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CHANGES IN A PROTECTED, OLD-GROWTH *Abies alba - Fagus sylvatica* FOREST IN THE ROMANIAN CARPATHIANS FOR 37 YEARS

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Abstract

The vegetation of an old mixed beech and fir forest from the foothills of the Bucegi Mountains, in the South Carpathians, Romania, was first surveyed in 1972-1973 and secondly in 2008-2009 in 121 sample plots from a representative area displaying a heterogeneous structure, the oldest trees being over 250 years. The research sought to assess the changes in the structure of tree populations and herbaceous layers after 37 years, and the relationships between the characteristic and dominant species. The results show that the Piatra Arsă forest has a diversified structure and is rich in terms of species and habitat diversity. Certain structural changes in the forest may be related to climate change, resulting in an increased proportion of beech at the expense of fir. Except for the shift in tree species dominance after 37 years under conservation, the Piatra Arsă forest displayed rather small structural changes (shifts of the micro-habitats), which may be attributable to natural fluctuations characterizing a natural old-growth forest.

Key words*: mixed fir-beech stand, species composition, stand structure, uneven old-growth forest.*

INTRODUCTION

Vegetation changes over time because of natural ecological succession are a well-established concept developed at the beginning of the 20th century (Clements, 1916). Forests are dynamic open systems, showing continuous changes in time of their structure and functions (Doniță et al., 1977; Chiriță, 1981).

Component organisms are ecologically interdependent through their complex interspecific relationships (Zaitsev et al., 2014; Manu et al., 2017; 2019; 2020; 2021) making the natural forest, as a whole, relatively stable (Giurgiu, 1978). In forest ecosystems, significant successional changes might take hundreds of years, but the vegetation heterogeneity may strongly affect the trajectory and speed of successions due to synergic effects of human and natural disturbances. Over the past century, observed changes in forest composition (Paluch, 2007; Cicșa et al., 2022), and area (Hédl, 2004; Sobala et al., 2017) appear mainly caused by anthropogenic impacts.

The "old-growth forests" are defined as climax successional stage (late successional development) (Nagel et al., 2014; Lábusová et al., 2019; Gray et al., 2023); the major climax forest communities, such as montane spruce- and beech-dominated forests, are robust in terms of stability and integrity and, at large spatial scales, their structure is little affected by disturbance that strongly impact other ecosystems over small scales (Chiriță, 1981; Cicșa et al., 2021).

In recent decades, many socioeconomic and institutional changes that affect forest management practices have occurred around the world, resulting in the shrinkage of the area of old-growth forests at an alarming rate (Gilg, 2004; Keeton et al., 2013; Sobala et al., 2017; Parobeková et al., 2018; Nikolakis & Innes, 2020). In the past, Romanian old-growth forests were part of a more extensive European formation (Doniță et al., 1992), but between 2000 and 2010, their cover declined by 1.3% (Knorn et al., 2012).

In a mixed stand of fir (*Abies alba* Mill.) and beech (*Fagus sylvatica* L.) the responses of tree growth to the most important environmental factors (precipitation and temperature) vary between individual trees and stands due to competition, health of the trees, size of their photosynthetic leaf area and their genetic constitution (Negulescu et al., 1973; Doniță et al., 1977). Environmental factors correlate more closely with the relative growth rate of large-diameter beech trees (i.e. dominant trees) than that of smaller subdominant trees (Knott, 2004). Temperature appears to have a more significant influence on large-diameter fir trees than does precipitation when compared with smaller diameter *Abies* trees (Pach & Podlaski, 2015).

The dynamics of understorey vegetation (herbaceous layer) in the forest reflect differences in habitats from particular phytogeographical regions (Ujházy et al., 2005) and are significantly influenced by the dynamics of the dominant tree species (Šamonil & Vrška, 2007) and by changes in space and time of the abundance of different component species (Ulrich, 2008; Morlon et al., 2009; Barbara et al., 2012; Su, 2018).

The biomass (above- and below-ground parts) of herbaceous species is an important indicator of mineral cycling in forest ecosystems (Brumme & Khanna, 2009; Heiri et al., 2009; Pesklevits et al., 2011; Durak, 2012), even though it is quantitatively lower than the biomass of tree and shrub layers (Schulze et al., 2009).

In the Carpathians, at lower altitudes, increasing drought (changes in temperature and precipitation) coupled with increased pests/ pathogens and danger of fire, lead to shifts in species composition in forests that might in turn result in species and community collapse, especially where connectivity and ecological corridors are limited (Werners et al., 2014).

Our research was designed to document the changes in the structure of an uneven-aged, mixed forest stand. In particular, the present study sought to reveal the changes in the structure of tree populations and, especially, the herbaceous layer after 37 years of total protection.

MATERIALS AND METHODS

The Piatra Arsă forest was formerly private property of King Mihai I (part of his residence in Sinaia), but state-owned in our days, under the National Forestry Administration and included in the Bucegi Natural Park as a zone of special conservation.

The total area of this forest is 5250 hectares and is located at the foot of the Piatra Arsă mountain in the Bucegi Mountains (Massif), in the upper basin of the Prahova River (western slope), near Sinaia town (N: 45^0 22' 28.9” and E: 025^0 32' 16.6") (Figure 1).

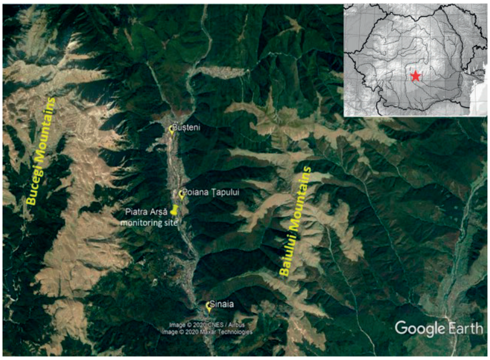

Figure 1. The location of the research area

Lying between 800-1100 m altitudes, the study area is widest to the north (where it is more sheltered and comprises a plateau, 10 km wide) and narrow to the south (where the land is lower, more open and with sunny slopes, 4 km width). The soil is a mollic eutricambosol (brown forest soil) typical of mountainous areas covered with beech and coniferous forests; it is deep, saturated, eubasic, with medium texture and high nutrient content. The climate is moderate continental; the annual mean temperature in the period 1967-1970 was 5.6^{0}C, but 1.9^{0}C higher in 2008. The rainfall ranged between 848 mm and 1025 mm in the interval 1967-1970, and over 50% of precipitation fell in summer. In 2008 it was 950 mm, 100 mm more than in 1972.

Within this uneven-aged mixed beech (*Fagus sylvatica*) and fir (*Abies alba*) forest, it has been selected 1 ha permanent monitoring plot with the oldest trees exceeding 250 years, we performed our measurements in the sampling periods 1972-1973 and 2008-2009.

Syntaxonomically, this stand is classified within the *Pulmonario rubrae-Fagetum* (Soó 1964) Täuber 1987 community type. *Fagus sylvatica* and *Abies alba* are the characteristic co-dominant species of the phytocoenosis, together with *Pulmonaria rubra* in the ground layer and other species from the *Symphyto-Fagion* Alliance, *Fagetalia* Order and *Querco-Fagetea* class (Sanda et al., 2008), The Romanian habitat in which we can frame this plant association is R4101 South-East Carpathian forests with spruce (*Picea abies*), beech (*Fagus sylvatica*) and fir (*Abies alba*) with *Pulmonaria rubra*. This habitat is in turn part of the NATURA 2000 type 91V0 Dacian beech forest (*Symphyto-Fagion*) (Doniță et al., 2005; Gafta & Mountford, 2008).

The main factors affecting the study site are: the occasional illegal logging by local people, small ravines formed on steeper slopes, and the proximity to the national road (DN1) which is usually overloaded with high traffic leading to pollutant depositions in the nearby forest.

Circular areas of 500 m^2, in five locations, were used to survey the tree layer (living trees), stumps (dead trees) and undergrowth (1-10 years old living young trees). The diameters of living trees were measured at the breast height (DBH) with a graduated tape. For stumps, we measured two diameters at right angles to one another, and for undergrowth we employed the inventory procedure used by Popescu-Zeletin et al. (1973; 1979) i.e. recording: all individuals from the tree layer with DBH larger than 4 cm; and all young trees aged 1-10 years individuals from the undergrowth layer (saplings) with DBH less than 4 cm but with heights more than 1 m.

On the entire area of the permanent plot, for surveying the herbaceous layer, we systematically (regular grid) installed 121 semi-permanent 1 m^2 sub-plots where we inventoried all individuals: herbaceous and 0-1 year young trees (seedlings). The sub-plots were marked with wooden sticks, positioned at equal distance (10 m) to be able to use the same micro-sites again for long term monitoring. On these permanent marker sticks, we used a metal circle (1 m^2) to delimit a precise area for the inventory of individuals of each species (herbaceous and young trees).

The distinctions of individual herbaceous plants were made visually, where an entire plant or a detached above-ground shoot could be clearly defined; these were recorded as individuals for subsequent counting. In a very few species, such as *Oxalis acetosella*, whose leaves emerge directly from the soil without a recognisable stem, each leaf was treated as an individual.

For each vascular plant species, the following data were taken from the literature (Sârbu et al., 2013): a) the ecological indicator values (for light, air temperature, soil nitrogen, moisture, and reaction) i.e. original Ellenberg's values adapted to the pedo-climatic conditions of Romania; and b) the phytocoenological category.

All statistical analyses were performed using the software package PAST (Hammer et al., 2001). The variables involved in various analyses included the number of plant species, the abundance (count of individuals) of each plant species, species diversity (Shannon-Wiener index), dominance (D index) and equitability (J index). All indices were tested for homogeneity of variance and subsequently subjected to t-test and ANOVA.

The environmental and species data were subjected to correspondence analysis (CA) and canonical correspondence analysis (CCA) (Legendre & Legendre, 1998). For the first five parameters, the mean values were graphically represented, including the standard error (± STE).

The correspondence analysis was based on the number of recorded individuals; both the Jaccard similarity index and Spearman rank correlation coefficient were used to estimate the floristic resemblance and species association.

RESULTS AND DISCUSSIONS

Tree layer

In Piatra Arsă forest, the number of mature trees is low, with individual trees of fir having diameters up to 113.38 cm and beech up to 105.41 cm. The total density of individuals in the tree layer decreased substantially over 37 years and the balance in abundance between the two dominant species changed through reduction of *Abies alba* and increase of *Fagus sylvatica* (Figure 2).

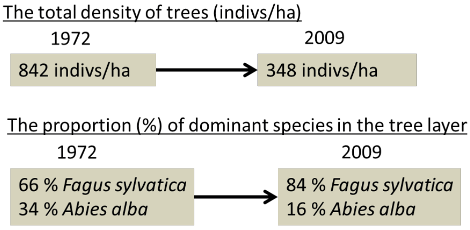

Figure 2. The change in density and proportion of tree species individuals over 37 years

The undergrowth layer was well-developed and mirrored the pattern of tree layer, except that *Abies alba* was better represented. The total density of inventoried stumps indicates low mortality (natural or man-induced) with higher incidence on fir.

The within-species diameters vary greatly with the age of individuals. The inter-specific variation is higher, the average diameters of fir individuals being larger than those of beech individuals in both layers. The distribution of trees by diameter class showed almost the same range in 1972 and in 2009. Trees belonging to three diameter classes (40-50 cm, 90 cm and bigger than 100 cm) were more frequent in 2008 (Figure 3). Tree individuals with low DBH remained very frequent, reflecting continuous and active regeneration.

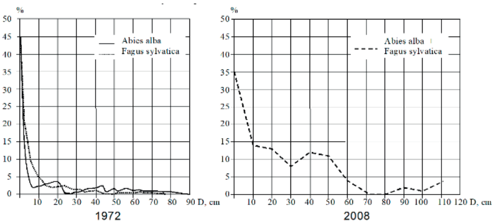

Figure 3. Distribution of the relative frequencies of beech and fir trees by diameter class

The number of mature large trees is low, but the variety of the diameter classes is high, showing a high vertical diversity and heterogeneity that is characteristic of an uneven-aged forest. The density of individuals in the tree layer was typical for old-growth forests (Popescu-Zeletin et al., 1973; 1979).

Structural changes in the forest depend greatly on the range and distribution of diameters in the dominant fir and beech trees. Pach & Podlaski (2015) reported that the more complex distribution of DBH occurs in unmanaged forests, characterised by a higher proportion of living trees in large DBH classes. Unmanaged forests, with their more complex structure, are generally richer in species and display higher microhabitat heterogeneity.

Hilmers et al. (2019) studied the productivity of mixed beech-spruce-fir forests from different mountain areas over 30 years and concluded that, despite a significant increase in annual mean temperature and stable precipitation, the average productivity of the European mixed montane forests has not changed significantly over recent decades. The volume and productivity growth were reported to be stable, and the volume increment of fir and beech was negatively correlated. Increased diversity of tree species composition might increase the adaptability of the forest to climatic and anthropogenic changes. Numerous studies have shown that the response of the component species to climate change is different, resulting in an increased proportion of beech at the expense of fir (Šamonil & Vrška, 2007; Diaci et al., 2007; Vrška et al., 2009; Klopcic & Boncina, 2011; Horvat et al., 2018). Some of the trends at Piatra Arsă may arise in part from climate-change but this cannot be confirmed from the data gathered and results.

In the Piatra Arsă forests, the tree layer was affected by a shift in species dominance over a period of 37 years: beech (34% in sampling period 1972-1973 increased to 84% in sampling period 2008-2009) overtaking the fir (66% in sampling period 1972-1973 decreasing to 16% in sampling period 2008-2009) and the overall density diminished. Only fir stumps were found, reflecting greater mortality in that species, which may be a consequence of global warming. Although the number of *Abies alba* trees was small, they were the largest individuals. Where the density of tree saplings is sufficiently high, this may affect the light reaching the forest floor and hence the dynamics of the herbaceous layer. The proportion of seedlings of different woody species was highly variable in time, resulting in a very dynamic recruitment of trees. The recruitment dynamics of the trees is very changeable in time due to variation in the number of seedlings and the proportion of the seedlings of different woody species.

The inventory of the undergrowth layer represented by juveniles (saplings and seedlings) of fir (*Abies alba*), beech (*Fagus sylvatica*) and sycamore maple (*Acer pseudoplatanus*) showed a greater abundance of fir saplings than those of the other two species in 1972. In contrast, in the last inventories (2008-2009), the beech saplings were the most abundant, followed by. the sycamore maple saplings and finally, by the fir saplings (Figure 4).

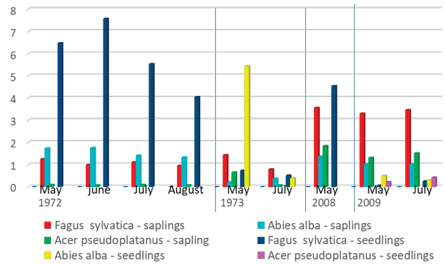

Figure 4. Density of tree species juveniles (by 1 square metre) in the undergrowth layer.

The seedlings of beech occurred in high densities in 1972 but decreased in time (Figure 5). Fir seedlings were absent in the first year (1972) but quite frequent in the second year (1973). Similar variation was encountered in the second monitoring period (2008-2009). Seedlings of sycamore maple were absent during the first monitoring period and in 2008, but present in 2009.

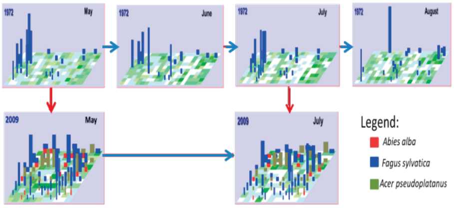

Figure 5. Abundance distribution of beech, fir and sycamore maple seedlings at 1 m^2 grain scale throughout the study plot in different months and years

The abundance distribution of fir, beech, and sycamore maple seedlings in the undergrowth layer (Figure 5) also showed great variation in time. In the first year of assessment (May→August 1972), the short-term (annual) intra-specific variation was high, and only beech seedlings were observed in the investigation plots.

The variation of abundance distribution of dominant trees seedlings is high both among seasons of the same year and especially between time periods (May 1972→May 2009 and July 1972→July 2009). Long-time investigation showed an increase in taxonomic diversity of seedlings.

Herbaceous layer

The frequency and diversity of species in the herbaceous layer varied greatly among the 121 semi-permanent plots established in the 1 ha forest plot. The most frequently recorded were some character species of the *Symphyto-Fagion* alliance i.e. *Fagus sylvatica, Abies alba, Cardamine glanduligera,* and *Pulmonaria rubra.* Species typical of the *Fagetalia* order with high frequency were: *Lamium galeobdolon, Galium odoratum, Mercurialis perennis, Isopyrum thalictroides* and *Stellaria nemorum.* Species representative of the *Querco-Fagetea* Class with high frequency and high abundance were: *Impatiens noli-tangere, Galium schultesii* and *Viola reichenbachiana.* Some herbaceous species were present in the sampling period 1972–1973 but were not found again after 37 years: *Dryopteris dilatata, Veronica urticifolia, Chaerophyllum aureum, Moehringia trinervia, Lapsana communis, Stellaria holostea* and *Cardamine amara.* Other herbaceous species were recorded in the sampling period 2008–2009 but had not been recorded in the baseline period 1972–1973: *Hepatica transsilvanica, Euphorbia carniolica, Anemone ranunculoides, Carex sylvatica, Senecio ovatus, Allium ursinum, Corydalis cava, Geranium phaeum, Maianthemum bifolium, Glechoma hirsuta, Galium schultesii, Scilla bifolia, Epipactis helleborine, Ranunculus ficaria, Carex remota, Leucanthemum vulgare* and *Solanum dulcamara.* As a proportion of the overall ground layer flora, relatively few species appeared or disappeared over the 37 years of the study. The number of species varies significantly from one season to another and from one year to another, pointing to active dynamics within the herbaceous layer (Figure 6). Overall, there were significant differences between the average numbers of individuals of herbaceous species.

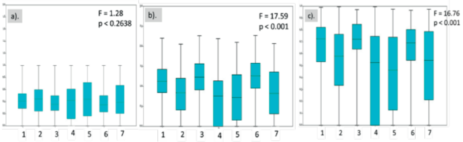

Figure 6. Box plots representing the number of species recorded at 1 m^2 scale in the seven months of assessment (1-May 1972; 2-July 1972; 3-May 1973; 4-July 1973; 5-July 2008; 6-May 2009; 7-July 2009)

Species dominance did not significantly differ in time (Figure 6a). However, both species richness (at 1 m^2 scale) and equitability did differ significantly between timings of observation. The richness and evenness also varied from one month to another (spring-May and summer-July) in the same year. The indices showed similar patterns in the same season from different years, as no major differences were detected between the corresponding distributions (Figure 7).

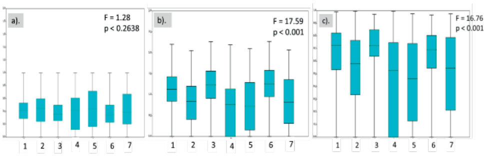

Figure 7. Box plots representing the distributions of the dominance index (a), Shannon-Wiener index (b) and equitability index (c) calculated at 1 m^2 scale in the seven months of observations (1-May 1972; 2-July 1972; 3-May 1973; 4-July 1973; 5-July 2008; 6-May 2009; 7-July 2009)

The ordination diagram (Figure 8) may suggest different groups of species related to the season of inventory within the same sampling period (i.e. July, May). Axis 1 may be related to flowering and growth seasons, with vernal species to the right and aestival species to the left. Thus, vernal species were well-developed in the spring inventory but some withered individuals persisted and were still recorded in July. For example, *Anemone ranunculoides* (Ane_ran) has the following frequencies: 3.08% in July 2008, 23.14% in May 2009 and 1.65% in July 2009. On the other hand, *Ranunculus ficaria* (Ran_fic) was found only in May 2009 and had 11.57% frequency. Axis 2 may depend on soil fertility i.e. species of infertile soils toward the bottom of the ordination diagram and those of more nutrient-rich soils toward the top of the ordination.

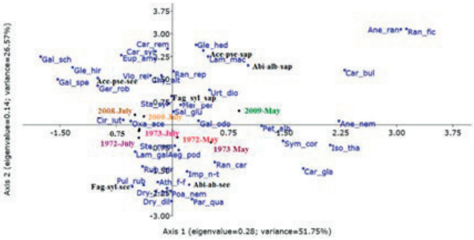

Figure 8. Correspondence analysis of plant species from the seven investigated months

Abies alba seedlings had a higher abundance in spring but diminished in summer. The same trend was observed in *Fagus sylvatica* seedlings, but *Acer pseudoplatanus* seedlings displayed a different pattern, with smaller density in spring and increased density in summer. The within-year variation in seedling abundance was similar between tree species. However, the abundance of fir and beech seedlings remained stable, whereas the abundance of sycamore maple seedlings increased in 37 years.

The shade-tolerant beech seedlings were associated with shade-tolerant herbaceous species like *Galium odoratum* (Gal_odo), *Mercurialis perennis* (Mer_per), *Impatiens noli-tangere* (Imp_n-t) and *Stellaria nemorum* (Ste_nem) (Figure 9).

Fir seedlings were associated with: *Rubus hirtus* (Rub_hir), *Pulmonaria rubra* (Pul_rub), *Athyrium filix-femina* (Ath_f-f) and *Poa nemoralis* (Poa_nem), which are semi-shade (moderate shade) tolerant species (Figure 9).

Sycamore maple seedlings (Ace_pse_see) were strongly associated with *Euphorbia amygdaloides* (Eup_amy), *Galium schultesii* (Gal_sch) and *Glechoma hederacea* (Gle_hed), once again semi-shade to shade-intolerant species (Figure 9).

The saplings of fir, beech and sycamore maple were associated with a mixture of species that have different tolerance to shade: *Ranunculus repens* (Ran_rep), *Carex remota* (Car_rem), *Carex sylvatica* (Car_syl), *Galeopsis speciosa* (Gal_spe), *Geranium robertianum* (Ger_rob).

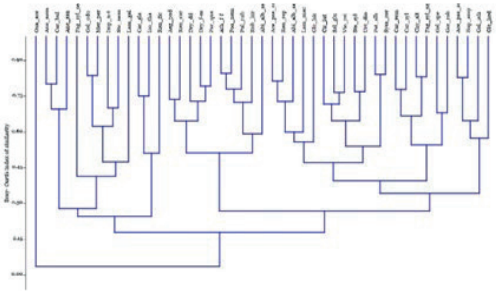

Figure 9. Bray-Curtis index of similarity among numerical abundances of investigated plant species from all 7 investigated months, in Piatra Arsă

In mixed fir and beech forests, the productivity of above-ground biomass of the herbaceous layer undergoes annual variation over the growing season, but also multiannual variation due to stochastic factors like the weather conditions specific to every year (Brezeanu et al., 1972; 1975) and heterogeneity in the microhabitats (Ciocârlan, 2009). Some herbaceous species showed a high correlation between density and frequency, especially during spring when species such as *Oxalis acetosella* and *Galium odoratum* have great abundance.

The frequency of the recorded species (clustered and random) will be determined by site conditions. We may suggest two probable categories although the environmental data that would support this inference are not available, and it is at least as likely that the observed patterns in species frequency arise from variation in seed dispersal and vegetative reproduction. For example, *Oxalis acetosella* and *Cardamine glanduligera* are spread over the entire investigated area whilst species such as *Mycelis muralis* and *Myosotis sylvatica* have their greatest frequency in limited areas, presumably due to the specific conditions present there in terms of soil moisture, pH, fertility, or light (Paucă-Comănescu et al., 1977; 1978; 1979).

Both the species that were newly recorded in 2008-2009 and those that apparently disappeared after being found in 1972-1973 are generally typical of the *Querco-Fagetea* class. However, the number of character species of the *Symphyto-Fagion* alliance declined to some extent. One explanation for this slight change in the composition of the ground herbaceous flora might be that the tree canopy cover was lower in 2008-2009 than in 1972 and, therefore, there was lighter available at soil level. The qualitative change of species composition was generated not only by "companion" species, but also by character species of *Symphyto-Fagion* and *Fagetalia*. The maximum values of the relative abundance of some species (*Oxalis acetosella* > 70%, *Lamium galeobdolon, Mercurialis perennis* and *Galium odoratum*) varied little over the sampling periods. Species richness and relative abundance differed between phenological seasons (spring-May and summer-July) of the same year.

Environmental changes

The environmental changes are indirectly inferred from the weighted means of the ecological indicator values (L, T, U, R, N) (Figure 10).

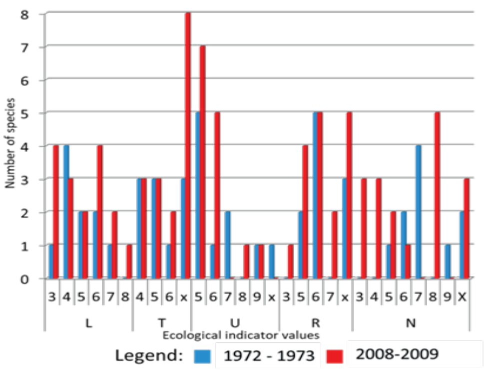

Figure 10. Changes in ecological indicator values of species (L - light availability, T-air temperature, U - soil moisture, R - soil reaction N - soil nitrogen) over the investigated periods

Most plant species recorded in the plot over the two inventorying periods, including the dominant trees, are especially associated with the mountainous (T=4) or sub-mountainous (T=5) regions of Romania. They grow on soil that can be well drained, moderately moist, or even damp (U=5-7) but not wet, prefer soils that range from moderate to low acidity to neutral (R=4-7), but some are indifferent (R=x).

In the period 1972-1973, the most abundant species where those growing in shadowed (L=3-4) and moderate shading (L=5-6) micro-habitats highlighting the high density of the tree layer (Figure 2). In the period 2008-2009, the light preferences of the recorded species are diverse, showing the high diversity of micro-habitats from the forest.

The preferences for temperature of the species are quite similar for both inventorying periods, in the recent years (2008-2009) increasing the frequency of the species that have no particular preferences for temperature (T=x).

In the period 1972-1973, the plat species diversity and frequency were quite general distributed among the values of soil moisture ecological indicator but still showing a slight dominance of the preferences for moderate humid soils (U=5-6), but there are also species preferring humid well-drained soils (U=8-9) and some are indifferent (U=x). In the period 2008-2009, changes occurred toward dominance of the plant species living in moderate humid micro-habitats and in small number in other type of micro-habitats.

The pattern of species preferences for soil reaction is almost the same for both investigated periods, excepting the increased frequency of the indifferent species (R=x). Some species as *Ranunculus carpaticus* (present in 1972 and 2009) and *Maianthemum bifolium* (present only in 2009) prefer acidic soils (R=3) and have low frequency in the plot, their distribution depending mainly on the occurrence of micro-sites with acidic soils.

Almost all the species recorded prefer soils rich in mineral nitrogen (N=5-7 even 8), but in the period 2008-2009 there were newcomers that typically prefer low nitrogen content in the soil (N=3): *Maianthemum bifolium, Glechoma hirsuta, Leucanthemum vulgare*. In contrast, some species found at the site prefer very fertile (high nitrogen content) in the soil (N=9): *Chaerophyllum aureum* (low frequency in July 1972), *Sambucus nigra* (juveniles) (low stable frequency during sampling periods). This analysis shows the high variability of nitrogen input in the forest over decades.

Forest stability is defined as the ability of the system to persist through its component populations that show adaptability to environmental conditions (Puhe and Ulrich 1994). Long-term studies in beech-fir forests are still not long enough relative to the longevity of the trees themselves. Despite this limitation, some authors (Diaci et al. 2007; Heiri et al. 2009; Petritan et al. 2015) tested various hypotheses and in developed new theories that might help our understanding of mixed beech-coniferous forest dynamics.

CONCLUSIONS

Piatra Arsă forest comprise a low number of mature trees with large diameters, but over 37 years, the total density of mature tree individuals decreased substantially and the balance in abundance between the two dominant species changed through reduction of *Abies alba* and increase of *Fagus sylvatica*. The within-species diameters vary greatly with the age of individuals. The inter-specific variation is higher, the average diameters of fir individuals being larger than those of beech individuals in both layers. The high vertical diversity and heterogeneity is characteristic of an uneven-aged (old-growth) forest.

The undergrowth layer was well-developed and mirrored the pattern of tree layer, except that *Abies alba* was better represented. After 37 years, the juveniles (saplings and seedlings) of fir (*Abies alba*) showed a greater abundance than those of beech (*Fagus sylvatica*) and sycamore maple (*Acer pseudoplatanus*).

The seedlings of dominant trees have a great variation among consecutive years and in long time. Long-time investigation showed an increase in taxonomic diversity of seedlings.

The frequency and diversity of species in the herbaceous layer varied greatly among the semi-permanent plots established in the 1 ha forest plot. As a proportion of the overall ground layer flora, relatively few species appeared or disappeared over the 37 years of the study. The number of species varies significantly from a season to another and from one year to another, pointing to active dynamics within the herbaceous layer.

The variation of richness and evenness is significantly between timings of observation and from one month to another (spring-May and summer-July) in the same year, but species dominance did not significantly differ in time.

Seedlings of *Abies alba* and *Fagus sylvatica* display a higher abundance in spring but diminished in summer, *Acer pseudoplatanus* seedlings had a vice versa display. However, the abundance of fir and beech seedlings remained stable, whereas the abundance of sycamore maple seedlings increased in 37 years.

The values of ecological indicators reveals that in Piatra Arsă forest all species are associated with the mountainous or sub-mountainous regions of Romania and are characteristic to the micro-habitats created naturally in the forest. The frequency of the recorded species (clustered and random) is determined by site conditions.

Our study highlighted that Piatra Arsă site is notably species-rich but with variation in species number dependent on season and inventorying year and showing a very dynamic herbaceous layer.

Within the study site, there is variation of the environmental factors at micro-site level, and this is reflected in changes of species in time and space, related in part to the vegetation season and species life cycle or between periods. At the forest level, the impact of environmental factors is quite stable, with no drastic changes happening in the herbaceous vegetation structure.

Except for the shift in tree species dominance after 37 years under conservation, the Piatra Arsă forest displayed rather small structural changes (shifts of the micro-habitats), which may be attributable to natural fluctuations characterising a natural old-growth forest.

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