Physiological reaction and energy accumulation of dominant plant species in fir-beech ecosystems affected by air pollution

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Abstract

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Physiological reaction and energy accumulation of assimilatory organs of 4 dominant plant species were studied in fir-beech ecosystems (80-100-year-old stand) and parallel clear-cut area (10-year-old stand) in locality Hliníky situated in the buffer zone of the Slovenský raj National Park. Studied ecosystems are located on the area affected by human activities. The achieved results showed that the influence of stand climate and habitat conditions differentiated the measured characteristics. Significantly higher photosynthesis was observed in assimilatory organs of *F. sylvatica*, *R. idaeus*, *D. filix-mas* and *V. myrtillus* species in the fir-beech stand (range 5.79–16.10 µmol CO_2 m^{-2} s^{-1}) compared to the clear-cut area (range 4.54–12.89 µmol CO_2 m^{-2} s^{-1}). *V. myrtillus* and *F. sylvatica* species reacted sensitively with significantly lower values of stomatal conductance (0.24, respectively 0.26 mol m^{-2} s^{-1}) in clear-cut area; *R. idaeus* and *D. filix-mas* showed significantly lower values (0.57, respectively 0.29 mol m^{-2} s^{-1}) in the fir-beech stand. Reduction of Fv/Fm under physiological limit was found for *F. sylvatica* species growing in clear-cut area, where the file of habitat conditions was not probably optimal. Energy content in phytomass of studied plant species varied from 18,511 J g^{-1} (*R. idaeus*) to 20,551 J g^{-1} (*V. myrtillus*). Significantly higher was only the mean value found for *D. filix-mas* species growing in the fir-beech stand (19,049 J g^{-1}) compared to the clear-cut area (18,561 J g^{-1}).

Keywords

ash, chlorophyll fluorescence, energy, fir-beech ecosystems, photosynthesis, stomatal conductance

Introduction

Vegetation reacts sensitively to environmental pollution by growth slowing, production reducing, morphological leaf changes, mortality of sensitive species and reduction of species diversity. Pollutants after release from the source do not remain in the air without changes, physical changes are in progress (movement and distribution in space, turbulent diffusion, changes in the concentration by dilution and other) as well as chemical changes. Industrial pollution is the most serious threat because industry uses highly toxic contaminants and many of them are released to the atmosphere (Atkinson and Winner, 1990; Fiala et al., 1989; Vacek et al., 1999, etc).

On the basis of literature can be assumed that forest stands with its structure and canopy of trees are influencing and changing the ground vegetation and its biomass. Natural forests have strong effects on the composition and structure of plant species mainly because they are the result of changes in light availability (Ódor and Standovár, 2001). Significant disruption of phytocoenoses composition only occurs after the removal of their woody components (Kukla et al., 2003; Kellerová and Janík, 2011). In natural conditions there are often more stressors simultaneously (strong radiation, high temperature, deficiency of water). If the file of habitat conditions is not optimal, the photosynthetic efficiency of plants is reduced (Ammann et al., 1999; Shparyk and Parpan, 2004).

Studied fir-beech ecosystems are located on the area affected by human activities. The Central Spiš has been subjected to a long-term negative influence of mining, metallurgy and engineering industry. The air in forest ecosystems is polluted mostly by harmful substances produced by ore-working and wood-working industry (Kukla et al., 2011). The main components of the pollutants are sulphur (SO_2), nitrogen (NO_x), CO emission and also heavy metals.

With this background, the present study was undertaken with the objective to compare physiological characteristics (photosynthesis, transpiration, stomatal conductance, chlorophyll fluorescence) and energy accumulation of assimilatory organs of *Fagus sylvatica* L., *Dryopteris filix-mas* (L.) Schott, *Vaccinium myrtillus* L. and *Rubus idaeus* L. species in fir-beech stand and parallel clear-cut area, both located on the area affected by air pollution in the buffer zone of Slovenský raj National Park (Western Carpathians).

Materials and methods

Study plots

The research was performed in the first half of July 2012 on 2 monitoring plots situated in the buffer zone of Slovenský raj National Park (NP), Table 1. G1 – clear-cut area is represented by 10-year-old fir-beech stand; G2 is represented by 80-100-year-old fir-beech stand. Soil reaction of Dystric Cambisol on the G1 (pH_{H_2O} 5.1) is strongly acidic and G2 (pH_{H_2O} 4.2) is very extremely acidic. Values of C/N ratio in the upper layers of soils (0–5 cm) on both plots reach the value 10.3. The studied plots are situated in the cool climatic region, with the mean temperature in July, 12–16 °C. The mean annual temperature is 4–5 °C, and average annual precipitation reaches 700–800 mm (Miklós and Hrnčiarová, 2002).

Methods

The soils were classified according to World Reference Base for Soil Resources 1994 (Bedrna et al., 2000). Values of soil reaction were determined potentiometrically – using a digital pH meter Inolab pH 720. Total content of N and C was determined by NCS analyzer type FLASH 1112.

The forest ecosystems were classified according to Zlatník (1976) and the names of plant taxa were given according to Marhold and Hindák (1998). On monitoring plots (area about the size of 400 m^2), leaves of *Fagus sylvatica* (from the bottom third of the tree crown), *Dryopteris filix-mas* (leaves), *Vaccinium myrtillus* (green twigs) and *Rubus idaeus* (shoots growing from creeping root) were sampled. All plant samples were dried at 80 °C for 48 hours and homogenised with a Fritsch planetary micro mill (<0.001 mm).

The gas exchange – rate of photosynthesis (P_N), rate of transpiration (E), stomatal conductance (g_S) and

Table 1. Ecological characteristics of studied forest ecosystems in buffer zone of Slovenský raj National Park (Western Carpathians)

Locality		Hliníky	
Study plot		G1 – clear-cut area	G2 – fir-beech stand
Age		10	80–100
Altitude [m]		960	950
Geographical coordinates		20°32’07’’E 48°51’51’’N	20°32’12’’E 48°51’49’’N
Exposure		S	SW
Stocking	upper storey	0.6–1.0	0.5
	lower storey	–	0.6–0.7
Canopy	upper storey	60–100	50
	lower storey	–	60–70
Vegetation unit		*Abieti-Fageta inferiora*	
Parent rock		quartz conglomerates	
Soil subtype		Dystric Cambisol	
pH_{H_2O} in 0–5 cm		4.2	5.1
pH_{KCL} in 0–5 cm		3.3	4.1
C : N in 0–5 cm		10.3	10.3

the intercellular CO_2 concentration (ci) were measured on the upper surface of leaves (the middle part of the leaf blade) *in situ* using the portable gas exchange system *LCpro+* (*ADC BioScientific Ltd.*, Hoddesdon, Great Britain). This instrument measures the gas exchange based on method of an open system (Šesták et al., 1966; Holá et al., 2010). Measurement time was set according to the work by Tucci et al. (2010). These physiological characteristics were measured under adjusted light and temperature conditions, the irradiance was 650 μmol m^{-2} s^{-1} of photosynthetically active radiation, the temperature in the measurement chamber was 25 °C, the CO_2 concentration was 420 ± 35 vpm (μmol mol^{-1}), the air flow rate was 205 ± 30 μmol s^{-1} and the duration of the measurement of each sample was 20 min after the establishment of steady-state conditions inside the measurement chamber. The value of VSD (vapour pressure deficit) was 0.85 ± 0.15 kPa. Measured parameters of gas exchange rates were calculated using the following formulas:

Rate of photosynthesis (P_N) [μmol CO_2 m^{-2} s^{-1}]

$$P_N = u_s \,.\, \Delta c, \qquad (1)$$

where Δc is the difference in concentration of CO_2 on input and output of the chamber [μmol mol^{-1}] and u_s is concentration – air flow per the m^2 leaf area [mol m^{-2} s^{-1}].

Rate of transpiration (E) [mmol H_2O m^{-2} s^{-1}]

$$E = u_s \,.\, \Delta W, \qquad (2)$$

where ΔW is the difference in concentration water vapor [mol mol^{-1}] and u_s is concentration – air flow per the m^2 leaf area [mol m^{-2} s^{-1}].

Stomatal conductance (g_s) [mol m^{-2} s^{-1}]

$$g_s = 1 : r_s, \qquad (3)$$

where r_s is stomatal resistance.

The minimum Chl fluorescence (F0) and the maximum Chl fluorescence (Fm) were also measured *in situ* with the portable 1 Chl fluorometer *ADC:OSI 1 FL* (*ADC BioScientific Ltd.*, Hoddesdon, Great Britain) with 1 s excitation pulse (660 nm) and saturation intensity 3,000 μmol m^{-2} s^{-1} after 20 min dark adaptation of the leaves. The maximum quantum efficiency of Photosystem (PS) II was calculated as Fv/Fm (Fv = Fm – F0).

Gas exchange and chlorophyll fluorescence were always measured from 9:00 to 12:30 h. CET. The natural intensity of PAR of the lower storey and the upper storey was 320 ± 55 μmol m^{-2} s^{-1} and 650 ± 50 μmol m^{-2} s^{-1}, respectively.

The energy content of phytomass (J g^{-1} of dry matter) was determined using an adiabatic calorimeter IKA C-4000 (software C-402). The samples weighing 0.7–1 g and homogenised were pressed into a form of briquette, dried up to a constant weight at 105 °C and burnt in pure oxygen under a pressure of 3.04 MPa (DIN 51900). The ash content was determined gravimetrically, by total oxidation of specimens in a muffle furnace at 500 °C (Javorský et al., 1987).

Statistical analyses were made with the use of a statistics program Statistica 9 software and the variability in mean physiological and energy characteristics of plant species between monitoring plots was tested by anova model. The significance of differences was verified by Fisher-LSD test. The measured characteristics included rate of photosynthesis, rate of transpiration, stomatal conductance, chlorophyll fluorescence, energy and ash accumulation. Average ± standard deviation for the physiological characteristics was within 15–30 individuals while preserving three repetitions. For each plant species, three measurements of energy value were made.

Results

Rate of photosynthesis (P_N)

Rate of photosynthesis of plant species growing in clear-cut area fluctuated in the interval from 4.51 μmol CO_2 m^{-2} s^{-1} (*D. filix-mas*) to 13.29 μmol CO_2 m^{-2} s^{-1} (*F. sylvatica*). In the case of fir-beech stand, photosynthesis was the lowest in ferns (5.74 μmol CO_2 m^{-2} s^{-1}) and the highest in beech leaves (16.31 μmol CO_2 m^{-2} s^{-1}). On studied plots, herb and shrub species showed lower photosynthesis compared to the assimilatory organs of *F. sylvatica*, Fig. 1. Significantly lower photosynthesis was observed in assimilatory organs of beech, male fern, bilberry and raspberry in clear-cut area in comparison with the fir-beech stand (significance level $\alpha < 0.001$). In the case of *F. sylvatica* species, the difference between compared plots was the highest (3.21 μmol CO_2 m^{-2} s^{-1}), Fig. 1.

Rate of transpiration (E)

On studied plots, herb and shrub species showed lower rate of transpiration (0.49–1.65 mmol H_2O m^{-2} s^{-1}) compared to the assimilatory organs of *F. sylvatica* (2.10–2.11 mmol H_2O m^{-2} s^{-1}), Fig. 2. In the case of *F. sylvatica*, *D. filix-mas* and *V. myrtillus* species higher rate of transpiration was found in clear-cut area; *R. idaeus* species showed higher transpiration in the fir-beech stand. Significantly different were only the mean values found for *D. filix-mas* species (significance level $\alpha < 0.001$). For other plant species differences between compared plots were not statistically significant.

Stomatal conductance (g_s)

The values of stomatal conductance on studied plots varied from 0.19 ± 0.05 mol m^{-2} s^{-1} (*D. filix-max*) to 0.57 ± 0.23 mol m^{-2} s^{-1} (*R. idaeus*), Fig. 3. In the case of *F. sylvatica* and *V. myrtillus* species higher values of

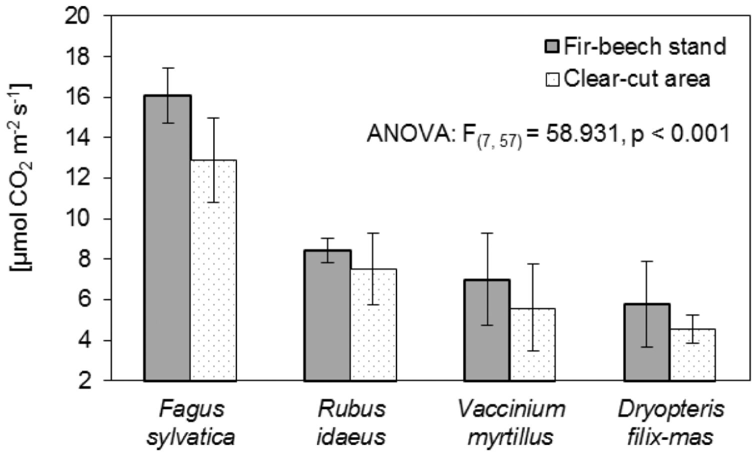

Fig. 1. Rate of photosynthesis (P_N) of assimilatory organs of dominant plant species (mean value ± SD).

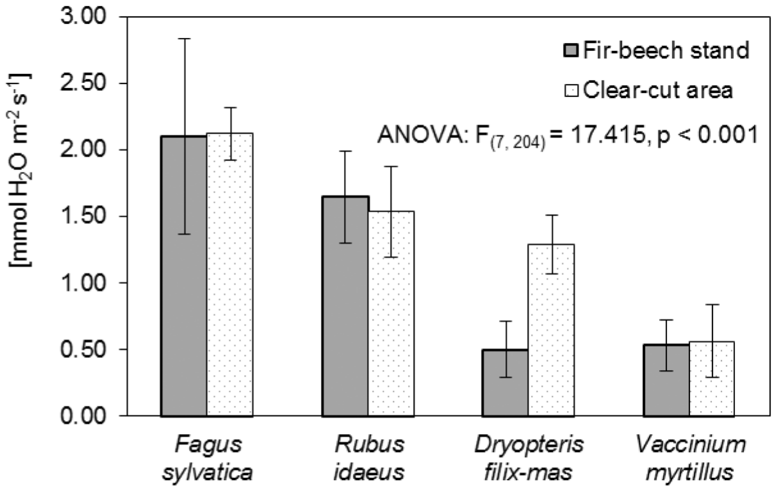

Fig. 2. Rate of transpiration (E) of assimilatory organs of dominant plant species (mean value ± SD).

stomatal conductance were observed in the fir-beech stand; *R. idaeus* and *D. filix-mas* species showed higher values in clear-cut area. Significantly different were only the mean values found out for *F. sylvatica* and *V. myrtillus* species, where the differences between plots were the highest (significance level $\alpha < 0.001$). The rate of gas exchange can be limited not only by stomatal conductance (gs), but also by substomatal CO_2 (ci). The values of substomatal CO_2 varied from 490 ± 35 vpm to 610 ± 35 vpm.

Chlorophyll fluorescence

Studied plant species showed lower values of fluorescence in clear-cut area (range 0.57–0.80) in comparison with the fir-beech stand (range 0.80–0.81). The lowest value of fluorescence was found out for *F. sylvatica* species (0.57 ± 0.02) growing in clear-cut area, the highest for *R. idaeus* species in the fir-beech stand (0.81 ± 0.18). The highest difference between studied plots showed assimilatory organs of *F. sylvatica* (29%), the lowest *R. idaeus* species (1.3%), Fig. 4.

Energy and ash accumulation

Higher values of energy were observed in assimilation organs of beech, male fern and blueberries (range 19,984–20,551 J g^{-1}) in the fir-beech stand; raspberry showed slightly higher value of energy in clear-cut area (18,562 ± 325 J g^{-1}) in comparison with the fir-beech stand (18,511 ± 251 J g^{-1}), Fig. 5. Significantly lower was only the mean calorific value found for *D. filix-mas* species (18,562 ± 310 J g^{-1}) growing in clear-cut area compared with the value in fir-beech stand (19,049 ± 251 J g^{-1}), (significance level $\alpha < 0.05$). Ash content of analysed plants in the fir-beech stand varied in the following order [mg g^{-1}]: 31.6 (*F. sylvatica*) < 41.0 (*V. myrtillus*) < 46.1 (*R. idaeus*)

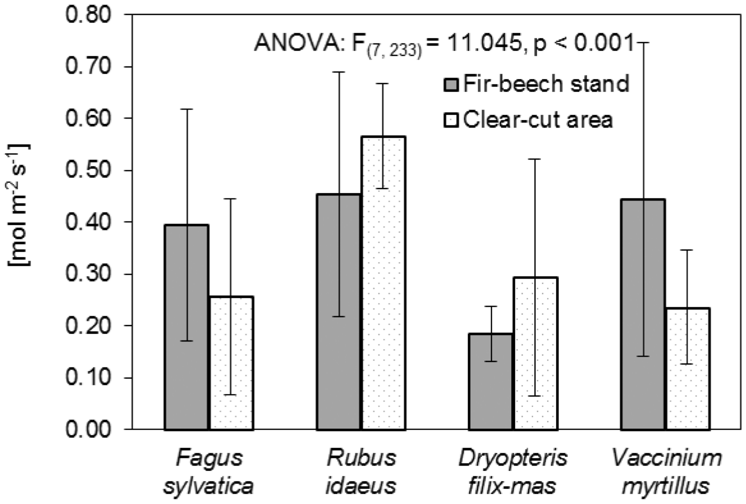

Fig. 3. Stomatal conductance (g_s) of assimilatory organs of dominant plant species (mean value ± SD).

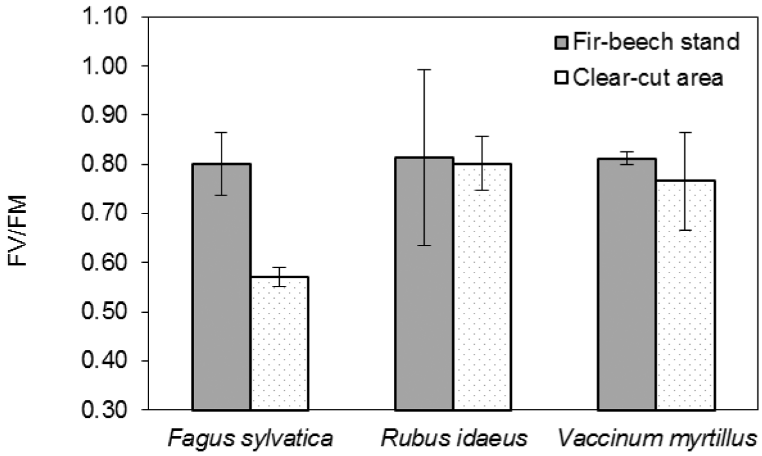

Fig. 4. Potential photochemical efficiency of electron transport in photosystem II of dominant plant species (mean value ± SD).

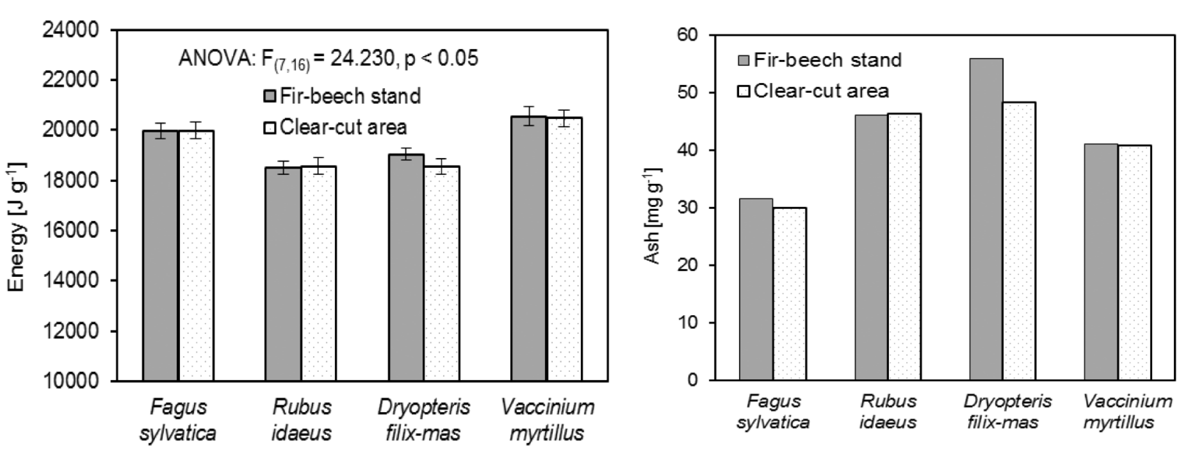

Fig. 5. Energy and ash contents of assimilatory organs of dominant plant species (mean value ± SD).

< 55.9 (*D. filix-mas*); in clear-cut area the values were as follows [mg g^{-1}]: 30.0 < (*F. sylvatica*) < 40.8 (*V. myrtillus*) < 46.4 (*R. idaeus*) < 48.4 (*D. filix mas*). Slightly higher was ash content in the dry matter of assimilatory organs of male fern in the fir-beech stand, only.

Discussion

Plants are in the course of their lives exposed to changing conditions of environment. Adverse environmental conditions (excessive radiation, high temperature, lack of water, lack of nutrients in the soil, etc.) can slow down their vitals, but also damage the organs of plants. Physiological reaction and quality of dry matter of assimilatory organs can also be influenced by acidic precipitations with increasing industrial and agricultural activities of man. The airborne pollutants deposited on the leaf surface can affect the plant´s metabolism by blocking light, obstructing stomatal apertures, increasing leaf temperature and altering mineral contents of the leaf (Hope et al., 1991; Pavlík et al., 2012). For all studied plant species in buffer zone of Slovenský raj NP, decrease of photosynthesis was recorded in clear-cut area (10-year-old stand), where obviously more radiation and thus heat penetrate to stand compared with the 80-100-year-old stand. With inappropriate environmental conditions the rate of photosynthesis decreases (Matyssek et al., 1993). Drought stress or sudden radiation, are considered to be the most important factors, which plants encounter in nature. In the absence of water in the plant, it loses its ability to cool off, in particular sunlit part of its phytomass and this leads to its overheating (Penka, 1985).

Direct irradiation of plant affects not only the plant itself, but also changes the microclimate around it, which in turn may affect its growth (Anderson, 1964). Light intensity in forest stand is lower than in the open area and its quality varies depending on the absorption and reflection of leaves. In submontane beech stands of different stocking in Kremnické vrchy Mts, e.g. Janík (2009) observed at the stage of full foliage values from 2.3 to 8.2% of open area lighting.

In the case of *F. sylvatica* species, the difference in rate of photosynthesis between compared ecosystems was the highest. Deteriorated physiological status of juvenile beech trees on the site with higher immission load is stated by Kmeť and Ditmarová (2001). According to the results of Woo (2009), beech belongs to the sensitive woody plant species on air pollution which is also reflected in the change of rate of photosynthesis. Takagi and Gyokusen (2004) observed higher rate of phytotosynthesis of leaves of the *Ilex* trees in the urban core, whereas in the suburban area it was the lowest. The authors found, that rate of photosynthesis was negatively correlated with sun light conditions and positively with air pollutant concentrations.

According to Farmer (1993), airborne dust in forest ecosystems also influences transpiration rate. *R. idaeus* species showed higher transpiration in the older fir-beech stand, only. All other plant species had higher values on plot with lower stand density (clear-cut area). The most sensitively to file of habitat conditions reacted *D. filix-mas* species, where the difference between compared plots was the highest, which could also be caused by higher input of risk elements from polluted atmosphere (leaching of risk elements captured in fir-beech stand canopy).

Stomatal conductance is one of the important factors affecting the metabolism of plant (Agbaire, 2009). Interspecies differences in stomatal conductance are confirmed by Tomasevic et al. (2005), who observed the influence of pollutants on the characteristics of hazel and horse chestnut. Changes in the values of substomatal CO_2 (ci) depending on environmental factors are stated by Kumaravelu and Ramanujam (1998), Liang, et al. (2008). Differences in stomatal conductance depending on the locality were studied by Nizzetto and Perlinger (2012). According to Dohmen et al. (1990) and Leuzinger and Körner (2007), stomatal conductance usually due to air pollution and the increasing concentration of CO_2 decreases, but the reaction in plants is not always reflected. From measured values of stomatal conductance of plant species in Slovenský raj NP is evident not only effect of site, but also the differences between plant genotype. In assimilatory organs of beech and blueberry, stomatal conductance was higher in the 80-100-year-old ecosystem; in the mature stand, leaves of raspberry and male fern answered more sensitively, which can again be related to airborne pollutants (leaching of risk elements captured in fir-beech canopy). Prasad (1995) for example states, that the plants from the control conditions have higher stomatal conductance in comparison with the plants from the stressed conditions.

Chlorophyll fluorescence is also an important physiological indicator, which appears to be a suitable indicator of plant reaction to stress (Maxvell and Johnson, 2000). Negative impact of stressors on chlorophyll fluorescence was studied by Gamon and Surfus (1999) and Takayama and Nishina (2009). These authors state that not always, these changes are accompanied by visible signs of damage leaves. Studied plant species in Slovenský raj NP showed lower values of fluorescence in clear-cut area in comparison with the fir-beech stand. The most sensitively to file of habitat conditions answered *F. sylvatica*, the least *R. idaeus* species. In the case of C3 plants, photosynthesis cycle as a "normal" is the range from 0.79 to 0.84 (Maxwell and Johnson, 2000). Measured values of potential photochemical efficiences of electron transport in photosystem II (Fv/Fm) showed decrease in this parameter under physiological limit for *F. sylvatica* species growing in clear-cut area.

On studied plots significantly higher was only the mean value of energy found for *D. filix-mas* in the fir-beech stand compared with clear-cut area. This fact was indeed associated with ecological demands of species (semi-shadow, water moderately demanding species). Calorific values for all other species were similar (slightly higher in clear-cut area, where more light was available for production of dry matter) and did not significantly differ between plots. Bobkova and Tuzhulkina (2001) state that combustion heat as a physical parameter is characterized by a relatively high variability, being dependent on plant species, growing conditions, morphological structure, age, period of sampling, and other factors. Bublinec et al. (2011), Schieber and Kováčová (2002) e.g. in mature beech forest in Kremnické vrchy Mts found out lower energy contents (by 5–22%) in herb species compared to plants growing on anthropogenic influenced plot (clear-cut area). Negative effects of pollutants on herb understorey in Bielovodská dolina valley, the High Tatras Mts, were observed by Kukla et al. (2003). In case of blueberry, the authors found higher energy values in undamaged stands opposite those anthropogenic affected areas. Ash contents of plant organs in studied stands are typical for the plants growing on acidic soils, poor in nutrients. On studied plots in Slovenský raj NP, slightly higher value of ash was showed by the dry matter of assimilatory organs of male fern in the fir-beech stand, only.

Conclusions

Evaluation of growth processes occurring in phytocoenoses is an important source of information about the degree of threat to forest ecosystems, depending on the intensity of the negative external environmental impacts. The research conducted in fir-beech ecosystems affected by air pollution, situated in the locality Hliníky (the buffer zone in Slovenský raj NP) showed, that forest stands with its structure, density of canopy of trees and habitat conditions differentiated the measured physiological characteristics. From measured values of gas-exchange parameters is evident not only effect of site, but also the differences between plant genotypes. Environmental conditions (radiation, drought, airborne pollutants) mainly influenced rate of phytotosynthesis and rate of transpiration by relatively lower values of plants in the juvenile stand (clear-cut area). In the case of *F. sylvatica* the difference of photosynthesis between compared ecosystems was the highest. The most sensitively to file of habitat conditions reacted *D. filix-mas* species with significantly lower value of transpiration in the engaged stand, which could be caused by impact of airborne pollutants (leaching of risk elements captured in fir-beech canopy). The results showed significant effect of site, but also plant genotype on stomatal conductance of plants (*V. myrtillus* and *F. sylvatica* answered sensitively with significantly lower values in clear-cut area, *R. idaeus* and *D. filix-mas* in the mature stand). Reduction of the potential photochemical efficiency (FV/FM) under physiological limit was found for *F. sylvatica* species in clear-cut area, where the file of habitat conditions for growth of juvenile beech trees was not probably favourable. Approximately 7–10% higher energy contents on studied plots were showed by assimilatory organs of beech and blueberry compared to raspberry and fern. Significantly higher was only the mean calorific value found for *D. filix-mas* species in the mature stand, where habitat conditions for growth of this species were obviously optimal.

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