

Impact of deforestation and different agronomical management regimes on biodiversity of mountain ecosystem

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Abstract

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The experiment was carried out on a mountain grassland established at an altitude of 845 m in 2006–2008, after deforestation of naturally reforested pasture. The aim of research was to examine the changes in plant diversity and concentration of ergosterol in above-ground phytomass of grassland with different regimes of management. The increasing of phytodiversity on variants P (grazing), KP (grazing followed by mowing) and BM (control) during 3 years after deforestation is connected with a relatively low proportion of grasses dominated by *Agrostis capillaris*. A high percentage of legumes, particularly *Trifolium repens*, was the only presented on variant VP (grazing on burn plots). Higher number of species on variants KP and PP (grazing after reseeding with clover-grass mixture) resulted in a higher index of diversity. The values of Shannon's index of diversity (H) increased the most on variant PP over the years, where high-value species of grasses sown also influenced the grassland quality (E_{GQ}). The lowest values of H were achieved on variant VP, where the grassland quality value (E_{GQ}) was surprisingly the highest due to the dominance of *Trifolium repens* monoceonosis. On variant BM (without management) the diversity index has decreased due to reforestation (7 kinds of plants). The concentration of ergosterol in the above-ground phytomass of grassland depended on quantity of pathogenic fungi in the soil, and moisture conditions in the autumn of 2006–2008.

Key words

Cambisol, deforestation, ergosterol, forage quality, fungi, grassland diversity

Introduction

Grasslands, important ecosystems in Europe, are serving essential functions by producing feed and providing other ecosystem services (SMIT et al., 2008). They are a very important part of landscape, markedly contributing to biodiversity (ISSELSTEIN et al., 2005). Central and Eastern Europe has a relatively rich biodiversity in respect to geological, historical and economic factors

(YOUNG et al., 2007), such as the grassland communities in Slovakia with the highest phytodiversity, containing up to 74 vascular plant species per 1 m², ranking among the communities with the highest species diversity in Europe (NOVÁK, 2008). The processes in naturally non-forest and man-made deforested ecosystems are usually studied in tropical (JACKSON and ASH, 1998; NANGENDO et al., 2005) and subtropical (FYNN et al., 2004; FAIRFAX et al., 2009) areas, especially in savanna stands (SCHOLLES

and ARCHER, 1997; REICH et al., 2001), where fire plays an important role in formation of open woodland ecosystems. This type of disturbance is an inevitable factor for grassland regeneration. The local deforestation can lead to an increase of species diversity in a grassland community (NOVÁK, 2009).

Soil texture does not influence only the contamination load by cattle, but also the quality and quantity of above-ground phytomass (ACHILLES et al., 2002). The traditional management of semi-natural grasslands is based on low soil loading caused by cattle (ISSELSTEIN et al., 2005). In case of a year-round cattle breeding, the load on pasture lands varies over the year. The trampling by cattle enables contact between damaged assimilatory plant organs and soil.

Soil is considered to be the refuge for almost all main taxonomic groups of fungi (THORN, 1997). They develop in the above-ground phytomass of grassland in autumn. Fungus and blight growth can be supported by dense vegetation as well as high air humidity (GIESLER et al., 1996). Fungal mycelia release mycotoxins attacking dead bodies, over seasoned and lodged plants in the humid environment (WHEELER, 1968; OPITZ VON BOBERFELD, 1997, 2000). Their negative influence is expressed by health problems, reduction of food intake, diarrhoea, reduction of animal production efficiency, etc. (KALAČ and MIKA, 1997; DEVEGOWDA, 2002). Ergosterol is a prevailing steroid fungus component and an important membrane component. It also serves as a chemical indicator for quantitative determination of actual mycelia condition caused mainly by species as: *Penicillium*, *Aspergillus*, *Fusarium*, *Alternaria*, *Mucor* (SHAPIRO and GEALT, 1982; YOKOKAWA, 1987; WEETE, 1989).

The aim of this study was to describe initial vegetation changes caused by deforestation of a formerly naturally reforested temperate pasture and different agronomical regimes. There were examined the following features: plant species composition, occurrence of fungi species in soil, diversity of plant species expressed by Shannon index (H) and grassland quality (E_{GQ}).

Material and methods

Study site

The experiment was conducted on a reforested pasture in the Slovenské Rudohorie Mts (48°33' N, 19°46' E) in 2006–2008. The altitude of the locality is 845 m a.s.l., the average annual precipitation reaches 927 mm and the mean annual temperature is 5.1 °C. The soil type is a Cambisol (KMm^c). The chemical characteristics of the soil determined in 2006–2008 are shown in Table 1.

The experimental area (formerly common land) was regularly grazed by sheep and cattle for centuries. In the 1950s, when the large-scale agriculture management was established, this low productive pasture was abandoned and naturally reforested. After more than 50 years a woody plant community composed of *Betula pendula*, *Corylus avellana*, *Populus tremula* and *Fagus sylvatica* was developed. Herbaceous vegetation consisted mainly of *Fragaria vesca*, *Galium odoratum*, *Cruciata laevipes*, *Veronica chamaedrys* and *Avenella flexuosa*. In 1993 grazing management by Charolais cattle started in this forest. The stocking rate varied from 0.30 to 0.60 cattle unit per hectare. Two grazing cycles were applied during the experiment.

The experimental area was established, and tree and shrubs were cut down and removed in May 2006. There were five treatments applied: P – grazing, KP – grazing after mowing, PP – grazing after seeding of clover-grass mixture, VP – grazing on burnt area, BM – unmanaged control (Table 2).

Table 2. Experimental variants

Variant	Applied treatment
P	Grazing
KP	Grazing followed by mowing
PP	Grazing after reseeding with clover-grass mixture
VP	Grazing on burnt plot
BM	Unmanaged control

Table 1. Chemical characteristics of the studied Cambisol

Horizon	Depth [m]	C _{ox} [%]	Hm [%]	HK/FK	pH		mmol kg ⁻¹			V [%]
					H ₂ O	KCl	H	S	T	
Aoq	0–0.05	2.59	4.46	0.42	4.94	4.79	73.80	64.30	138.10	46.60
Bv ₁	0.06–0.45	1.66	2.86	0.51	5.19	4.95	63.90	50.00	113.90	43.90
Bv ₂	0.46–0.75	0.81	1.40	0.65	5.14	4.90	43.80	62.70	106.50	58.90

Aoq, humus ochric silicate horizon; Bv, cambic horizon (horizon of intrasoil weathering); Cox, organic carbon; Hm, humus; pH/H₂O, active soil reaction; pH/KCl, exchangeable soil reaction; HK/FK, ratio of humic to fulvic acids; H, hydrolytic acidity; S, sum of exchangeable basic cations; T, total sorption capacity; V, base saturation percentage.

The experiment was arranged in three randomized blocks (5 treatments \times 3 replicates = 15 plots) with study plots 2 m \times 2 m in size. The plots of PP variant were reseeded with grass/clover mixture (seeding rate 12.64 kg ha⁻¹) consisting of *Festuca pratensis*, *Phleum pratense*, *Poa pratensis*, *Dactylis glomerata*, *Trifolium repens* and *Lotus corniculatus* (Table 3).

Table 3. Composition of clover-grass mixture

Species – Variety	Share in mixture [%]	Quantity of seeds [kg ha ⁻¹]
<i>Festuca pratensis</i> Huds. – Otava	19.51	3.96
<i>Phleum pratense</i> L. – Sobol	12.20	1.24
<i>Poa pratensis</i> L. – Slezanka	12.20	1.65
<i>Dactylis glomerata</i> L. – Rela	7.32	0.99
<i>Trifolium repens</i> L. – Ovčák	41.45	4.21
<i>Lotus corniculatus</i> L. – Polom	7.32	0.59
Σ		12.64

Field activities and laboratory analyses

Phytocenological records were made on 15 permanent subplots (1 m \times 1 m) placed in the central parts of the study plots (2 m \times 2 m). The percentage cover (KLAPP, 1965) of all vascular plant species over the area of 1 m² was recorded. The phytocoenoses were estimated visually before grazing in May of each year. An initial estimation was made in the spring of 2006 – to obtain baseline data for each plot. The nomenclature of plant species follows MARHOLD and HINDÁK (1998). The floristic composition of grassland expressed in percents also serves for evaluation of the grassland quality (E_{GQ}), by NOVÁK (2004). For evaluation of floristic diversity we used the Shannon's index of diversity (H) according to BEGON et al. (1997).

Herbs under protective cages (0.50 m \times 0.50 m) were harvested before each grazing cycle on each study plot, with exception of the phytocenological subplots. After the harvest, the cages were replaced into another part of the study plot. The pasture lands were grazed by beef cattle of the Charolais breed. Above-ground phytomass samples were taken in 2006–2008 at the end of October and at the beginning of November (before snowing) in three repetitions. They were dried at 60 °C, homogenised (200 g) and sent to the JLU Giessen laboratory (Germany). The concentration of ergosterol was determined after extraction by High Performance Liquid Chromatography and UV detector according to SCHWADORF and MÜLLER (1989).

Data analyses

Repeated measurements were carried out for evaluation of the species diversity and functional group data by ANOVA. The community response was analysed with

using constrained ordinations. The redundancy analysis (RDA) by LEPŠ and ŠMILAUER (2003) in the CANOCO package (TER BRAAK and ŠMILAUER, 2002) followed by a Monte Carlo permutation test was used to evaluate trends in plant species composition, because of linear species responses and a rather homogenous species composition over the plots (LEPŠ and ŠMILAUER, 2003). A split plot design was used in the permutation type to cope with repeated measurements. We used 999 permutations in all performed analyses restricted to three split-plots, freely exchangeable whole plots, time series or linear transects at the split-plot level. Our data consisted of repeated observations of the baseline data (measurements performed before the grazing), in consequence of the interaction of treatments and year, were the most important variables. The sum of 100 subplots was used for statistical evaluation. Centring by species was applied. A standard biplot ordination diagram constructed by the CanoDraw program (TER BRAAK and ŠMILAUER, 2002) was used to visualize the results of the CANOCO analyses.

Results and discussion

Content of ergosterol in above-ground phytomass

LABUDA (2007) found the following fungal genera in a soil depth up to 0.75 m: *Penicillium* (14 species), *Trichoderma* (4 species), as well as *Mucor* and *Fusarium*. The soil contained also some other fungal species, such as *Cladosporium* (3 species), *Mortierella* (2 species) and species of genera *Absidia*, *Acremonium*, *Gelatinospora*, *Rhizopus*, *Talaromyces*, *Umbelopsis* and *Zygorrhynchus*. We assume that fungi in favourable soil conditions attack the above-ground phytomass (mainly *Penicillium* – most numerous species).

Ergosterol is a component of fungal cell membranes, and it is usually used as an indicator of fungal biomass in soil. Its concentration in above-ground phytomass of studied plots ranged from 62 to 323 mg kg⁻¹, depending on the moisture conditions in the autumn of 2006 and the variant of experiment (Fig. 1). We have put links between the ergosterol levels found in the above-ground phytomass of grassland and the presence of pathogenic fungi in soil, as it has been already reported by BEDRNA (2002) and JAVOREKOVÁ et al. (2008). Filamentous microscopic fungi support soil aggregates formation, but their pathogenic species can exert also negative influence on plants' growth (KRNÁČOVÁ et al., 2008). An environment favourable for fungi was created in long-lasting wet and cold weather periods in the different years. Increased occurrence of fungi in the soil was reflected in increased level of ergosterol in the above-ground phytomass. WOLF (2002) ascertained similar values in the above-ground phytomass of Festuco-Cynosuretum association (at altitude from

335 m to 460 m). We discovered fewer damaged parts of plants during the dry autumn in 2008. Ergosterol concentrations found in the above-ground phytomass in 2006 were significantly different those obtained in 2007 and 2008 (Table 4).

Floristic composition of grassland

In the studied area, fifty years after reforestation, was identified 31 herb species and 17 woody species. Due to mowing and cattle grazing the plant community *Agrostis-Festucetum rubrae* was dominated by *Agrostis capillaris* and *Festuca rubra* species for three years. GREMMEN et al. (1998) consider *Agrostis* spp. a fast-growing grass species, propagated by means of spurs. We agree with the author's argument, but also with the opinion of ESPIE and BARRATT (2006), that filling empty places in grasslands and creating stands by the species *Agrostis* spp. and *Festuca rubra* is influenced by environmental factors (altitude, moisture, temperature, light, high seed production, but mainly vegetative reproduction by spurs). Regarding the legumes, *Trifolium repens* species dominated. YU et al. (2008) found out that grazing of light-demanding grassland increases the portion of white clover. Among the other herbs species, the presence of *Lysimachia nummularia* and *Fragaria vesca* was significant in the first years. These species are more frequent in wet, shady and shrubby stands. The light-demanding plants began gradually to put across themselves, especially *Taraxacum officinale*, *Leontodon autumnalis* and others (Fig. 2).

During the first three years after deforestation, the proportion of grass species in phytocoenosis was relatively low (average 26.16%). Dominance of *Agrostis capillaris* was recorded in variants P, KP and BM. The highest proportion of this species was recorded on the plot with treatment KP (17.06%). Legumes had a high proportion on variant VP (up to 80.23%). Other herbs had higher proportions than the grasses (on average 44.25%) on variants P and KP. KRAJČOVIČ (1968) and BUCHGRABER (2002) presented that optimal proportion of high-quality grassland for animal feeding represents 50–60% grasses, 10–30% legumes, and the rest are other herbs. The highest number of species were on variants KP and PP (41), the lowest number was on the burnt out variant VP, where we recorded only 25 species and dominance of *Trifolium repens*, because of a higher content of ash in the soil (P, K nutrients). The unmanaged variant BM was left to self-growth and formed in the opposite way as the other plots influenced by mowing and grazing. After three years, the community left without treatment was composed of 33 plant species, from them 7 were woody species. The remarkable successional development of this community independent of all treatments as well as significant differences among the treatments was detected. The woody vegetation (*Populus tremula*, *Betula pendula*, *Cerasus avium*, *Carpinus betulus*, *Rosa* spp., *Rubus* spp.) tall grasses (*Poa trivialis*, *Avenella flexuosa*, *Calamagrostis epigjos*) and tall forbs (*Viscaria vulgaris*, *Galium mollugo*) had a higher abundance on the BM variant. We note, in agreement with LUOTO and PYKÄLÄ (2003) that woody

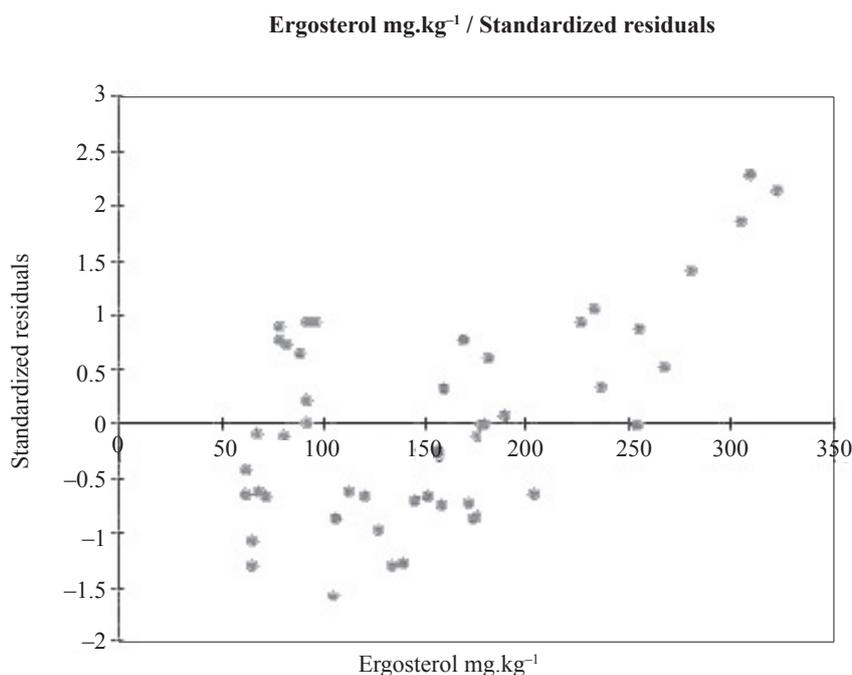


Fig. 1. Content of ergosterol in above-ground phytomass of differently treated grassland plots.

species left to self-growth are aggressive, their reproduction is fast and their succession is directed towards reforestation.

Table 4. Significance of difference in ergosterol content

Compared years	Ergosterol [mg kg ⁻¹]	
	Pr. > Diff	Significant
2008 ~ 2006	<0.0001	Yes
2008 ~ 2007	0.068	No
2007 ~ 2006	<0.0001	Yes

After introduction of grazers, the plant species richness per 1 m² increased. The grazing had a positive effect on species richness compared to the unmanaged grassland. Due to an ash layer and excessive nutrient content in the topsoil layer, the presence of vascular

plant species on the burnt plots was quite low. Only three years after abandonment of deforested sites, fast colonisation by shrub species (*Rubus idaeus*, *Rosa* spp.) and trees was re-started (especially by *Populus tremula*). Such afforestation is associated with a decline of grassland flora – as reported in many studies (MILBERG, 1994; SMITH AND RUSHTON, 1994; HANSON AND FOGELFORS, 2000; PYKÄLÄ et al., 2005). On the other hand, any type of defoliation management can prevent reforestation with sufficient efficiency. Because of creeping stems and clonal growth of woody species, the probability of removing the majority of above-ground biomass by defoliation is low, and the *Trifolium repens* species has an advantage to quickly colonize the bare ground (THÓRHALLSDÓTTIR, 1990). This was also the case of VP variant.

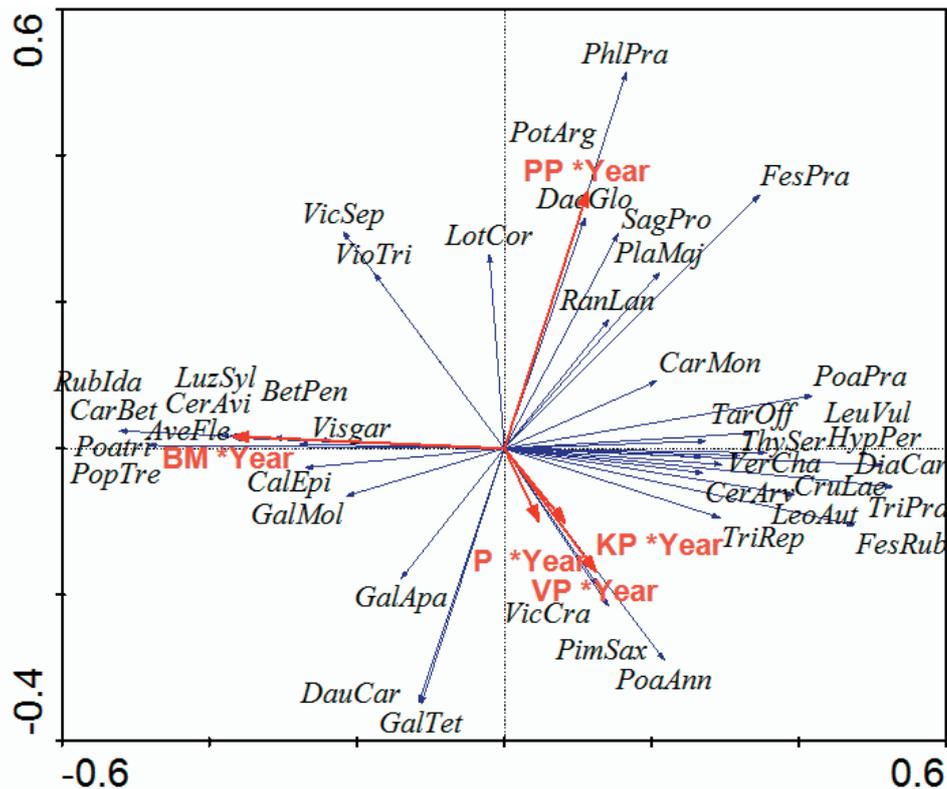


Fig. 2. Ordination diagram showing the results of RDA analysis.

Explanations:

P, grazing; KP, grazing followed by mowing; PP, grazing after reseeded; VP, grazing on burning; BM, unmanaged control; RubIda, *Rubus idaeus*; CarBet, *Carpinus betulus*; Poatr, *Poa trivialis*; PopTre, *Populus tremula*; LuzSyl, *Luzula sylvatica*; CerAvi, *Cerasus avium*; veFlex, *Avenella flexuosa*; BetPen, *Betula pendula*; Visgar, *Viscaria vulgaris*; CalEpi, *Calamagrostis epigejos*; GalMol, *Galium mollugo*; GalApa, *Galium aparine*; DauCar, *Daucus carota*; GalTet, *Galeopsis tetrahit*; PoaAnn, *Poa annua*; PimSax, *Pimpinella saxifraga*; VicCra, *Vicia cracca*; FesRub, *Festuca rubra*, TriRep, *Trifolium repens*; LeoAut, *Leontodon autumnalis*; TriPra, *Trifolium pratense*; CruLae, *Cruciata leavipes*; CerArv, *Cerastium arvense*; DiaCar, *Dianthus cartusianorum*; VerCha, *Veronica chamaedris*; HypPer, *Hypericum perforatum*; ThySer, *Thymus serpyllum*; LeuVul, *Leucanthemum vulgare*; PoaPra, *Poa pratensis*; TarOff, *Taraxacum officinale*; CarMon, *Carex montana*; RanLan, *Ranunculus lanuginosus*; PlaMaj, *Plantago major*; SagPro, *Sagina procumbens*; FesPra, *Festuca pratensis*; DacGlo, *Dactylis glomerata*; PotArg, *Potentilla argentea*; PhlPra, *Phleum pratense*; LotCor, *Lotus corniculatus*; VioTri, *Viola tricolor*; VicSep, *Vicia sepium*.

The successional development of grasses was similar, with exception of variant BM, the shrubs and trees on which suppressed their growth in the second year. However, the behaviour of dominant grasses was different. *Festuca rubra* showed a similar progress in all applied variants of treatments except VP variant. This species is known as a very adaptable under different management, and, therefore, it can well perform in various types of grasslands (HONSOVÁ et al., 2007; MAŠKOVÁ et al., 2009; KESTING et al., 2009; LANTA et al., 2009). The cover of the *Agrostis capillaris* species was supported by grazing or cutting/grazing management, on the other hand, it was reduced by reseeding a productive grassland mixture, excessive nutrient supply in soil after phytomass burning as well as by competitive abilities of shrubs on the unmanaged plots. This short grass species (GRIME, 1987) is known as an indicator of young grasslands (WAESCH and BECKER, 2009), being common especially in low productive temperate grasslands (HELLSTRÖM et al., 2003; LOUAULT et al., 2005; PAVLŮ et al., 2007; MAŠKOVÁ et al., 2009).

Ecological values in above-ground phytomass

The increase of Shannon's index of diversity (H) and value of grassland quality (E_{GQ}) was recorded on plots of P, KP and PP variants over three years (Table 5). The value of Shannon's diversity index (H) was the highest in case of PP variant (2.68). This was connected with relatively high species richness. The lowest diversity index was obtained on VP variant, where the E_{GQ} value was the highest due to the dominance of high-quality legume monoceonosis of *Trifolium repens* species. The values of diversity index over the years increased, except to the BM variant where the decrease was recorded due to reforestation. On variant PP with sown high-quality species (*Phleum pratense*, *Festuca pratensis*, *Dactylis glomerata*, *Poa pratensis*, *Trifolium repens*) was recorded a high value of E_{GQ} (58.92). BULLOCK et al. (2007) note that it is not possible to generalize that only sowing of a fodder plant mixture can increase the diversity of the species community. We agree with the opinion of BEGON et al. (1997) that the species diversity significantly affects not only the frequency, but also the cover of species occurring in the community and also with ISSELSTEIN et al. (2005) that use of grasslands can

preserve the plant species richness of the grassland. It resists the high competitive stress from invasive species and woody vegetation from the neighbouring areas.

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Table 5. Shannon index of diversity (H) and evaluation of grassland quality (E_{GQ})

Year	P		KP		PP		VP		BM	
	H	E_{GQ}								
2006	1.69	24.19	1.70	23.92	1.49	41.57	0.53	26.15	2.16	29.87
2007	2.14	39.03	2.29	36.57	2.44	58.92	0.75	74.14	1.92	19.34
2008	2.31	44.17	2.42	48.17	2.68	71.11	0.81	72.73	1.13	13.77

P, grazing; KP, grazing followed by mowing; PP, grazing after reseeding; VP, grazing after burning; BM, unmanaged control.

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Vplyv odlesnenia a rôznych agronomických opatrení na biodiverzitu horského ekosystému

Súhrn

Experiment bol realizovaný na odlesnenom horskom pasienku v 845 m n. m v rokoch 2006–2008. Na variantoch s pasiením (P), kosením a následným pasiením (KP) a bez manažmentu (BM) bol 3 roky po odlesnení pomerne nízky podiel tráv s dominanciou druhu *Agrostis capillaris*. Vysoké zastúpenie leguminóz, predovšetkým druhu *Trifolium repens*, bolo len na spálenisku (variant VP). Vyšší počet druhov na variantoch využívaných kosením a pasiením (KP) a pasiením po príseve ďatelinovo-trávnej miešanky (PP) sa odrazil vo vyššom indexe diverzity. Hodnoty hydrolytickej kyslosti sa v priebehu rokov najviac zvyšovali na variante PP, kde vysiate vysokohodnotné druhy ovplyvnili aj kvalitu pastvy (E_{GQ}). Najnižšie hodnoty H boli na variante VP, aj keď hodnota E_{GQ} bola najvyššia v dôsledku dominancie monocenózy druhu *Trifolium repens*. Na variante BM ponechanom na samovývoj (bez manažmentu) poklesli hodnoty vplyvom prirodzenej obnovy drevín (7 druhov drevín). Koncentrácia ergosterolu v nadzemnej fytomase trávneho porastu súvisela s existenciou patogénnych húb v pôde a závisela najmä od vlhkostných podmienok v jesennom období 2006–2008.

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