map and extent of deforested area in the early 20th century); nomenclatural status of the name: *Piceo-Abietetum carpaticum* Kulczyński 1928, nom. inval., ICPN Art. 3e (Weber et al. 2000). In the adjacent mountain range of the Spišská Magura, superiority of *Fagus sylvatica* (*Abies alba*) in the natural development of forest vegetation is confirmed up to the uppermost elevation (1 259 m) (Kučera 2012b). Equivalently, the occurrence of a natural *Picea abies* forest zone is not expected in the other adjacent mountain range of the Beskid Sądecki (Kučera 2012a), although development of a such altitudinal vegetation zone was presumed there by Pawłowski (1925) already above elevation 1 100–1 130 m and by Myczkowski – Grabski (1962) above (1 000) 1 050 or 1 100 m; Fabijanowski (1962) expected typical *Picea* forests even at altitudes 780 m and 915 m.

## 7 CONCLUSIONS

According to the field research in the region of Vysoké skalky with data on the occurrence of *Piceetum excelsae* published by Kulczyński (1928) I conclude:

1. Almost all stands of *Picea abies* on the slopes of Vysoké skalky are plantations – cultures unstable to environmental influences.
2. Only one local *Picea abies* stand (Figure 1: D, E) corresponds to the phytosociological data of Kulczyński (1928).
3. Data of altitudinal range of the occurrence of *Picea* stand corresponding to Kulczyński's data published by Matuszkiewicz (1982), Pancer-Kotejowa et al. (1982, p. 319) and Szeląg (1995) are incorrect.
4. The plant community of the Kulczyński's *Picea* stand is not a native *Picea abies* woodland, and should probably be evaluated as a natural, isolated refugium of occurrence of a *Picea abies* population within a mixed forest stand.
5. Nature conservation should consider an appropriate management in respect of the preservation of this presumably unique gene pool of *Picea abies* in the Malé Pieniny.

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## APPENDIX

Phytosociological relevés recorded in the region of Vysoké skalky

**Relevé No. 1:** 49°22.826' N, 20°33.360' E, ± 6 m, 1 023 m a.s.l. (upper limit), steep slope, aspect NE (46 °), relevé area 20 × 20 m, total cover 100%, E<sub>3</sub> 95%, E<sub>2</sub> 15%, E<sub>1</sub> 80%, E<sub>0</sub> 0%, rocks 1%, wood 0,5%, *Fagus* trees above 80–100 years, *Fagus* perimeter<sub>1.3</sub> [p.] 149 cm, height (h.) [29] m, age up to 80 years, 18. 7. 2011, P. Kučera, R. Rapant (PK220).

E<sub>3</sub>: *Fagus sylvatica* 5, *Acer pseudoplatanus* 1, *Picea abies* 1,

E<sub>2</sub>: *Fagus sylvatica* 2b, *Corylus avellana* r,

E<sub>1</sub>: *Acer pseudoplatanus* +, *Fagus sylvatica* +, *Lonicera xylosteum* r, *Picea abies* r, *Ribes alpinum* r, *R. uva-crispa* r,

*Lunaria rediviva* 3, *Galium odoratum* 2b, *Dryopteris filix-mas* 1, *Galeobdolon montanum* 1, *Geranium robertianum* 1, *Hordelymus europaeus* 1, *Hylotelephium argutum* 1, *Mercurialis perennis* 1, *Oxalis acetosella* 1, *Poa nemoralis* 1, *Roegneria canina* 1 (det. D. Bernátová), *Symphytum cordatum* 1, *Aconitum variegatum* +, *Actaea spicata* +, *Adoxa moschatellina* +, *Asarum europaeum* +, *Bromus benekenii* + (det. D. Bernátová), *Campanula trachelium* +, *Dentaria glandulosa* +, *Epilobium montanum* +, *Galeopsis speciosa* +, *Geranium sylvaticum* +, *Glechoma hirsuta* +, *Hypericum hirsutum* +, *Impatiens noli-tangere* +, *Lamium maculatum* +, *Lathyrus vernus* +, *Milium effusum* +, *Myosotis* sp. +, *Paris quadrifolia* +, *Polygonatum verticillatum* +, *Polystichum aculeatum* +, *Ranunculus platanifolius* +, *Rubus idaeus* +, *Senecio ovatus* +, *Silene dioica* +, *Stachys sylvatica* +, *Thalictrum aquilegifolium* +, *Viola reichenbachiana* +, *Calamagrostis varia* r, *Carduus personata* r, *Cicerbita alpina* r, *Cirsium erisithales* r, *Dryopteris carthusiana* r, *Petasites albus* r, *Primula elatior* r, *Ranunculus lanuginosus* r, *Salvia glutinosa* r, *Tithymalus amygdaloides* r, *Urtica dioica* r, *Veronica officinalis* r.

**Relevé No. 2:** 49°23.046' N, 20°33.163' E, ± 8 m, 890 m a.s.l., aspect variable (NNW, 328 °), total cover 100%, E<sub>3</sub> 45%, E<sub>2</sub> 20%, E<sub>1</sub> 93%, E<sub>0</sub> 20%, fallen wood 1%, age of *Picea* trees up to 80 r., *Picea* p. 96 cm, h. [22] m, age up to cca 80 years, 17. 7. 2011, P. Kučera, R. Rapant (PK217).

E<sub>3</sub>: *Picea abies* 3,

E<sub>2</sub>: *Picea abies* 2b (in spots), *Lonicera nigra* 1, *Fagus sylvatica* +, *Salix caprea* +, *Salix* × *subcaprea* r,

E<sub>1</sub> *Picea abies* 2a, *Lonicera nigra* 1, *Acer pseudoplatanus* +, *Fagus sylvatica* +, *Ribes petraeum* +, *Salix caprea* +, *Sorbus aucuparia* +, *Abies alba* r, *Corylus avellana* r,

E<sub>1</sub> A: *Vaccinium myrtillus* 4, *Homogyne alpina* 2b, *Rubus idaeus* 2a, *Dryopteris dilatata* 1, *Gymnocarpium dryopteris* 1, *Lycopodium annotinum* 1, *Maianthemum bifolium* 1, *Oxalis acetosella* 1, *Valeriana tripteris* 1, *Avenella flexuosa* +, *Calamagrostis villosa* +, *Luzula luzuloides* +, *L. sylvatica* ssp. *sylvatica* +, *Orthilia secunda* +, *Poa nemoralis* +, *Vaccinium vitis-idaea* +, *Deschampsia cespitosa* r, *Urtica dioica* r,

E<sub>1</sub> B: *Agrostis capillaris* +, *Athyrium filix-femina* +, *Chrysosplenium alternifolium* +, *Fragaria vesca* +, *Gentiana asclepiadea* +, *Paris quadrifolia* +, *Senecio ovatus* +, *Alchemilla* sp. r, *Calamagrostis varia* r, *Dentaria glandulosa* r, *Geum rivale* r (lower border of the relevé plot), *Epilobium montanum* r, *Primula elatior* r, *Prunella vulgaris* r, *Streptopus amplexifolius* r, *Veronica officinalis* r,

E<sub>0</sub>: *Dicranum scoparium* 2a, *Polytrichum formosum* 1, *Sphagnum quinquefarium* 1, *Plagiochila porelloides* +, *Plagiomnium affine* +, *Plagiothecium denticulatum* +, *Tritomaria quinquedentata* +, *Calypogeia muelleriana* +, *Cephalozia bicuspidata* +, *Eurhynchium angustirete* +, *Lepidozia reptans* +.

Inside of the chosen relevé plot (of standard size 20 × 20 m) a slightly different microhabitat (E<sub>1</sub> B, more wet and nutrient, with two fallen *Picea* trunks) fell within in the lower part of the plot.

**Relevé No. 3:** 49°23.031' N, 20°33.169' E, ± 8 m, 900 m a.s.l., aspect variable (NNW, 347 °), relevé area 15 × 20 m, total cover 100%, E<sub>3</sub> 40%, E<sub>2</sub> 18%, E<sub>1</sub> 90%, E<sub>0</sub> 10%, fallen wood 2%, *Picea* p. 121 cm, h. [22,5] m, age up to 60–80 years, *Fagus* trees (at left bottom part) age up to 30 years, h. 6 (7) m, 17. 7. 2011, P. Kučera, R. Rapant (PK218).

E<sub>3</sub>: *Fagus sylvatica* 2b, *Picea abies* 2b (before disturbance 3),

E<sub>2</sub>: *Picea abies* 2a, *Corylus avellana* 1, *Fagus sylvatica* 1, *Acer pseudoplatanus* +, *Sorbus aucuparia* +, *Abies alba* r,

E<sub>1</sub>: *Lonicera nigra* 1, *Picea abies* 1, *Acer pseudoplatanus* +, *Daphne mezereum* +, *Fagus sylvatica* +, *Rosa pendulina* +, *Salix*×*subcaprea* +, *Sorbus aucuparia* +, *Abies alba* r,

*Vaccinium myrtillus* 2b, *Calamagrostis arundinacea* 2a, *Rubus idaeus* 2a, *Athyrium filix-femina* 1, *Calamagrostis varia* 1, *C. villosa* 1, *Dryopteris filix-mas* 1, *Galeobdolon montanum* 1, *Galium odoratum* 1, *Gentiana asclepiadea* 1, *Homogyne alpina* 1, *Oxalis acetosella* 1, *Senecio ovatus* 1, *Valeriana tripteris* 1, *Clematis alpina* +, *Dryopteris dilatata* +, *Fragaria vesca* +, *Galium rotundifolium* +, *Gymnocarpium dryopteris* +, *Luzula luzuloides* +, *Maianthemum bifolium* +, *Mycelis muralis* +, *Prenanthes purpurea* +, *Rubus fruticosus* agg. +, *Agrostis capillaris* r, *Avenella flexuosa* r, *Chamerion angustifolium* r, *Deschampsia cespitosa* r, *Epilobium montanum* r, *Hieracium* sp. r, *Hypericum maculatum* r, *Urtica dioica* r, *Vaccinium vitis-idaea* r, *Veronica officinalis* r.

E<sub>0</sub>: *Eurhynchium angustirete* 1, *Dicranum scoparium* 1, *Barbilophozia barbata* +, *Plagiochila porelloides* +, *Plagiomnium affine* + *Sphagnum quinquefarium* +.

**Relevé No. 4:** 49°22.999' N, 20°332.208' E, ± 14 m, 927 m a.s.l., gentle slope, aspect NNE (27 °), relevé area 20 × 20 m, total cover 98%, E<sub>3</sub> 70%, E<sub>2</sub> 25%, E<sub>1</sub> 75%, bryophytes only on rocks, rocks 8%, *Fagus* trees (up to) 60–80 years, *Fagus* h. above 20 m (also *Acer*), age up to 70–80 years, 17. 7. 2011, P. Kučera, R. Rapant (PK219).

E<sub>3</sub>: *Fagus sylvatica* 4, *Acer pseudoplatanus* 2b, *Picea abies* 1,

E<sub>2</sub>: *Corylus avellana* 2b, *Fagus sylvatica* 2a, *Lonicera nigra* +,

E<sub>1</sub>: *Fagus sylvatica* (1) 2a, *Acer pseudoplatanus* 1, *Fraxinus excelsior* +, *Lonicera nigra* +, *Picea abies* +, *Rosa pendulina* +, *Abies alba* r, *Sambucus racemosa* r, *Sorbus aucuparia* r, *Viburnum opulus* r,

E<sub>0</sub>: *Galium odoratum* 2b, *Dryopteris filix-mas* 2a, *Calamagrostis varia* 1, *Galeobdolon montanum* 1, *Melica nutans* 1, *Oxalis acetosella* 1, *Rubus fruticosus* agg. 1, *Vaccinium myrtillus* 1, *Valeriana sambucifolia* 1 (det. D. Bernátová), *Brachypodium sylvaticum* +, *Calamagrostis arundinacea* +, *Chaerophyllum hirsutum* +, *Clinopodium vulgare* +, *Cruciata glabra* +, *Dentaria glandulosa* +, *Epilobium montanum* +, *Festuca* sp. +, *Fragaria vesca* +, *Galium anisophyllum* +, *Hordelymus europaeus* +, *Hypericum maculatum* +, *Luzula luzuloides* +, *Maianthemum bifolium* +, *Mycelis muralis* +, *Paris quadrifolia* +, *Poa nemoralis* +, *Polypodium vulgare* +, *Polystichum aculeatum* +, *Prenanthes purpurea* +, *Primula elatior* +, *Ranunculus platanifolius* +, *Rubus idaeus* +, *Sanicula europaea* +, *Senecio ovatus* +, *Valeriana tripteris* +, *Viola reichenbachiana* +, *Ajuga reptans* r, *Athyrium filix-femina* r, *Epipactis helleborine* r, *Hylotelephium maximum* r, *Polygonatum verticillatum* r, *Urtica dioica* r, *Veronica chamaedrys* r.

Outside of the relevé plot at the slope beginning down from this lateral ridge: *Lonicera xylosteum*, *Ribes alpinum*.

**Relevé No. 5:** 49°22.998' N, 20°33.170' E, ± 6 m, 935 m a.s.l., steep slope, aspect NNE (25 °), relevé area 20 × 20 m, total cover 100%, E<sub>3</sub> 40% (formerly above 75%), E<sub>2</sub> 55%, E<sub>1</sub> 80%, E<sub>0</sub> cca 15%, *Fagus* tree probably above 80 years, h. 16–17 m, *Picea* trees were up to 60–80 years old, most of them are dead, *Picea* h. above 20 m (21–22 m), 18. 7. 2011, P. Kučera, R. Rapant (PK223).

E<sub>3</sub>: *Picea abies* 3, *Fagus sylvatica* 2a (2b), *Abies alba* 1 (young), *Acer pseudoplatanus* 1 (young),

E<sub>2</sub>: *Lonicera nigra* 3, *Corylus avellana* 2a, *Fagus sylvatica* 2a, *Picea abies* 2a, *Sorbus aucuparia* ssp. *aucuparia* 1, *Abies alba* +, *Acer pseudoplatanus* +,

E<sub>1</sub> (partly mixed types): *Picea abies* 2a, *Lonicera nigra* 1, *Abies alba* +, *Acer pseudoplatanus* +, *Fagus sylvatica* +, *Ribes alpinum* +, *Sorbus aucuparia* +, *Corylus avellana* r, *Salix silesiaca* r,

*Vaccinium myrtillus* 3, *Galium odoratum* 2a, *Gymnocarpium dryopteris* 2a, *Calamagrostis varia* 1, *Dryopteris dilatata* 1, *Hordelymus europaeus* 1, *Galeobdolon montanum* 1, *Oxalis acetosella* 1, *Rubus idaeus* 1, *Vaccinium vitis-idaea* 1, *Valeriana tripteris* 1, *Asplenium viride* +, *Athyrium filix-femina* +, *Chamerion angustifolium* +, *Dentaria glandulosa* +, *Dryopteris filix-mas* +, *Epilobium montanum* +, *Fragaria vesca* +, *Maianthemum bifolium* +, *Petasites albus* +, *Phegopteris connectilis* +, *Poa nemoralis* +, *Polystichum aculeatum* +, *Senecio ovatus* +, *Pyrethrum clusii* +, *Viola reichenbachiana* +, *Avenella flexuosa* r, *Carex digitata* r, *Clematis alpina* r, *Cystopteris fragilis* r, *Hieracium* sp. r, *Hypericum maculatum* r, *Mercurialis perennis* r, *Ranunculus platanifolius* r, *Sanicula europaea* r, *Streptopus amplexifolius* r, *Urtica dioica* r,

E<sub>0</sub>: *Rhizomnium punctatum* 1, *Eurhynchium angustirete* 1, *Dicranum scoparium* 1, *Blepharostoma trichophyllum* +, *Calypogeia muelleriana* +, *Cephalozia bicuspidata* +, *Lepidozia reptans* +, *Lophozia ventricosa* +, *Plagiochila asplenioides* +, *Polytrichum formosum* +, *Sphagnum quinquefarium* +, *Tetraphis pellucida* +.

**Relevé No. 6:** 49°22.996' N, 20°33.143' E, ± 5 m, 940 m a.s.l., slope variable (total [40] °), aspect N (350 °), relevé area 20 × 20 m, total cover 97%, E<sub>3</sub> (68%), E<sub>2</sub> (35%), E<sub>1</sub> (40%), E<sub>0</sub> (35% everywhere on rocks), *Fagus* h. even 20 m, *Picea* h. above 20 m, 18. 7. 2011, P. Kučera, R. Rapant (PK222).

E<sub>3</sub>: *Fagus sylvatica* 4, *Picea abies* 2b [formerly 3],

E<sub>2</sub>: *Lonicera nigra* 2b, *Ribes alpinum* 2a, *Corylus avellana* 1, *Picea abies* 1, *Salix caprea* 1, *Sambucus racemosa* 1, *Sorbus aucuparia* +, *Lonicera xylosteum* r, *Salix* × *subcaprea* r,

E<sub>1</sub>: *Sambucus racemosa* 1, *Ribes alpinum* 1, *Lonicera nigra* 1, *Sorbus aucuparia* +, *Rosa pendulina* +, *Acer pseudoplatanus* +, *Picea abies* r, *Abies alba* r,

*Gymnocarpium dryopteris* 2a, *Oxalis acetosella* 2a, *Geranium robertianum* 2a, *Dryopteris filix-mas* 1, *Galeobdolon montanum* 1, *Galium odoratum* 1, *Rubus idaeus* 1, *Urtica dioica* 1, *Vaccinium myrtillus* 1, *Athyrium filix-femina* +, *Brachypodium sylvaticum* +, *Cardamine flexuosa* +, *Cystopteris fragilis* +, *Dryopteris dilatata* +, *Epilobium montanum* +, *Fragaria vesca* +, *Hylotelephium argutum* +, *Luzula luzuloides* +, *Paris quadrifolia* +, *Poa nemoralis* +, *Polypodium vulgare* +, *Polystichum aculeatum* +, *Senecio ovatus* +, *Valeriana tripteris* +, *Actaea spicata* r, *Asplenium trichomanes* r, *A. viride* r, *Cardaminopsis arenosa* r, *Dentaria glandulosa* r, *Dryopteris expansa* r, *Valeriana sambucifolia* r.

**Relevé No. 7:** 49°22.993' N, 20°33.053' E, ± 5 m, 932 m a.s.l., gentle slope, aspect NNW/N (348 °), relevé area 20 × 20 m, total cover 97%, E<sub>3</sub> 68%, E<sub>2</sub> 2%, E<sub>1</sub> 95%, E<sub>0</sub> cca 15%, wood 5%, *Picea* trees 60–80 years, *Picea* p. 129 cm, h. [25,5] m, age 60–80 r., 18. 7. 2011, P. Kučera, R. Rapant (PK221).

E<sub>3</sub>: *Picea abies* 4, *Larix decidua* r,

E<sub>2</sub>: *Fagus sylvatica* +, *Acer pseudoplatanus* r, *Fraxinus excelsior* r, *Lonicera nigra* r,

E<sub>1</sub>: *Fagus sylvatica* 1, *Acer pseudoplatanus* +, *Fraxinus excelsior* +, *Abies alba* r, *Lonicera nigra* r, *Sambucus racemosa* r, *Sorbus aucuparia* r,

*Rubus idaeus* 3, *Senecio ovatus* 3, *Oxalis acetosella* 2b, *Galeobdolon montanum* 2a, *Asarum europaeum* 1, *Athyrium filix-femina* 1, *Dryopteris dilatata* 1, *D. filix-mas* 1, *Galium rotundifolium* 1, *Geranium robertianum* 1, *Lysimachia nemorum* 1 (det. D. Bernátová), *Adoxa moschatellina* +, *Cardamine flexuosa* + (det. D. Bernátová), *Festuca gigantea* +, *Galium odoratum* +, *Mercurialis perennis* +, *Mycelis muralis* +, *Poa nemoralis* +, *Primula elatior* +, *Silene dioica* +, *Urtica dioica* +, *Viola reichenbachiana* +, *Actaea spicata* r, *Chamerion angustifolium* r, *Epilobium montanum* r, *Fragaria vesca* r, *Hylotelephium argutum* r, *Platanthera bifolia* r, *Prenanthes purpurea* r, *Roegneria canina* r (det. D. Bernátová), *Salvia glutinosa* r.

E<sub>0</sub>: *Brachythecium reflexum* 1, *B. starkei* 1, *Rhizomnium punctatum* 1, *Dicranum scoparium* +, *Plagiothecium denticulatum* +, *Plagiomnium affine* +.

# Micropropagation of Leuce-poplars and evaluation of their development under sandy site conditions in Hungary

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**Abstract** – Leuce-poplars are a native stand-forming tree species throughout Hungary. Several species or selections of them are used as ornamental plants in parks or to line streets and highways. They cover approximately 4.0 per cent of the total forested area in Hungary (70 000 ha). The white (grey) poplar belongs to the Leuce poplars and plays a significant role in sand fixation, regional forestation, and nature conservation. The National Agricultural Research and Innovation Centre, Forest Research Institute or NARIC-FRI (formerly known as the Forest Research Institute) is involved in long-term breeding work for the selection of fast-growing white poplar trees under dry conditions. In vitro multiplication of trees is applied mainly to fruit growing trees in Hungary; in forestry research it is used primarily for selective breeding. This paper presents a short overview of the most important issues concerning the biotechnology of different *Populus* species, the related research on micropropagation trials, and the results of field investigations of micropropagated Leuce-poplar clone experiments.

***Populus* sp. / biotechnology / in vitro / rejuvenation / selected clones / field trials**

**Kivonat – Leuce-nyárok mikroszaporítása és homokterületen történő termesztésük kiértékelése Magyarországon.** A Leuce-nyárok őshonos állományalkotó fajok Magyarországon. Néhány fajukat, illetve szelekciójukat parkokban, utcák, utak mentén díszfaként használják. Magyarországon az összes erdőszült területnek kevesebb mint 4%-át teszik ki (70 000 ha). A Leuce-nyárokhoz tartozó fehér (szürke) nyár jelentős szerepet játszik a homoktalajok megkötésében, azok erdősítésében, és természetvédelmi szempontból is fontos. Száraz termőhelyi viszonyokra szelektált, gyorsan növő Leuce-nyárok hosszútávú nemesítési munkája folyik a - korábban Erdészeti Tudományos Intézet, ma már - a Nemzeti Agrárkutatói és Innovációs Központ Erdészeti Tudományos Intézetében (NAIK-ERTI). Magyarországon fás növények esetében in vitro szaporítási eljárásokat főleg a gyümölcsstermesztésben alkalmaznak, az erdészeti kutatásban elsősorban a szelekciós nemesítés során használják. Jelen tanulmány rövid áttekintést nyújt a különböző nyár fajok legfontosabb biotechnológiai vonatkozásairól, a kapcsolódó mikroszaporítási kísérletekről, valamint mikroszaporítással előállított Leuce-nyár klónok termesztési kísérleteinek eredményeiről.

***Populus* sp. / biotechnológia / in vitro / rejuvenilizáció / szelektált klónok / terepi kísérletek**

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## 1 INTRODUCTION

Inside the section of Leuce poplars, the white poplar (*Populus alba* L.), and its most important natural hybrid the grey poplar, (*Populus* × *canescens*) are native to Hungary. The area of native poplar stands and plantations was about 77 000 ha in 2012 (4.0 per cent of the total forested land), with a standing volume of 13.0 million m<sup>3</sup> (169 m<sup>3</sup> ha<sup>-1</sup>).

The NARIC Forest Research Institute is the most important national institution for poplar breeding and improvement. Investigations of selected individuals and populations of native poplars laid the foundation for their in situ and ex situ conservation.

Clonal selection is very effective on additive and non-additive genetic components. Asexual propagation is extremely important for the conservation of selected genotypes. Traditional vegetative propagation methods, like root cuttings and greenwood cuttings, are acutely limited, especially at the beginning of the production. The role of micropropagation can be important at this stage. This was started in the previous Research Institute for Fruitgrowing and Ornamentals in Budapest, which is now known as the NARIC Fruitculture Research Institute.

Poplars were one of the first objectives of in vitro propagation trials (Gautheret 1934, Mathes 1964, Winton 1968, 1970, 1971, Wolter 1968, Chalupa 1974). Cambial tissues were used as basic material in these experiments. First callus improvement, then shoot or root development was induced on the callus surface. Sometimes it occurred without any outside effect. The connection between the regenerated shoots and roots was occasionally unclear. The collected opinions founded the vegetative propagation established from a single bud and different originated callus based plant regeneration (Noh – Minocha 1986).

Whitehead and Giles (1977), Christie (1978), and Ahuja (1983, 1984) micropropagated sterile bud originated plantlets of poplars for the first time. Difficulties of culture establishment and genetically determined differences between the species were pointed out in the presentation of their results. Success of establishment also depends on the age of the mother plants.

Development of micropropagation methods of poplars for commercial purposes was the aim Barocka et al. (1985). Medium optimisation was continued by Chun et al. (1986). Wann et al. (1988) improved the micropropagation method of the tetraploid clone 'Ta – 10' for the substitution of vegetative propagation by grafting. Development of the micropropagation procedure for different poplar species/clones continues today. Coleman – Ernst 1990, Iordan-Costache et al. 1995, Zhang et al. 2000, Noël et al. 2002, Phan et al. 2004, Kang et al. 2009, Thakur et al. 2012 summarised the multiplication procedure of several poplar species that were inoculated or not inoculated with ectomycorrhizal fungus for different experimental purposes.

As the use of micropropagated plantlets became more and more popular, questions of genetic uniformity arose. Rahman – Rajora (2001) used microsatellite analysis to examine a phenotypic homogenous population of *Populus tremuloides* originated from a single mother plant produced by micropropagation. No difference could be detected in 8 out of 10 loci, but differences were observed in the cases of 2 loci. These results offer the possibility of genetic modification during the micropropagation procedure, without any phenotypic change. Genetic uniformity of the traditional methods of propagated plants (grafting, cuttings) was not controlled in the same time.

Breeding work based on in vitro explants started almost parallel with the development of an in vitro mass-propagation procedure for poplars. Protoplast and cell suspension production followed by plant regeneration were the aims of Douglas's (1982) and Ahuja's (1983) research work. Savka et al. (1985) improved the efficacy of breeding methods with early stage embryo isolation and nurturing under in vitro conditions. In the nineties, regeneration trials

for breeding purposes began (Son – Hall 1990, Altman 2003, Confalonieri et al. 2003, Häggman et al. 2007, Yadav et al. 2009, Maheshwari – Kovalchuk 2011) followed with publications on agrobacterium mediated transformation (McCown et al. 1991, Leplé et al. 1992, Song et al. 2006, Nishiguchi et al. 2006).

White poplar selected clones were micropropagated in the laboratory of NARIC Fruitculture RI in collaboration with the NARIC Forest RI. Plant tissue culture methods provide us with new means to speed up the vegetative propagation of selected clones and give us the opportunity to establish new clone experiments and seed orchards (Rédei – Balla 2007).

## 2 MATERIALS AND METHODS

### 2.1 Plant material for micropropagation trials

All the clones/cultivars originated from the clone collection of NARIC Forest RI:

*Populus alba* clones, number: 101. 'H 337' (*P. alba* × *P. grandidentata*),  
103. 'H 384' (*P. alba* × *P. grandidentata*),  
109. 'H 425-4/1' (*P. alba* × *P. alba*),  
111. 'H 425-4' (*P. alba* × *P. alba*),  
112. 'K-1' *Populus hupehensis* and  
114. 'K-2' *Populus alba* originated from China.

#### *Plant material and growing site for the field evaluation*

In the clone trial, clone 101 'H 337' (*P. alba* × *P. grandidentata*), 103 'H 384' (*P. alba* × *P. grandidentata*), 104 'H 325' (*P. alba* × *P. grandidentata*) and 111 'H 425-4' (*P. alba* × *P. alba*) and *Populus* × *canescens* seedlings can be found as controls. The experiment was set up in the spring of 2004 with one-year-old micropropagated plants (for the clones) and one-year-old seedlings (for the control). A randomised block system with three replications was used. The initial spacing was 2.5 × 2.0 m. In every replication, 30 plants were planted. The total experimental area was 0.43 ha. The one-year-old seedlings were produced by the Kiskunsági Forestry Joint Stock Company.

The experiment itself was allocated in subcompartment Kecskemét 40A in the Danube - Tisza interflow region (in central Hungary). The main ecological characteristics of the studied area are: forest steppe climate zone; humidity of the air is less than 50% in July at 2 pm; during the period at the test area the annual precipitation is between 263.8–560.3 mm; hydrology: free draining; genetic soil type: humid sandy soil with very shallow rootable depth. The latitude and longitude coordinates of the experimental area are *N* 46.883547, *E* 19.588868.

### 2.2 Micropropagation

Micropropagation is a useful method for breeders, because a large quantity of uniform plants can be produced in a short time from a single tree or bud.

Steps of the micropropagation method:

1. Establishment of sterile in vitro culture
2. Improvement of multiplication procedure in a growth - room of 22 °C, 1500 Lux light intensity and 16/8 photoperiod.
3. Development of rooting
4. Acclimatization to greenhouse conditions
5. Adaptation under field conditions
6. Plantation

## 2.3 Field evaluation

### *Tree classification on the field*

Characterization of stem quality, including their health condition, was defined by using the following stem quality classes:

*Class 1* – The stem is straight, cylindrical, healthy and reaching the top of the crown. Crooks are tolerated in one dimension only, up to a bend of less than twice the stem diameter. The lower two-third of the bole is free of live branches.

*Class 2* – The stem is straight and healthy, forks are tolerated, but only if they are in the uppermost third of the tree. Crooks are tolerated in one dimension only, up to less than 4 times the stem diameter.

*Class 3* – The stem is crooked, leaning and more or less damaged. Crooks may reach 6 times the stem diameter in one dimension and minor crookedness in a second dimension is tolerated.

*Class 4* – The stem is very crooked in more than one dimension and heavily damaged. Low branching, forked trees sometimes with broken crown.

The stem quality index was estimated by the arithmetic average of the stem quality classes.

## 3 DESCRIPTION OF THE METHOD USED AND DEVELOPED FOR MICROPROPAGATION

### 3.1 Establishment of sterile culture

Three different explants were used for the establishment of a sterile culture of poplar clones:

1. Winter buds were collected in February. After a preliminary disinfection, buds were forced flushing in a thermostat with high relative humidity under artificial light. Shoots developed under controlled conditions were disinfected and planted into growing medium.  
This method was found to be successful by disinfection in the case of white poplar. Most of the shoots became sterile, but they stopped growing under in vitro conditions and died after a short time.
2. Shoots developing in springtime, following a careful disinfection, grow quickly.
3. Woody cuttings are not sensitive for disinfection, but often stop further development under in vitro conditions.

Actively growing shoot-tips of a young tree are suitable for culture establishment. In the case of breeding, when the mother plants are about 30 years old or older, the yearly growth is usually only a few millimetres. The early closing buds begin to grow only after a long rejuvenation period.

### 3.2 Rejuvenation

*Rejuvenation can be carried out under 1) in vivo or 2) in vitro conditions.*

- 1) In vivo rejuvenation means involving horticultural methods into the procedure of establishment:
  - Rooted cuttings originated from the selected mother tree are grown in a greenhouse (*Figure 1*), with regular plant protection, they are prepared for the establishment (*P. alba* clones number: ‘109’ and ‘111’).
  - Graftings are used as explants ( $P \times$  euramericana cv. ‘Kopecky’).

In vivo rejuvenation is difficult, because any kind of traditional vegetative propagation with adult trees is hard work.



Figure 1. Forced flushing of *Leuce-poplars* for in vitro establishment

- 2) Rejuvenation in vitro means that the sterile cultures are transferred to fresh medium daily at the beginning, later weekly as far as the fenolic compounds differ into the medium and decrease their inhibition for development (Figure 2). Growth regulators induce flushing at the same time (*P. alba* clones number 101, 103, 112, 114).

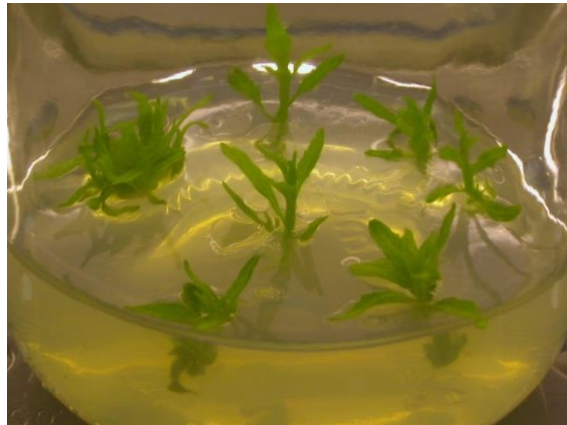


Figure 2. The sterile cultures start to grow

### 3.3 Multiplication

Experiments for propagation under in vitro condition start when enough sterile culture is prepared. All the cultures are grown in a culture room under controlled conditions: 22 °C, 16/8 day/night photoperiod, 1500 Lux warm white light intensity.

Medium experiments are based on MS (Murashige – Skoog 1962) and WPM (Lloyd – McCown 1980). Poplar shoots are very sensitive to the nitrogen form and concentration as well as for the added growth regulators. They have to be determined clone by clone.

The multiplication rate is typical for the species, clones/cultivars. With poplars, 5–8 new shoots develop monthly. The typical propagation rate can be detected 3 to 5 months following establishment when the cultures were adapted to the in vitro conditions. The developed new shoots isolated from the mother plants are the base of the next propagation phase. The multiplication phase has to be repeated till the required shoot number for rooting is produced. The shoots growing on the propagation medium are elongated and suitable for rooting (Figure 3-4).



Figure 3-4. Leuce-poplars in the propagation phase

Vitrification often occurs in white poplar cultures causing a depression of the multiplication rate. Keeping the concentration of major and minor elements as well as growth regulators at a low level can help the cultures avoid these plant-physiological abnormalities. The vitrified cultures have to be eliminated.

### 3.4 Rooting

Spring time, from February until May, is the best period for root induction.

15–20 mm high, 3-week-old shoots can be rooted in half-strength major and minor elements, reduced sugar containing medium supplemented with a low concentration of auxine.

Rooting rate of *P. alba* clones is about 80–90 percent (Figure 5-6), *P* × *euramericana* cv. 'Kopecky' about 60 percent.

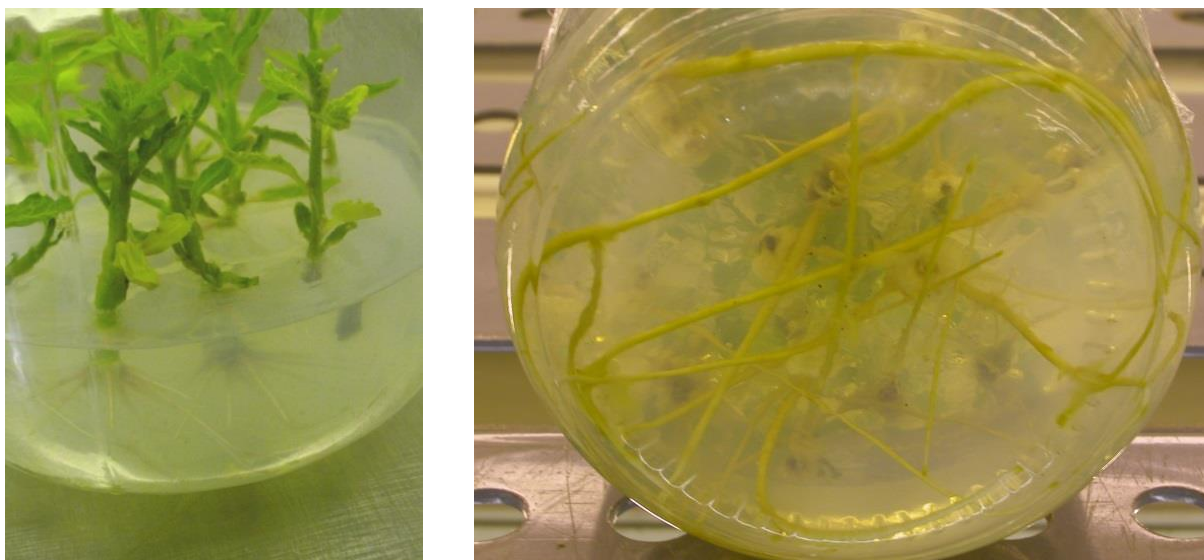


Figure 5-6. Rooted Leuce-poplars in vitro

During weeks 3–4, the roots develop and growth of the plantlets achieves the size appropriate for acclimatization (Figure 7).



Figure 7. Rooted poplar plantlets ready for acclimatization

### 3.5 Acclimatization

Acclimatization means the period (4–6 weeks) where the plantlets – developed under sterile conditions – are adapted to semi-sterile greenhouse conditions. This is one of the most difficult parts of the micropropagation procedure. Spring is the best time for acclimatization, because days grow longer and the greenhouse temperature can be kept between 15–30 °C.

The quality of the substrate has to be chosen to fit the requirement of the plant species. With trees, usually fibre, – reach substrate with high water capacity and a pH of about 6–6.5 can be used. Nutrition can be contained in the substrate or can be added together with the water supply.

Following the transplantation of plantlets into the substrate, a relative humidity of 90-95% has to be ensured for them until the stomas begin to work and the development of plant skin is finished (about 10 days). After then the relative humidity can be reduced to the normal greenhouse level. Protection against direct sunlight is also important during the whole procedure.

Attention must be paid to plant protection, mainly against fungal infection. By the end of the acclimatization, the plantlets grow up to 15–20 cm (Figure 8), which is a good size for transplantation under shaded and irrigated nursery conditions (Figure 9).



Figure 8. Acclimatization of different poplar clones in the greenhouse



Figure 9. Growth of the white poplars in the nursery garden

Plantlets acclimatized in the mentioned period in the first growing season grow to the size fit for grafting in August–September in the case of rootstocks, or for reforestation in October–November in case of forest-trees (Figure 10, 11, 12).



Figure 10. Micropropagated H-325 *Leuce poplar* clone in the nursery garden



Figure 11. Growth of the H-384 clone in the nursery



Figure 12. H-337 white poplar clone in the nursery

Hot summer and dark winter periods are not fit for acclimatization. Tree plantlets, acclimatized in autumn are difficult to keep during the winter period, so it is not suggested.

#### 4 RESULTS

The evaluations have proved that the clones *P. alba* × *P. grandidentata* H-337 and H-384 are the most suitable for poplar growing under unfavourable ecological factors, while the clone *P. alba* × *P. alba* H 425-4 could be considered as an alternative for wood production. The abovementioned promising clones may also be planted in roadside plantations and parks because of their decorative value.

Table 1 shows the most important stand structure parameters of the *Leuce-poplar* clones at the age of 8 (KecsKemét 40A). Based on the data concerning the growth in height, clones H-337 and H-384 provided the best results. They surpassed the control by 51% and 22% respectively. Regarding the growth in DBH (diameter at breast height) the abovementioned clones surpassed the control by 30% and 25%; the tendency was also the same with mean tree volume values. The effect of differences in DBH on the mean tree volumes seems to be very considerable (an additional 102% and 49% for the above-mentioned clones). Where the stem quality index is concerned, the succession from best to worst is: H-337, H-384, H 425-4, H-325 and the *control*. According to the significance test at  $P = 5\%$  level, significant differences

were found in height ( $SD_{5\%} = 2.07$  m), in DBH ( $SD_{5\%} = 2.11$  cm) and in the mean tree volume values ( $SD_{5\%} = 21.2$  dm<sup>3</sup>) (Table 2).

Table 1. Some yield data and stem qualification of Leuce-poplar clones at age of 8 years (Kecskemét 40A, 2011)

Name of clone	Mean height		Mean diameter at breast height		Mean tree volume		Stem quality index (1–4)
	(m)	%	(cm)	%	(dm <sup>3</sup> )	%	
H-325	8.05	111	7.40	102	25.8	102	2.14
<b>H-337</b>	<b>10.95b</b>	<b>151</b>	<b>9.44b</b>	<b>130</b>	<b>51.3b</b>	<b>202</b>	<b>1.33b</b>
<b>H-384</b>	<b>8.84</b>	<b>122</b>	<b>9.08</b>	<b>125</b>	<b>37.9</b>	<b>149</b>	<b>1.61b</b>
H 425-4	7.66	106	7.82	108	26.6	105	1.91
<b>Control</b>	<b>7.24a</b>	<b>100</b>	<b>7.26a</b>	<b>100</b>	<b>25.3a</b>	<b>100</b>	<b>2.28a</b>
<b>SD<sub>5%</sub></b>	<b>2.07</b>		<b>2.11</b>		<b>21.2</b>		<b>0.52</b>

Between the values marked by different letters significant differences were found at  $P=5\%$  level.

Table 2. ANOVA values according to the investigated parameters.

	Height			
	SS	DF	MS	F
Total	50.93	20		
Replication	3.43	2		
Treatment	30.82	6	5.1366	
Error	16.68	12	1.39	3.69
$SD_{5\%} = 2.07$				
	DBH (diameter at breast height)			
	SS	DF	MS	F
Total	32.21	20		
Replication	2.32	2		
Treatment	12.48	6	2.08	
Error	17.41	12	1.4308	1.43
$SD_{5\%} = 2.11$				
	Mean tree volume			
	SS	DF	MS	F
Total	0.003774	20		
Replication	0.000307	2		
Treatment	0.001761	6	0.00029	
Error	0.001706	12	0.000142167	2.06
$SD_{5\%} = 21.2$				
	Stem quality			
	SS	DF	MS	F
Total	3.270000	20		
Replication	0.200000	2		
Treatment	1.890000	6	0.3150	
Error	1.180000	12	0.0983	3.20
$SD_{5\%} = 0.52$				

$P < 0.05$

The collected data were analysed using the STATISTICA 8.0 (data analysis software system -StatSoft, Inc. 2008) programme including correlations and regression analysis.

The annual ring evaluation analyses show that selection surplus can also be detected under marginal site conditions. According to the average annual ring width (growth in diameter at breast height), clone H-384 surpassed the control by 52.7% while clone H-337 surpassed it by 46.2% (Figure 13).

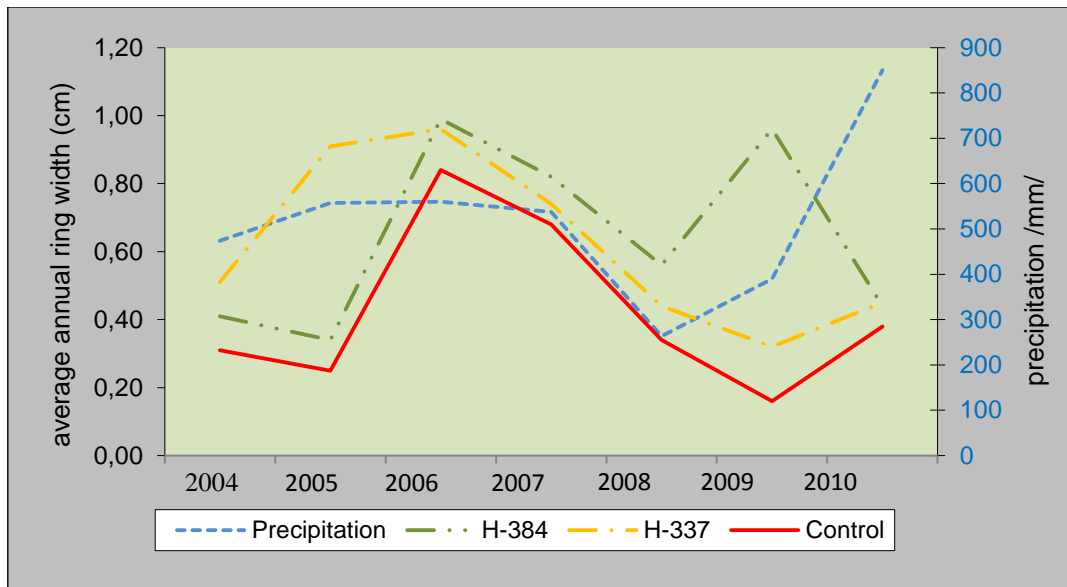


Figure 13. Annual ring analysis of the 2 most promising *Leuce-poplar* clones at the age of 7 (Kecskemét 40A, 2010)

We have done discriminant analysis considering the DBH (diameter at breast height), height, rate of growth, stem quality, and health condition as quantitative variables – according to the different clones – as observation groups. Figure 14 shows the ordination diagram based on canonical discriminant functions of the variety comparison clone trial.

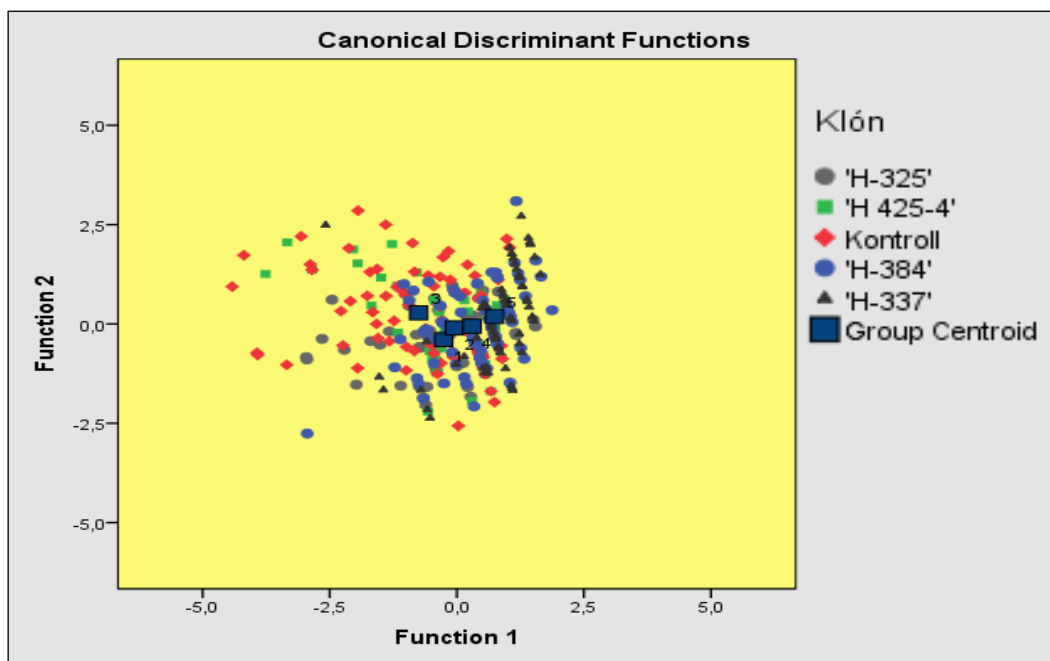


Figure 14. The common ordination diagram of the 5 groups (clones) on the basis of the investigated characteristics

In the course of the discriminant analysis it can be said that if the number of groups are more than 2 – as in this case – the discrimination of cases (variables) belonging to the single groups (clones) can be done with the help of so-called canonical variables.

*Figure 14* indicates – though not clearly – that the groups separate from each other considering the first function (horizontal axis). The figure also shows that the control (commercial) white poplar specimens show the highest deviation according to the investigated characteristics compared to the other clones.

## 5 DISCUSSION

The H-384 and H-337 clones presented the most homogeneous distribution. These results also prove the excellent stem, form, yield properties, health condition, and the outstanding rate of growth of these 2 promising *Leuce-poplar* clones (Keserű 2013).

In spite all of the efforts to reduce the cost of the micropropagation, using micropropagated plants for reforestation seems to exorbitantly expensive given present economic circumstances. Today a micropropagated plant costs 2 EUR, while the price of a commercial seedling is much lower (cca. 0.1 EUR). In Hungary and also in other countries (Haapala et al. 2004) mother plantations are established from micropropagated plants and from there trees for forestation are propagated year by year using conventional vegetative propagation methods, usually by cuttings (Lubrano 1992, Confalonieri et al. 2003).

Among the forest species, poplars belong to a well-known group and can be used as a model tree for other species (Taylor 2002). The production of clones that grow quickly in shorter periods of time, are adaptable to different sites like poor, polluted soil or drought conditions, and can yield more wood is currently ongoing through the application of biotechnological methods that are based on micropropagation (Lubrano 1992, Giri et al. 2004, Doty 2008, Chen – Polle 2010). Experiments on the production of genetically modified resistant clones against fungal and lepidopteran infections using *Agrobacterium* mediated transformation methods are improving (Zhang et al. 2005). The important role of poplars among the energy tree species must be stressed (Polle – Douglas 2010, Sanningrahi et al. 2010).

## 6 CONCLUSIONS

The early evaluation demonstrated in the paper showed that the micropropagated clones H-337 and H-384 seem to be suitable for poplar growing under unfavourable site conditions, while clone H 425-4 could be considered as an alternative for wood production. The experiments have also demonstrated that micropropagated plants can be successfully transplanted into soil, hardened, and grown in the field. Micropropagated trees have been exhibiting normal growth and appearance since they were planted. Hungary has a great deal of experience growing white poplars. A selected clone that has a really high growth capacity, as is demonstrated in the present publication, should be used for reforestation or ornamental purposes despite the higher cost.

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# Multi-Frequency Electrical Impedance Measurement on a Wooden Disc Sample

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**Abstract** – Non-invasive measurement techniques are widely used for the investigation of living tree and wood material. Principles and methods from other fields of science form the basis of these techniques. Electrical impedance measurement, which is adopted mainly from applied geophysics, plays an important role in the investigation of living trees and wood material. Unlike other measurement techniques that provide mainly physical information, electrical impedance measurement is able to gather either physical or chemical information. There are several published accounts of the use of Electrical Impedance Tomography mainly for examining internal decay, estimation of sapwood, and heartwood width. The advantages of the method are its low costs and rapid execution; however, low spatial resolution is still one of the most glaring limits of the method. Use of a new, self-developed, high resolution measurement system provides new opportunities to gain better insights into the characterisation of wood material. As well as high accuracy and resolution, the measurement system is able to operate in a wide frequency range. This expands the examined parameters making it possible to extend the gained information and develop new methods for the investigation of living trees and wood material. The aim of our publication is to introduce some experimental results in the investigation of spatial resolution our system measured on a wooden disc.

**multi-frequency Electrical Impedance Measurement / spatial resolution / wood disk**

**Kivonat** – Fakorongok multi-frekvenciás elektromos impedancia mérése. Az él fák és faanyagok vizsgálatához különböző mérés-technikai eljárásokat alkalmaznak. Ezek alapját elsősorban más tudományterületekről hozott elvek és módszerek képezik. Az elektromos ellenállás (impedancia) mérés, melyet a geofizikai gyakorlatból adoptáltak, ezek között nem kevésbé fontos szerepet tölt be, ugyanis míg a mérési módszerek többsége szerkezeti, fizikai információt szolgáltat, addig az elektromos módszerek a fizikai mellett kémiai információszerezésre is alkalmasak. Az Elektromos Impedancia Tomográfiát több ízben publikálták él fák esetében üregek, illetve szíjács és geszt vastagság becslésre. Bár a mérési eljárás elnevezés közé sorolható a relatív alacsony költségigény és gyorsaság, összevetve azonban más képalkotó eljárásokkal, a kisebb térbeli felbontás szab határt az egyes alkalmazások elterjedésének. Egy új, saját fejlesztésű nagy felbontóképességű mérési eszköz alkalmazása új lehetőségeket nyújthat a felbontás javításában és a faanyag jellemzésében. A mérési eszközre jellemző, hogy a nagy pontosságú és felbontóképességű méréseket igen széles

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frekvencia-tartományban képes kivitelezni. Ez egy új dimenziót nyit meg az él fa és faanyag vizsgálat módszertanában, mely egy új mérési eljárás fejlesztését alapozhatja meg. Jelenlegi publikációnk célja mér eszközünk térbeli felbontásának kísérleti ellen rzése faanyagon.

### **multi-frekvenciás elektromos impedancia mérés / térbeli felbontás / fakorong**

## **INTRODUCTION**

Non-destructive measurement techniques play a crucial role in the evaluation of living trees. These methods are developed mainly from imaging techniques used in applied geophysics (Szalai et al. 2002) and medical diagnostics. Imaging techniques from the adaptation of seismic methods are the most improved. (Divos et al. 1994) These measurements are made by using acoustic waves. The result of acoustic tomography is generally an approximation of the spatial distribution of wave velocity in the investigated tree. There are several other non-destructive imaging methods published mostly for the detection of decay in trees. (Divos et al. 1991, Nicolotti et al. 2003, Bucur 2003) These investigation methods are X-ray, gamma ray, computer tomography, nuclear magnetic imaging, thermal techniques, ground penetrating radar, microwave methods and electrical tomography.

Electrical tomography methods are used in geophysical sub-surface explorations by performing on-surface measurements. As an early woodmaterial investigation method, electrical resistivity measurements were published in (Shortle et al. 1997). The researchers (Shortle et al. 1997) examined the spatial distribution of electrical resistance in a wooden disc. The authors performed the published measurements on samples taken from the wooden disc. Later, other authors published electrical measurement methods applied to several goals, for example finding discoloured wood, red-heart in beech (Weihs, 2001), detection of decay in trees (Dubbel et al. 1999; Rust et al. 2007, Brazee et al. 2011); estimation of sapwood and heartwood width (Bieker et al. 2010.); variation of electrolytes in *Quercus robur* (Bieker et al. 2010.). For these purposes, Electrical Resistance Tomography (ERT) as a non-invasive measurement technique was used. In the case of these investigations, the measurements and evaluations were performed with single frequency measurements.

In multi-frequency Electrical Impedance Tomography, we create an electrical field in the segment of a living tree that is to be investigated. This field depends on spatial distribution of admittance (or impedance), which is characteristic for the tissue of the tree. Admittance (reciprocal of impedance) is a physical quantity, which measures how easily an investigated material allows the injected current to flow through it. The electrical field can be characterised by the measuring of electrical potentials on the boundary of domain. Through data collection and evaluation, the spatial distribution of admittance (or impedance) can be approximated. The speciality of mFEIT is the injected current, which is a monochromatic sine wave. Thus the measured material can be characterised in time and frequency domain. (Woo et al. 2007) Testing in time domain includes the EIT in classical meaning. By adding the measurements in frequency domain, we can measure the transfer function of the object. Thus we gain a more extended evaluation method for the investigation of living trees and wood materials that includes the measurement of spatial distribution of admittance (or impedance) depending on the frequency. The investigated structure responds dissimilarly in every frequency point.

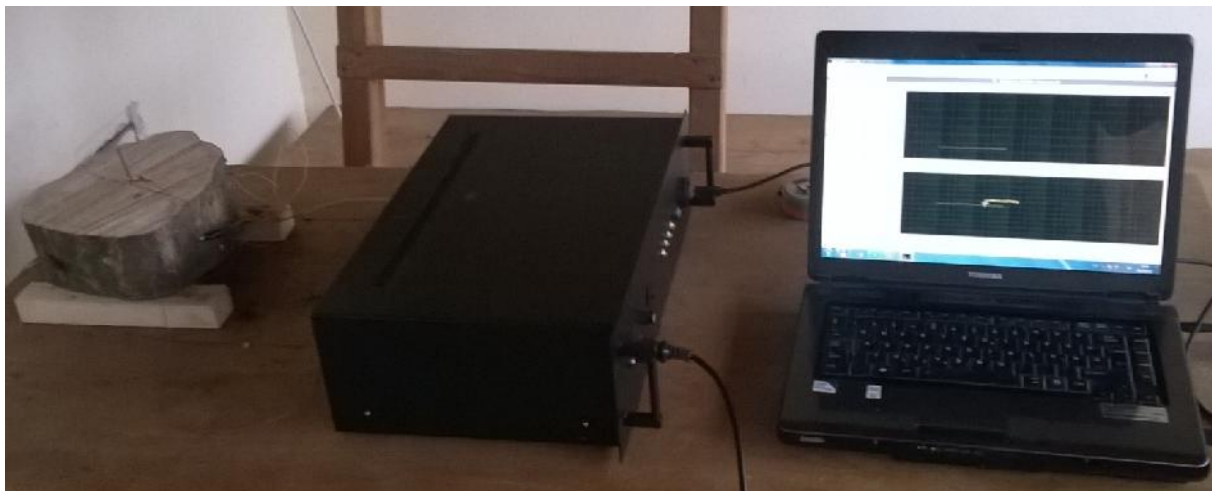
The aim of mFEIT is to locate several types of anomalies in living trees in the best spatial resolution possible. To achieve this, equipment with high resolution and an appropriate electrode configuration is essential. A publication of Beck - Williams (1996) compares the advantages and disadvantages of several techniques applied in industrial tomography.

According to their statements (1996), EIT is a relatively cheap, quick, suitable, and simple imaging technique; but there is a need for research in the refinement of spatial resolution which is relatively small when compared to other techniques. The spatial resolution of EIT is determined as the ratio of diameter of the smallest object that can be located and the average diameter of material in which the measurements are made. According to Beck - Williams (1996), the EIT spatial resolution is about 10%, the worst imaging technique in industrial tomography. A spatial resolution of 10% means that EIT as an imaging method has the capability to localize a 20 mm object (anomaly) in a 200 mm diameter sample.

## MATERIALS AND METHODS

The measurement system consists of a high precision generator and a measuring board. The generator, which can be operated as a current or voltage generator, provides a low-distortion monochromatic sine wave for the excitation of a measured object in a large frequency range, between 20 mHz and 90 kHz. The measurement board is in general a data acquisition system that measures the real and imaginary part of complex voltages with 48 bit resolution. Amplitude measurement accuracy is 1 ppm and phase measurement accuracy is  $0.01^\circ$ . The system can execute spectral (Fourier spectrum of signal), spectroscopic, and tomographic measurements. With wobbler function it is possible to record impedance spectrums and transfer functions. Tomographic measurements are realized with parallel measurements using 8 channels. By adding a multiplexer board, it is possible to extend the number of channels up to 64. Our primary aim is to develop a modern tomographic method, which is called multi-frequency Electrical Impedance Tomography (mfEIT), for the investigation of living trees using a measurement system with high accuracy and resolution. In our paper we demonstrate the results of the experiments dedicated to spatial resolution determination in wood material.

In order to verify the suitability of our measurement system for mfEIT measurement in living trees and wood material, we performed an experiment for spatial resolution determination. We measured wood samples several times – first without an anomaly and later with an artificial anomaly. A fresh (green) slice of hornbeam (*Carpinus betulus*) was chosen for the experiment. The height of the disc was 11 cm; the average diameter was 20 cm. The measurement setup can be seen in *Figure 1*.



*Figure 1. Measurement layout for experiments in hornbeam*

In the first case the log was measured without anomalies. These recordings were used as background information in the evaluation. Then an anomaly – a small hole – shown in *Figure 2*, was made in the sample. Later, additional holes were drilled in the hornbeam disk. The holes were placed on the line between the source electrodes; the distance of the negative pole of generator and the centre of the bore was 140 mm. The diameter of the first hole was 8 mm (4% of the sample diameter); this was increased to 10 mm (5%), 12 mm (6%), 16 mm (8%), 20 mm (10%).



*Figure 2. 8 mm diameter bore, drilled into slice of hornbeam for experimental measurements*

In this study we applied only 4 electrodes in addition to a ground electrode which was located at the centre of the disk. In the case of in-situ measurements, we will place the ground electrode in the soil, near the tree. The applied electrode configuration is shown in *Figure 3*.

The electrode configuration shown in *Figure 3* is called a null array in geophysical practice (Szalai et al. 2002), which indicates some types of anomalies very sharply. If the sought anomaly doesn't exist, the measured values are minimal corresponding to the inhomogeneity of measured material. When an anomaly can be found in the investigated material, the measured potential values respond with very sharp changes. *Figure 3* shows that the current generator was connected to the source electrodes (1 and 2) and the potentials were measured on electrodes 3 and 4. The lines, linking the source (1 and 2) and measuring (3 and 4) electrodes were perpendicular. That results in the appropriate sensitivity that is characteristic for null arrays.

We measured the complex voltages on the source and measuring electrodes by applying a current generator. For calculations we applied the complex voltage measured on source for reference voltage. The measurements were done with swept sine in a range of 0.1 Hz – 90 kHz, in 100 frequency points. The voltages, measured on electrodes 3 and 4, are dissimilar to the reference voltages in every frequency point. This difference, caused by the measured material, can be characterized by calculating the attenuation of amplitudes and phase difference. (*Figure 4*).

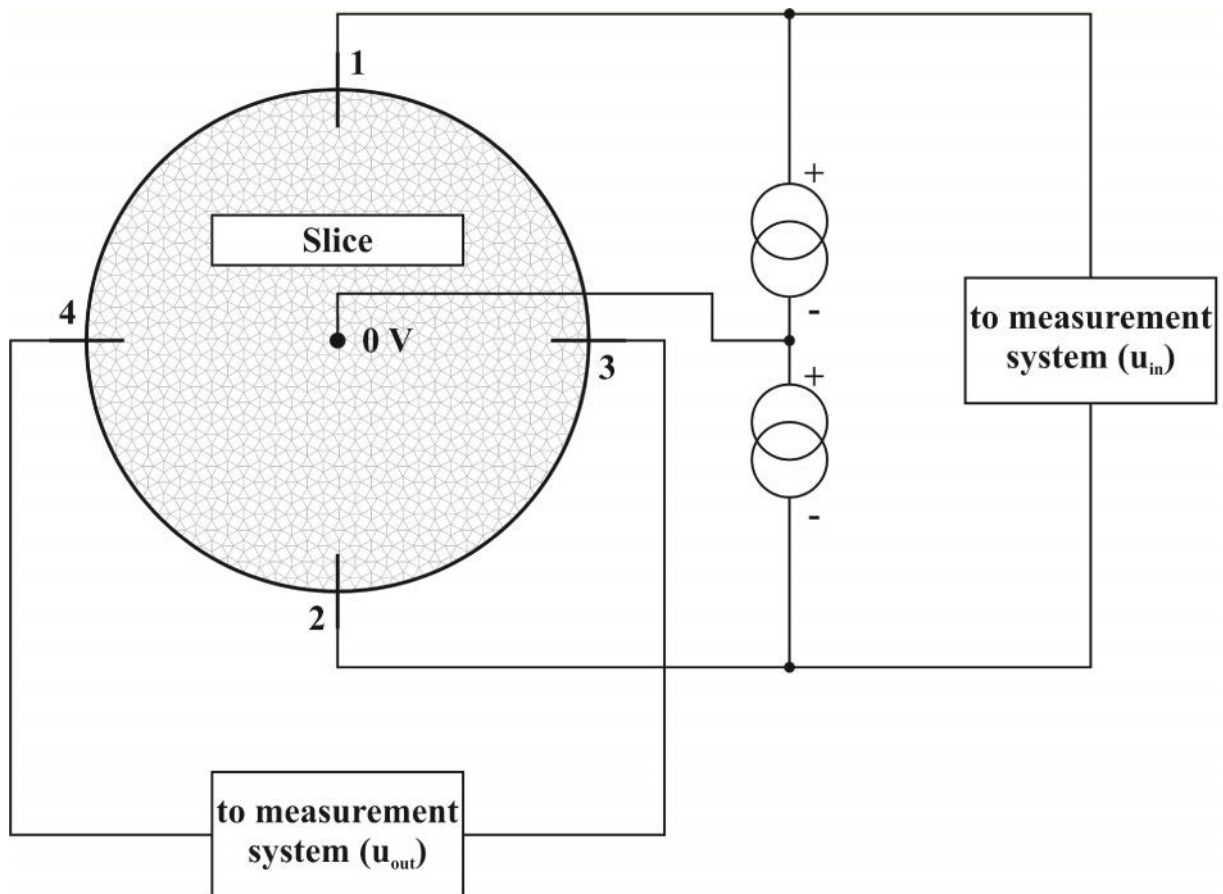


Figure 3. The applied electrode configuration

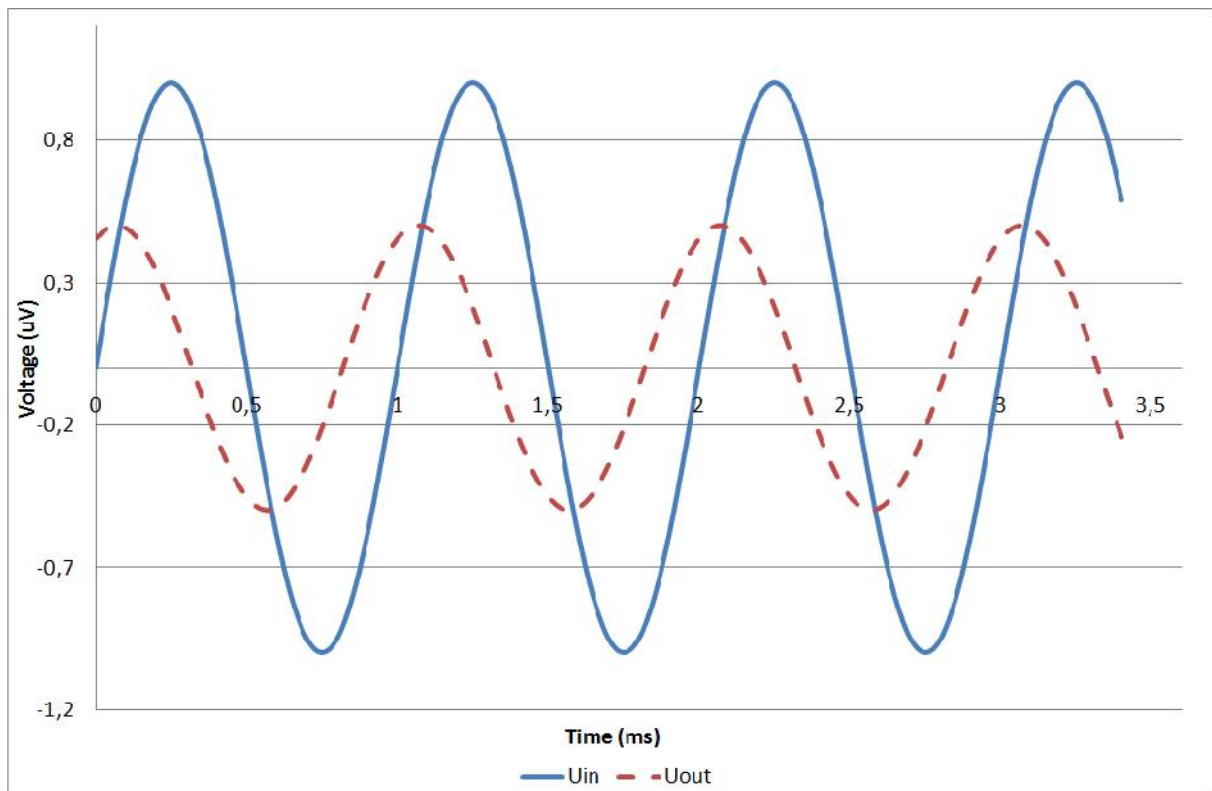


Figure 4. Illustration of the difference between the measured and reference voltages measured in linear material

The collected data is characteristic for the transfer function of the tested material. Thus the evaluation of collected data is possible in this way:

$$A = -20 \cdot \lg \frac{|u_{out}|}{|u_{in}|} \quad (1)$$

$$\phi = \phi_{in} - \phi_{out} \quad (2)$$

where

$A$  is the attenuation of signal in measured material (dB)

$|u_{out}|$  is the magnitude of voltage measured on electrodes 3 and 4

$|u_{in}|$  is the magnitude of voltage measured on source electrodes

$\phi$  is the phase difference

$\phi_{out}$  is the phase of voltage measured on electrodes 3 and 4 (°)

$\phi_{in}$  is the phase of voltage measured on source electrodes (°)

We assumed homogeneous electrical properties in the radial – tangential plane, although in reality, according to our experimental data, there is about a 10% relative difference between radial and tangential directions. At this stage we tried to ignore this effect, but in the future we will need to take this into consideration.

## RESULTS

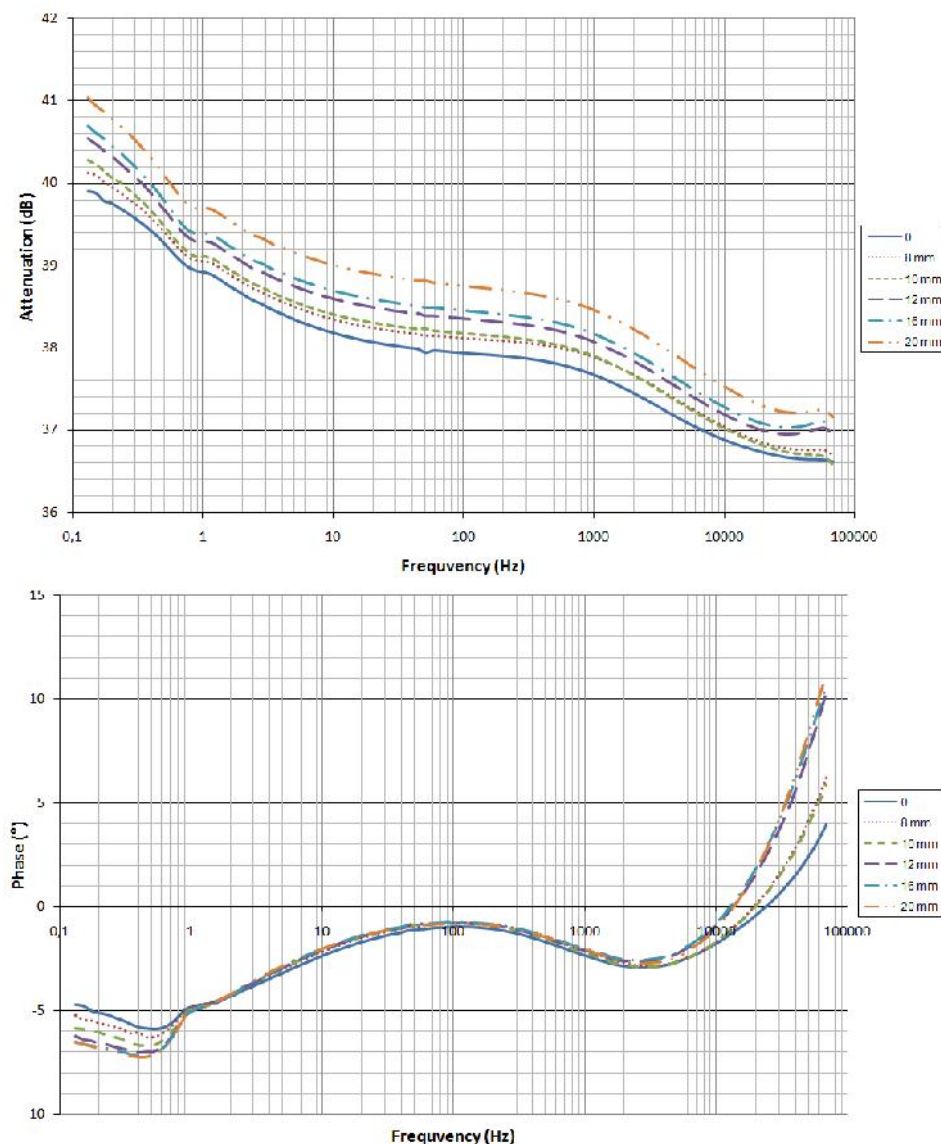
The first measurements were made in a log of hornbeam without artificial defect. The aim of these experiments was to gain information of the electrical stability of the measurement setup. These measurements were used as background data as well. For the characterisation of stability of the measurement set-up, single frequency recordings were made with 1 Hz, 5 s averaging time in 60 s lengths. Coefficients of variation (CV) were calculated. The CV values in general were better than 0,5% (at 1 Hz). Thereafter the transfer functions were measured. In every experimental case the transfer function was recorded 3 times. From these the average CV values were calculated, which were better than 0,05% for magnitude and better than 10% for phase (calculated on the total frequency range).

The measured transfer functions for all experimental cases are shown in *Figure 5*.

*Figure 5* shows the attenuation and phase versus frequency – called a Bode diagram. Both of these characteristics are specific for the investigated material. Thus, for the evaluation of the measurements, both of the curves have to be taken into consideration. The attenuation is represented in dB unit and the phase in degree. According to *Figure 5*, the attenuation curves have the same character. At low frequency, they have maximal values and they decrease monotonically. The shape of attenuation curves is specific for the material properties. The difference between maximum and minimum attenuation values is 3.29 dB in the case of measurement without bore. The phase diagrams are characteristic for the wood material. At low frequency the phase starts on the minimum values and then it increases until 100 Hz before it decreases and increases again. The dynamic range of phase alternation is 9.89°.

*Figure 5* clearly shows the differences between experimental cases. The hole drilled into the log in physical aspect is equivalent to placing a non-conductive media into a conductive. This coarse intervention should result in significant change in the measured attenuation. This can be seen in *Figure 5* as well. The smallest attenuation values were measured without an anomaly. Then with the addition of non-conductive media, the attenuation values increase

consequently. The experimental cases can easily be distinguished in *Figure 5*. The correlation of attenuation curves and the size of the bore can be clearly seen. The phase values indicate the anomaly clearly as well. By increasing the diameter of non-conductive media at low frequency, the phase is decreasing. Between 1 Hz and 4 kHz the phase curves have nearly the same values. Thus in this frequency range, the phase measurement isn't capable of distinguishing the diameter of an anomaly. At higher frequencies the phase curves become separated and they indicate the diameter of non-conducting media. Contrary to the change at low frequencies, at a higher frequency range according to the increasing diameter of an anomaly, the phase curves shift to higher values. It is evident that phase measurement provides new information about the investigation of trees and wood material. If an anomaly exists in investigated media, the measured phase curves will characterise the anomaly. Thus by combining the tomographic measurement with the recording of the transfer function of investigated tree or wood material, a characterisation of an existing anomaly can be gained.



*Figure 5. Transfer functions measured in hornbeam log without (0) and with non-conducting media of several diameters*

The obtained results clearly show that the multi-frequency approach offers more extended information about the investigated wood material than the traditional EIT, or ERT methods. Evaluating the attenuation of measured signals represents a possibility to draw conclusions about electrical properties, size and shape of an investigated anomaly. The published experiment shows that the attenuation curves (*Figure 5*) – according to the results performed without a bore – are in correlation with the diameter of bores: with the increasing diameter of a bore, the difference of actual and 0 curves is increasing. The phase curves (*Figure 5*) represent the extended information, which is a result of multi-frequency investigation. The 0 curve is characteristic for wood material. With the addition of an anomaly, a non-conductive media (bore), the shape of phase curves became distorted. This alteration is in correlation with material properties of the added anomaly. The phase curves can be examined in low, middle, and high frequency intervals because the response of curves is different in these intervals. This is an important opportunity for enhancing the selectivity of measurement.

Summarising the results: using the multi-frequency approach offers a possibility to develop a mEIT tool for tree evaluation with better – at least 4% – spatial resolution compared to the 10% reported in the literature, Beck (1996).

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## Guide for Authors

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## Contents and Abstracts of the Bulletin of Forest Science

Bulletin of Forest Science (Erdészettudományi Közlemények) is a new journal supported by the Hungarian Forest Research Institute and by the Faculty of Forestry of the University of West Hungary. The papers are in Hungarian, with English summaries. The recent issue (Vol. 5, 2015) contains the following papers (with page numbers). The full papers can be found and downloaded in *pdf* format from the journal's webpage ([www.erdtudkoz.hu](http://www.erdtudkoz.hu)).

### Vol. 5, 2015

Tamás MAJOR and Viktória CSANÁDY:

#### **Combined FEM-SPH simulation method for the modelling of the interaction of tillage tools and the soil ... 7-19**

**Abstract** – Earlier cultivation and tillage tools were designed on the basis of practical experience; testing and records of fundamental theoretical analysis of these tools is largely lacking. Today, the release of new equipment is always preceded by several years of laboratory tests and field experiments. By modelling the interaction between the soil and tillage tools, the time and costs of the development process can be significantly decreased. Thanks to the recent rapid advancement of high performance computers and numerical methods, it is possible to complete these model calculations more effectively now. With the help of a combined FEM/SPH simulation – in this case utilizing two different driving speeds of a tractor – we determined the magnitude of horizontal force acting upon the rotating tool as a function of time. Using the functions fitted to the simulation results, the authors have calculated the average and the maximum values of the horizontal force. Moreover, normal stress distribution in the soil has also been determined.

János RUMPF, Attila László HORVÁTH and Katalin SZAKÁLOSNE MÁTYÁS

#### **Tree utilization price revenue categories and quality classification of some trees and forest stands ... 21-41**

**Abstract** – The „Simplified grading selection method” (Szász és Burján 1975) developed decades ago by the Forest Research Institute (FRI) and founded upon an earlier range of selection methods and evaluations of the diameter at breast height for logging became obsolete and necessitated updating and modernization. This was adjusted to the prevailing current logging data in order to make valuable original data useful and useable for the design and evaluation of current logging practices. We developed a simple solution for this: the so-called two-step proportioning method. However, the use of FRI tables requires the need to know the quality of tree stands as well. For this purpose, we utilized the trunk rating system in the Forest Planning Guide, making some adjustments to suit the task at hand. The resulting selection structure multiplied by current average prices determined average revenue. Our

method proved suitable for different levels of aggregation including national, forest company, and forest subcompartment levels. The process can be considered as a „sales-centric quality classification” of stands. To calculate the overhead cost of logging operations, we utilized the suitable portion of the developed cutting organizational plan (Szasz, 1979), and named the modified version the "Sopron series analysis method".

Ferenc FACSKÓ

#### **The transparency of forestry companies in 2014 ... 43-53**

**Abstract** – The following report summarizes the results of a study which examined the level of transparency among state-owned forestry companies. Overall, it was discovered that value of forestry companies are around country average. The information content of forest company websites can be assigned to two categories: a wealth of information or very little information. It can be said that companies that do not meet their legal requirements are also more likely to provide little information or information of poor quality on their websites.

István BACH, Norbert FRANK, Beáta PINTÉR and Sándor BORDÁCS

#### **Changes in the production of reproductive material in forest management from the years 1982-2014 (Quo vadis forest reproductive material production?) ... 55-69**

**Abstract** – Over the past decades, the forest reproductive material sector has changed significantly in Hungary. Societal, economic, and political developments have decisively influenced forest management, the results of which are reflected in the statistical indicators of the production of forest reproductive material. Analyses of statistical data point out relevant trends and changes, especially in ownership structure, total number of nurseries, mean size of nursery area, and proportion of tree species produced. In the early 1990s the formerly dominant state-owned nurseries were replaced by privately-owned ones, and large or medium size nurseries were replaced by small or even micro size farms. Simultaneously, the mean area of nurseries decreased drastically for about 10 years. As a consequence of slight reduction in total number of nurseries, the mean area, as well as the total volume of reproductive material produced by each nursery, has increased slightly since the 1990s. In general, relevant modifications in forestry policy, such as the increasing importance of close-to-nature forestry, modified preferences in the use of non-autochthonous tree species, and essential changes in afforestation programs have significantly affected the production structure of the forest nursery sector as well. For example, the production volume of reproductive material of conifers is far less than it was in the 1990s, but the production volume of scattered broadleaves is far greater than it was in the 1990s. Further variations in the structure of tree species can be expected due to the varying needs of climate change.

Bence BÁRDOS, László NAHÓCZKI, Dénes MOLNÁR, Norbert FRANK, Zoltán KÖVESKUTI and Ádám FOLCZ

#### **Investigation of epicormic shoot growth of sessile oak in shelterwood cutting stands ... 71-83**

**Abstract** – This paper addresses the epicormic shoot growth of sessile oak (*Quercus petraea*) in natural regeneration stands which can have a strong effect on potential trunk quality at harvesting; the deterioration of trunk quality can cause significant economic losses for forest managers. During the course of our research, we investigated the epicormic shoot growth of sessile oaks in shelterwood cuttings in different ecological environments. We examined 487 specimens on 10 plots in 3 different Hungarian regions. The collected data were analyzed in relation to site conditions and silvicultural interventions. Our results show that epicormic shoot growth intensity is influenced by crown size and changes in environmental parameters.

Accordingly, it is recommendable to make more intensive cuttings in young stands in order to stimulate trees to grow larger crowns. Faster and more careful final cuts are also suggested. By using these principles, trunk quality deterioration caused by epicormic shoot growth can be mitigated.

Árpád SZALACSI, Szilvia VERES and Gergely KIRÁLY

**Gap cutting and its effects on the understory vegetation in the pedunculate oak-hornbeam forests of Szatmár-Bereg Plain (NE Hungary) ... 85-99**

**Abstract** – Lowland oak-hornbeam forests are one of the important sites of quality oak wood production. Consequently, this habitat has become a major conflict point between forest management and nature conservation in Hungary over the past 20 years. In order to develop a regeneration method based on natural processes, we tested the impacts of gap regeneration cuttings in three compartments in the Szatmár-Bereg Plain (NE Hungary). During the regeneration process, we found an insignificant number of invasive weeds in the gaps. Forest herbs were represented nearly uniformly in the gap parts variably exposed to the sun; the cover of native weed species was significantly higher in the central part of the gaps. Mixed tree species (especially hornbeam) have shown great vitality in the gaps. Thus, in order to ensure the natural regeneration of the pedunculate oak, protection against game damage and proper management are indispensable. Based on our observations, a recommended minimal starting gap size in lowland hornbeam-oak forests is 0.15 hectares. After they are created, further expansion of the gaps is necessary after 4-5 years. The final restoration subdivision (approx. 2 ha) can be reached in 2 steps within a span of 8-10 years. The forests created in this way are mixed and are of an appropriate vertical and horizontal structure.

Tivadar BALTAZÁR, Ildikó VARGA and Miloš PEJCHAL

**The impact of the European mistletoe (*Viscum album* L.) on woody host-plants: a study of the relationship between infection intensity and tree vitality ... 101-118**

**Abstract** – Our research aims were to examine the relationship between infection intensity of European mistletoe (*Viscum album*) and the physiological and biomechanical vitality of the potential host species. For this study 3039 individuals of nine host species (*Acer campestre*, *A. platanoides*, *A. pseudoplatanus*, *Crataegus monogyna*, *C. pedicellata*, *Juglans nigra*, *Robinia pseudoacacia*, *Tilia cordata* and *T. platyphyllos*) were examined; of these 1424 specimens were infected. The host trees are situated in the Castle Park in Lednice, Czech Republic. Based on our results, it can be concluded that there is a strong relationship between these three factors. The weakest relationship was observed in the case of field maple (12-17%) and the strongest relationship was in the case of the black walnut (32-39%). In spite of our findings, the exact role of tree vitality influencing mistletoe infection remains unclear.

Szabolcs SZANYI, Levente SZŐCS and Zoltán VARGA

**The zoogeographical and ecological characteristics of Macroheterocera fauna of the Bockerek forest reserve ... 119-128**

**Abstract** – The Bereg Lowland is the most humid and coolest part of the Great Hungarian Plain, and is also the richest in natural or close-to-natural forests. Among them, the Bockerek forest (between Tákos and Vámosatya) is one of the most important. It is a nature conservation area and a large portion is a forest reserve. Its condition needs to be continually monitored; thus, the Forestry Research Institute has been monitoring the forest with light traps for a considerable time. From this material we analysed the faunal list of the nocturnal macro-moths. Since we only used a taxonomic list of species from a 9-year period without

frequency data, we only surveyed the composition according to faunal types and ecological faunal components with special regard to significant pest species in forestry.

József VARJU and Ferenc JÁNOSKA

**Woody nutrient preferences and habitat use of the Eurasian beaver (*Castor fiber* Linnaeus, 1758) at the Moson Danube ... 129-144**

**Abstract** – Beavers (*Castor fiber*) inhabiting the Moson Danube region presently cause considerable damage to the forestry sector. In our study we discuss forest areas affected by the activities of the beavers. We marked out plots in economically significant forest cultures in riverside sectors inhabited by beavers along the Moson-Danube where we studied the beavers' nutrient preferences and habitat use based on the number and location of chewed trunks. We have identified positive preferences in the case of several tree and shrub species (*Corylus avellana*: 0.24 *Prunus padus*: 0.22 *Salix* sp.: 0.82) and we have determined the areas of these cultures that have been particularly affected. We found that 75% of the chewed trunks are within 10 metres of the riverbanks. As a result of our research, forests particularly sensitive to damage c