Species composition of an alluvial meadow after 40 years of applying nitrogen, phospohorus and potassium fertilizer

Druhové složení aluviální louky po 40 letech hnojení dusíkem, fosforem a draslíkem

Dagmar H o n s o v á[1], Michal H e j c m a n[2*], Michaela K l a u d i s o v á[1], Vilém P a v l ů[3], Dana K o c o u r k o v á[1] & Josef H a k l[1]

[1]Department of Forage Crops and Grassland Management, Czech University of Life Sciences, Kamýcká 129, CZ-165 21 Praha 6 – Suchdol, Czech Republic; [2]Department of Ecology and Environment, Czech University of Life Sciences, Kamýcká 1176, CZ-165 21 Praha 6 – Suchdol, Czech Republic, e-mail: hejcman@fle.czu.cz; [3]Department of Plant Ecology and Weed Science, Crop Research Institute Prague – Ruzyně, Rolnická 6, CZ-460 01 Liberec, Czech Republic
**Corresponding author*

Honsová D., Hejcman M., Klaudisová M., Pavlů V., Kocourková D. & Hakl J. (2007): Species composition of an alluvial meadow after 40 years of applying nitrogen, phospohorus and potassium fertilizer. – Preslia 79: 245–258

In 1966, the Černíkovice experiment, Czech Republic, was started when an alluvial meadow dominated by *Alopecurus pratensis* was subjected to the following fertilizer treatments: non-fertilized control, PK, N_{50}PK, N_{100}PK, N_{150}PK, and N_{200}PK. The experimental plots were cut three times per year in the initial phase of the experiment and twice per year since the late 1980s. In mid May 2005, plant cover was visually estimated, biomass yield and sward height measured in order to detect changes in the grassland ecosystem caused by this long-term fertilizer treatment. After 40 years treatment was a significant predictor of sward structure, explaining 32% of the variability in plant cover data in RDA. Legumes were not detected in the N_{200}PK treatment and achieved the highest cover in the control and PK treatment. Grasses had the lowest cover in the PK treatment and control, which significantly differed from all treatments with N. *Alopecurus pratensis* prevailed in all N_xPK treatments. Herbs had the highest cover in the control followed by the PK treatment and both treatments significantly differed from the N_xPK treatments. *Achillea millefolium* was recorded in all treatments, but the highest cover was recorded in the control treatment. Species richness of vascular plants ranged from 8 per m^2 in the N_{200}PK treatment to 24 in the control. A significant decrease in species richness with increase in sward height was detected. The cover of mosses ranged from 1 to 6% like sward height gradually increased with fertilizer application. Aboveground biomass yield was significantly lower in the control than all other treatments. Based on the results of the Černíkovice experiment and a comparison with other long-term fertilizer experiments it is concluded that naturally highly productive grasslands are much less threatened by the inappropriate application of fertilizers than low productive grasslands with a specific plant species composition.

K e y w o r d s : Czech Republic, long-term fertilizer experiment, nitrogen, phosphorus, potassium, species richness, sward height

Introduction

Long-term grassland fertilizer experiments were in most cases set up to determine whether different fertilizers improve the yield and quality of forage (Schellberg et al. 1999, Silvertown et al. 2006, Spiegelberger et al. 2006). In Central European countries, a series of long-term experiments aimed at investigating the rational use of N fertilizers were founded by agronomists in the 1960s and 1970s. According to the authors' information, the following experiments are still running: three in Austria (Trnka et al. 2006), two in Poland (Niczyporuk & Jankowska-Huflejt 2000, Kopeć & Gondek 2002) and one in the Czech Republic (Mrkvička et al. 2006). These experiments consist of an unfertilized control and N_xPK treatments, where the level of applied N ranged up to 400 $kg \cdot ha^{-1}$. This research focused predominately on nutrient leaching and cycling, biomass production, and forage quality. Although the results of these experiments are used to address basic questions about the ecology of grassland species, the effect of fertilizer on plant species composition has been ignored except for one of these experiments (Galka et al. 2005).

In the Czech Republic, most long-term fertilizer experiments, covering a gradient of ecosystem productivity from *Nardus stricta* to *Alopecurus pratensis* grasslands, ceased in the early 1990s as a consequence of agriculture restructuring caused by political change. No paper describing plant species composition was published for the series of experiments and it is incomprehensible that monitoring plots in these unique experiments in most cases were not permanently marked, so preventing study of residual after-effects of fertilizers on grassland functioning. The only exception is an experiment in Závišín, close to the town of Mariánské Lázně, where an increase in species number is reported after the cessation of fertilizer treatments (Královec & Prach 1997).

It is known that nutrients have a great influence on plant species composition and biomass production of a sward. The following generalizations are based on the results of numerous studies (Klečka & Fabian 1935, Klapp 1938, Larin 1956, Regal & Krajčovič 1963, Tilman 1982, Di-Tommaso & Aarsen 1989, De Kroon & Bobbink 1997): application of P and K increases the proportion of legumes; N application, when the P and K concentration in the soil is adequate, increases the proportion of grasses, predominantly tall ones, and reduces the proportion of legumes and short herbs.

The effects of short and long-term fertilizer applications on plant species composition frequently differ, hence the importance of long-term studies in plant ecology (Silvertown 1980, Güsewell et al. 2002, Hejcman et al. 2007b). A fertilizer experiment established in an alluvial meadow near Prague (Velich 1986) and reported in this paper is probably the longest still running well designed fertilizer experiment established in a wet meadow in Central Europe.

The aim of this study was to describe plant species composition and species richness after 40 years of fertilizer application. Within this context, the following question was addressed: How is plant species richness, sward height, biomass yield and plant species composition affected by long-term applications of N, P and K fertilizers?

Material and methods

Experimental setup

In 1966, a fertilizer experiment was set up in the Czech Republic at 363 m a.s.l. in a flat alluvial meadow near the village of Černíkovice, 35 km south of Prague (49° 47' N, 14° 45' E) (Velich 1986). At the study site, mean annual precipitation and temperature were 600 mm and 8.1°C, respectively (Benešov meteorological station 1961–2005). The soil type is a fluvisol/gleysol with a loamy texture and a $pH_{(KCl)}$ of 6.0. The experimental area was limed occasionally when a decrease in soil pH was detected. The depth of the underground water table varied between 0.1 and 0.7 m during the growing season and flooding occurred on several occasions. According to phytosociological nomenclature (Botta-Dukát et al. 2005) the vegetation of the experimental grassland was classified as *Deschampsion cespitosae*. At the start of the experiment, the alluvial meadow was dominated by grasses with a total cover of 68% (*Alopecurus pratensis* 17%, *Poa pratensis* 11%, *Festuca pratensis* 10%, and *Holcus lanatus* 7%). The cover of legumes was 11% (with the most abundant species *Trifolium hybridum, T. repens, T. pratense*) and that of other dicotyledonous species was 16% (with the most abundant species *Ranunculus repens*, *R. acris*, *Taraxacum* sp.). Before the first fertilizer application, aboveground biomass production under standard farm management was approximately 9.45 $t \cdot ha^{-1}$ of dry matter and the meadow was regularly treated with fertilizer (Velich 1986).

The experimental meadow was mown three times per year from 1966 to 1985 and since the late 1980s, only twice. The experiment was arranged in four randomized blocks each with the following treatments: unfertilized control, PK, N_{100}PK, N_{200}PK, N_{300}PK, N_{400}PK. N_{300}PK and N_{400}PK treatments were added in 1975. The area of individual plots was 5 × 6 m.

In the 1990s the dosages of N were reduced by half. The following treatments were then applied: control, PK, N_{50}PK, N_{100}PK, N_{150}PK, and N_{200}PK (Table 1, Fig. 1). Since the start of the experiment, the following fertilizers were used: saltpeter ammonium with lime ($NH_4NO_3 + CaCO_3$, 27.5% N, 10% Ca), super phosphate [$Ca(H_2PO_4)_2 + CaSO_4$, 8.5% P, 20% Ca, 10% S] and potash salt (KCl+NaCl, 50% K, 47% Cl). In each year, nitrogen fertilizer was applied in April and potassium and phosphorus fertilizers in October.

The cover of all the vascular plant species was visually estimated directly in percentages in each plot and cover estimates ranged from 0.5% to 100%. This was done before the first harvest in mid-May 2005. In each plot, two 1 × 1 m relevés were recorded (eight relevés per treatment, Fig. 1). Based on the descriptions of the vascular plants in the national flora (Kubát et al. 2002), all species within the study area were categorized according to their main traits. The following basic functional groups were recognized: legumes, herbs and grasses. Grasses and other herbs (not legumes) were further categorized, according to mean species height given in the national flora, into short graminoids, short herbs, tall graminoids and tall herbs (Appendix 1). The functional group rhizomatous grasses was as used by Velich (1986). To obtain total cover of functional groups, the covers of individual species belonging to a particular group were summed.

Actual sward height (ASH, syn. compressed sward height, Correl et al. 2003) was the mean of 12 measurements obtained using a rising plate meter within each 5 × 6 m plot.

To reveal the expected relationship between the average height of a particular species and its response to fertilizer treatment, the mean height of each species obtained from the local flora (Kubát et al. 2002) was weighted according to the total cover of each species in the relevé. This value is called the "potential sward height" (PSH; Pavlů et al. 2003). For example, the equation for a relevé composed of three species (A, B and C) is: PSH = [(cover of A × mean height of A) + (cover of B × mean height of B) + (cover of C × mean height of C)] / (cover of A + cover of B + cover of C). The intention was to generalize the reaction for all the sward components and investigate if some treatments stimulate the growth of short or tall plant species. Differences between actual and potential sward height were used as a measure of the so-called "environmental deficiency" in the different treatments. If this difference is positive, plants are generally taller than their mean height denoted in the national flora and vice versa.

Table 1. – Description of applied treatments and amounts of nutrients supplied annually.

Treatment	Applied nutrients [kg·ha^{-1}]					
	N	P	K	Ca	S	Cl
Control	0	0	0	0	0	0
P_{40} K_{100}	0	40	100	94	50	94
N_{50} P_{40} K_{100}	50	40	100	112	50	94
$N_{100}P_{40}K_{100}$	100	40	100	130	50	94
$N_{150}P_{40}K_{100}$	150	40	100	148	50	94
$N_{200}P_{40}K_{100}$	200	40	100	166	50	94

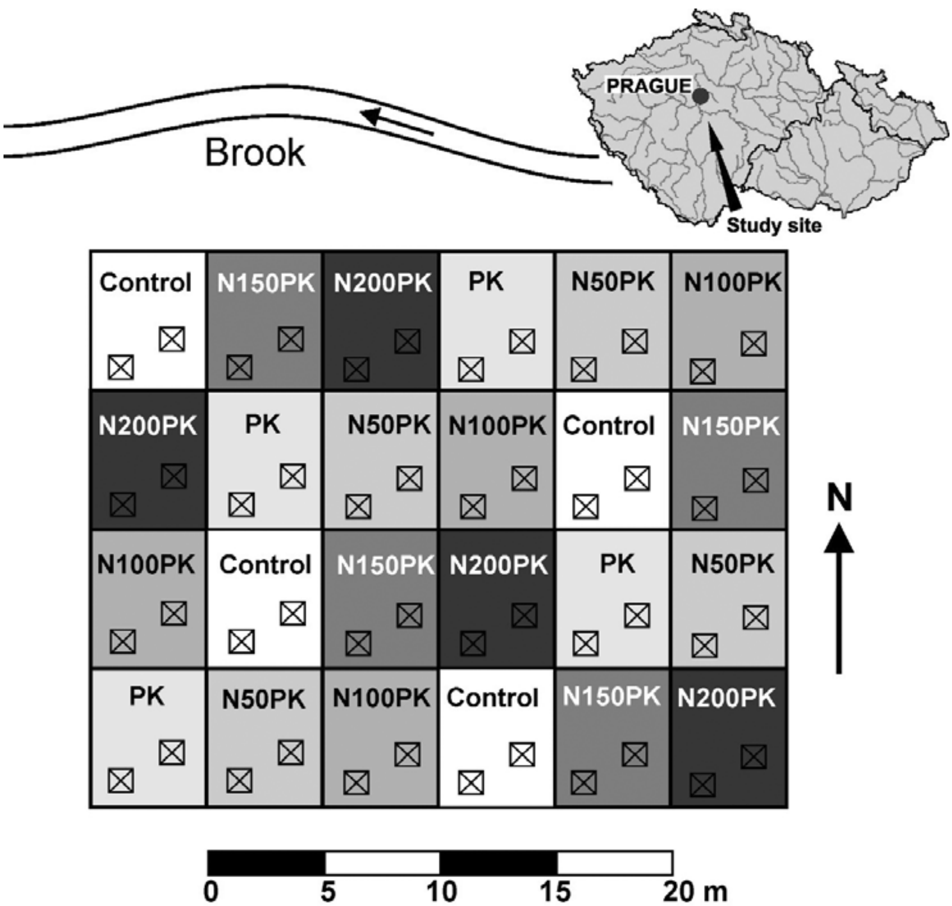


Fig. 1. – Location of the Černíkovice experiment in the Czech Republic and a diagram of the spatial arrangement in four randomized blocks. Positions of 1m^2 plots used for monitoring the vegetation are indicated by squares with diagonals.

Dry matter biomass yield was the sum of a first and a second harvest. In each plot, sward was mown by machine leaving a stubble height of 5 cm. The harvested biomass was immediately weighed and the percentage dry matter determined in the laboratory after 48 hours of drying at 85 °C. Biomass production per each plot was than recalculated as dry matter yield ($t \cdot ha^{-1}$).

Data analysis

Redundancy analysis (RDA) in the CANOCO 4.5 program (ter Braak & Šmilauer 2002) was used to evaluate multivariate plant cover data. This method was used because the data set was relatively homogeneous, and environmental variables and covariables were categorical. Monte Carlo permutation test with 999 permutations was used to reveal the significance of treatment effects and permutations were performed within blocks. The result of the RDA analysis was visualized in the form of an ordination diagram in CanoDraw program. All univariate analyses were performed using the STATISTICA 5.0 program (StatSoft 1995). One-way ANOVA followed by post-hoc comparison using the Tukey HSD test was applied to functional groups, species richness, ASH and PSH data. Relationship between ASH and plant species richness, and moss cover was evaluated by regression.

Results

In May 2005, 50 vascular plant species were recorded in the study area: 15 grasses, 27 dicotyledons, seven legumes and one *Luzula* species. Type of treatment was the most significant predictor of sward structure in the experimental area and explained 32% of the variability in plant cover (RDA, $F = 1.743$, $P = 0.002$).

The ordination diagram (Fig. 2) shows the divergence in species composition between the treatments without N (control, PK) and those with N_xPK. If the treatments with N were calculated separately, the main divergence is between the $N_{50}PK$ and $N_{100}PK$ treatments. Mean cover recorded for species is given in Appendix 1.

With the exception of short graminoids, treatment significantly affected the total cover of all functional groups in 2005 (ANOVA). Legumes were not detected in the $N_{200}PK$ treatment and achieved the highest cover in the control and PK treatment (Fig. 3). *Lathyrus pratensis* and *Trifolium repens* were the only legumes sporadically recorded in treatments with N up to a dose of 150 $kg \cdot ha^{-1}$. The functional group grasses, in contrast to herbs, had the lowest cover in the PK and control treatments, which significantly differed from all treatments with N. Short graminoids, *Agrostis stolonifera, Anthoxanthum odoratum*, and *Cynosurus cristatus*, were sporadically recorded only in the control, $N_{50}PK$, and $N_{100}PK$ treatments. Tall grasses, *Alopecurus pratensis* and *Poa pratensis* especially, prevailed in all N_xPK treatments and the lowest cover was recorded in the control and PK treatment. The percentage of rhizomatous grasses recorded by Velich (1986) along with the data collected in 2005 is presented in Fig. 4a. In the first 10 years, the percentage of rhizomatous grasses remained constant and thereafter there was a moderate decrease in the value of about 30%. This percentage was probably relatively stable because a similar value was recorded in 1983 and again in 2005. On the other hand, a great fluctuation was recorded in the PK treatment, with the peak in 1976–1977. After initial fluctuations, the most stable

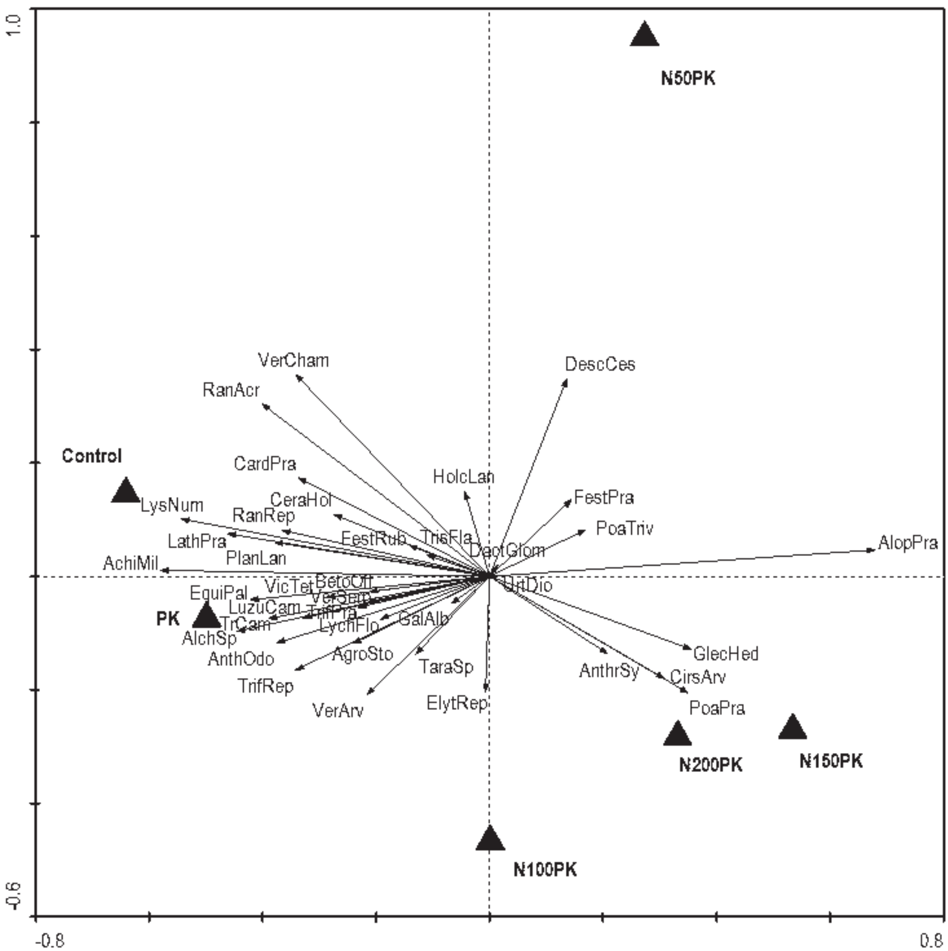


Fig. 2. – Ordination diagram showing the result of RDA analysis. Treatment abbreviations are given in Table 1. Species abbreviations: AgroSto – *Agrostis stolonifera*, AchiMil – *Achillea millefolium*, AlchSp – *Alchemilla* sp., AlopPra – *Alopecurus pratensis*, AnthOdo – *Anthoxanthum odoratum*, AnthrSy – *Anthriscus sylvestris*, BetoOff – *Betonica officinalis*, CardPra – *Cardamine pratensis*, CeraHol – *Cerastium holosteoides*, CirsArv – *Cirsium arvense*, DactGlom – *Dactylis glomerata*, DescCes – *Deschampsia cespitosa*, EquiPal – *Equisetum palustre*, ElytRep – *Elytrigia repens*, FestPra – *Festuca pratensis*, FestRub – *Festuca rubra*, GalAlb – *Galium album*, GlecHed – *Glechoma hederacea*, HolcLan – *Holcus lanatus*, LathPra – *Lathyrus pratensis*, LuzuCam – *Luzula campestris*, LychFC – *Lychnis flos-cuculi*, LysNum – *Lysimachia nummularia*, PlanLan – *Plantago lanceolata*, PoaPra – *Poa pratensis*, PoaTriv – *Poa trivialis*, RanAcr – *Ranunculus acris*, RanRep – *Ranunculus repens*, TaraSp – *Taraxacum* sp., TrifCam – *Trifolium campestre*, TrifPra – *Trifolium pratense*, TrifRep – *Trifolium repens*, TrisFla – *Trisetum flavescens*, UrtDio – *Urtica dioica*, VerArv – *Veronica arvensis*, VerCham – *Veronica chamaedrys*, VerSerp – *Veronica serpyllifolia*, VicTet – *Vicia tetrasperma.*

percentage of rhizomatous grasses exceeding 80% was recorded in the N_{200}PK treatment. Their dominance resulted in a simplification of sward structure as the cover of most other recorded species was very low.

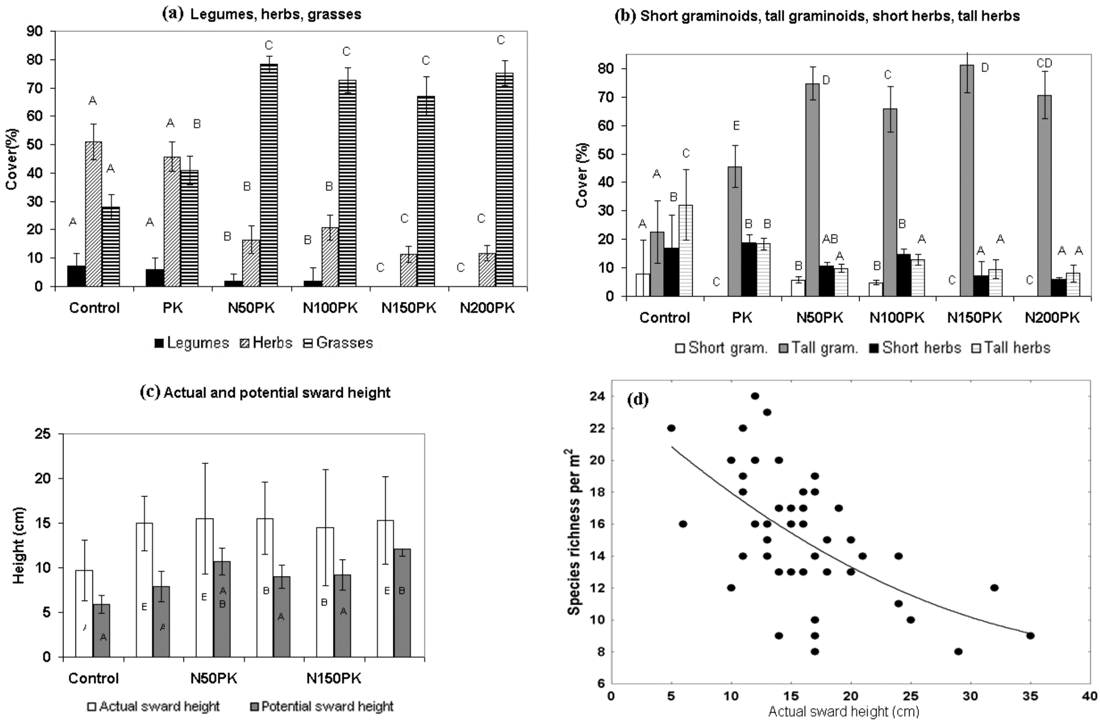


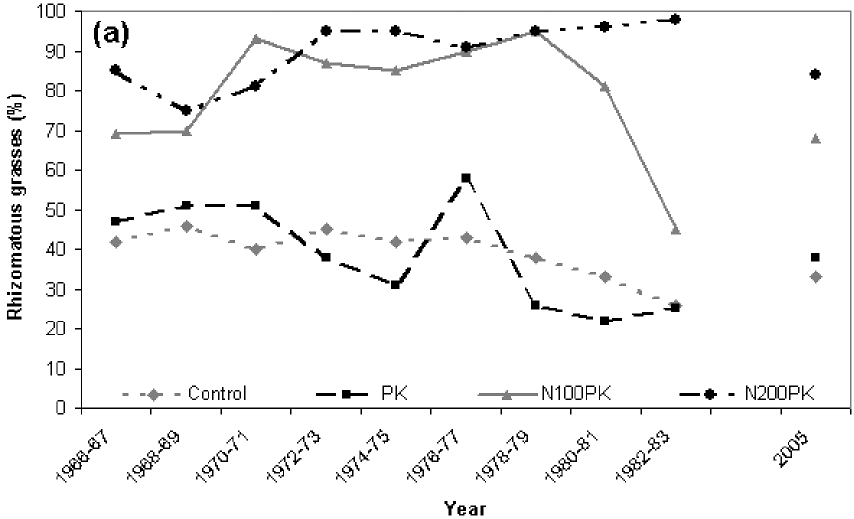
Fig. 3. – Percentage cover of different functional groups: (a) legumes, herbs and grasses; (b) short and tall graminoids, and short and tall herbs; (c) actual and potential sward height. (d) Plant species richness as a function of actual sward height. Treatment abbreviations are given in Table 1.

The herb functional group had the highest cover in the control followed by the PK treatment in 2005. Both these treatments differed significantly from all treatments with N. The highest cover of short herbs, *Lysimachia nummularia* and *Veronica chamaedrys* in particular, was revealed in the treatments without N, which differed significantly from other treatments. Tall herbs, *Achillea millefolium* especially, were recorded in all treatments, but the highest cover was found in the control, which significantly differed from other treatments.

Species richness of vascular plants ranged from 8 to 24 per 1 m^2 plot and the effect of treatment was significant. Species richness was highest in the control followed by the PK treatment and lowest in the N_{200}PK treatment, which differed significantly from the other treatments (Appendix 1). There was a significant decrease in species richness with increase in sward height ($r = 0.283$, $F = 4.01$, $P < 0.001$, Fig. 3).

The cover of mosses ranged from 1 to 6%, gradually increased with fertilization and was therefore highest in the N_{150}PK and N_{200}PK treatments. Significant increase in moss cover with increase in actual sward height was detected by regression ($r = 0.466$ $F = 26.123$, $P < 0.001$).

A significant effect of treatment on sward heights was revealed: heights were lowest in the control and actual sward height was higher than the potential in all treatments (Fig. 3). This indicated relatively suitable growth conditions in all treatments.



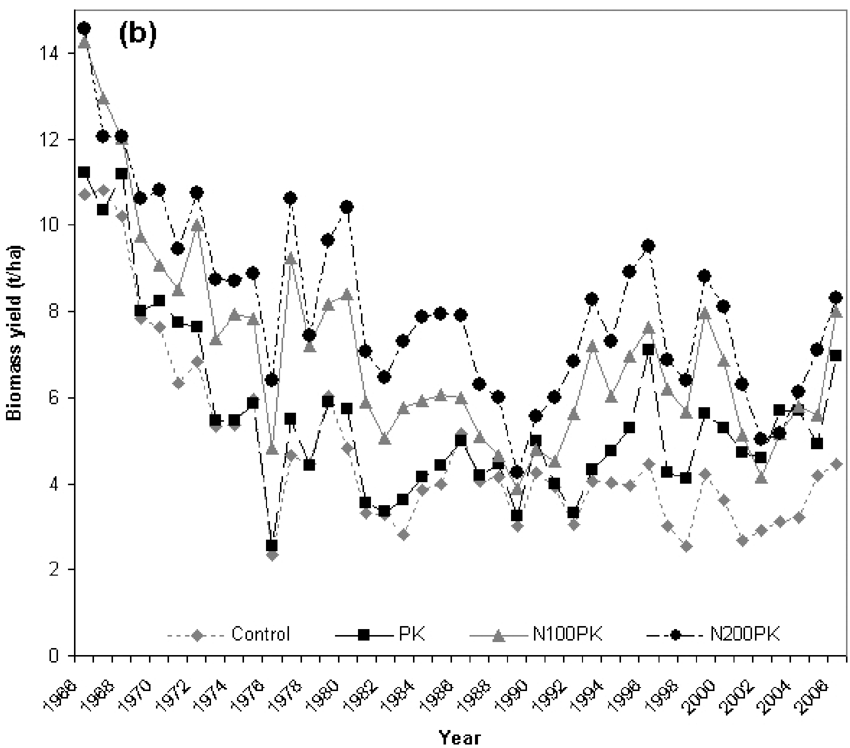


Fig. 4. – Long-term trend in the rhizomatous grasses, measured as two-year average percentage of the total cover (a) and biomass production in control, PK, N_{100}PK and N_{200}PK treatments (b). Baseline data were not determined before the application of fertilizer in 1965. Control and PK treatment plots remained the same from 1966 to 2006; Plots of N_{100}PK and N_{200}PK changed in 1990s because of the reduction in the N doses to half. Note that data from plots of former N_{200}PK and N_{400}PK treatments (now N_{100}PK and N_{200}PK treatments) are plotted for the period following this change in fertilizer application.

Aboveground biomass yield was lowest in control, which differed significantly from the other treatments in 2005 (Appendix 1). Figure 4b is based on data from several studies (Velich 1986, Mrkvička & Veselá 2002, Mrkvička & Veselá 2004) and that collected in 2005 and 2006. This comparison indicates that there was a (1) decrease in biomass yield in all treatments in the initial 10 years of the experiment, (2) high annual fluctuations in biomass yield contrasting with a relatively stable percentage of rhizomatous grasses in N_xPK treatments in particular, and (3) a divergence in the biomass production of the control and PK treatments, starting in the 25th year of the experiment.

Discussion

In Europe, a frequently discussed topic of agri-environmental policy is the amount of fertilizer that can be applied to grassland without affecting its structure and functioning. In the Černíkovice experiment, the relatively little effect of treatment on the presence of dominant grasses contrasts with the results of other long-term experiments on low productive grasslands, where the dominant species in the control disappeared from the fully fertilized treatments after several decades of nutrient application (Galka et al. 2005, Hejcman et al. 2007a). The low productive grasslands, with their specific plant species composition, are more threatened by fertilizer applications than naturally highly productive grassland. Although the initially dominant species persisted in all treatments, differing only in percentage cover, species richness was negatively correlated with the amount of N applied if P and K were not limiting. This indicates that high doses of N will also negatively affect plant species richness in naturally highly productive plant communities. This is consistent with the results of Prach (1993) who records a decrease in species richness from 50 to 27 after 40 years of intensive management of a wet meadow.

Although Velich (1986) investigated plant species composition regularly from 1967 to 1983, not only at Černíkovice, but also in six other experiments, collecting in total more than 890 relevés, this data has never been published. Velich (1986) concluded that initial changes in plant species composition caused by fertilizer application ceased within 3–6 years. The immediate response of the plant community is illustrated by increase in the cover of rhizomatous grasses from 68% before the start of the treatment to 80–98% in the first two seasons of the $N_{100}PK$ and $N_{200}PK$ application (Fig. 4a). This percentage did not change over the following 15 years. In 1982–1983 there was a decrease in rhizomatous grasses in the $N_{100}PK$ treatment, but the cause of this decrease is unknown. The grasses that responded positively to N_xPK were predominately rhizomatous, in particular *Alopecurus pratensis*, for which the highly positive and immediate effect of N application is known (Gaisler et al. 1998). In 2005, the tussock grass *Deschampsia cespitosa* was favoured by fertilizer application, however at high doses of N it was poorer competitor than *Alopecurus pratensis*. This species can be eliminated by ploughing and sward restoration, but not by the application of fertilizer or defoliation (Velich 1986).

In 2005, the absence of legumes in the two highest N treatments confirmed the previous results of Velich (1986), indicating that the decline of legumes in plots receiving high doses of N was caused indirectly by competition with tall grasses and directly due to their high sensitivity to the increased nitrate concentrations in the soil affecting the transport of assimilates from the leaves to underground organs. The only legumes able to survive moderate applications of N were *Lathyrus pratensis* and *Trifolium repens*. Tolerance of both these legumes to N fertilizer was recorded in the Rengen Grassland Experiment (Hejcman et al. 2007a). In 2005, the absence of a positive effect of the PK treatment on the cover of legumes in Černíkovice experiment contrasts with the conclusions of other authors (Dodd et al. 1994, Galka et al. 2005, Silvertown et al. 2006). It is suggested that a positive effect of P and K on the cover of legumes is most obvious for soils containing low concentrations of P and K at the start of fertilizer treatment (Velich 1986). Further, application of P and K in general increases the representation of legumes in grassland and year-to-year variability in the grass/legume ratio. There is no positive effect of P and K application on legumes in some years, although the mean percentage cover of legumes from 1969 to 1983 in the PK treatment at Černíkovice was 10.1% but only 4.2% in the control (Velich 1986). Fluctuations in the grass/legume ratio accord with the resource ratio hypothesis (Tilman 1982). In absence of limitation by P and K, nitrogen fixing legumes increase N availability, which favours the growth of grasses after N depletion.

The occurrence of the highest cover of herbs in the non-fertilized control accords with results of other long-term experiments (Silvertown et al. 2006, Hejcman et al. 2007a). Herbs, predominately short ones, compared to grasses, possess a lower competitive ability under intensive application of N, P and K fertilizers. Exceptions to this generalization were *Anthriscus sylvestris*, *Cirsium arvense*, *Glechoma hederacea* and *Urtica dioica*, growing in N_xPK treatments. The high cover of *Achillea millefolium* in the control was consistent with the results from the Park Grass Experiment (Grime et al. 1988) and Laelatu wooded meadow experiment in Estonia (Sammul et al. 2003). Pennings et al. (2005) report a decrease in *A. millefolium* in 7 out of 12 fertilizer experiments and concluded that the response of this species to N application depended on site conditions.

The decrease in species richness in the N_{200}PK treatment accords with the results of other long-term experiments (Sammul et al. 2003, Crowley et al. 2005, Galka et al. 2005, Hejcman et al. 2007a). The absence of a negative effect of PK treatment on species richness was probably because the limiting nutrient to biomass production in tall grasses was N. However, the increase in moss cover with that of fertilizer application was opposite to trends reported by other long-term experiments (Virtanen et al. 2000, Niinemets & Kull 2005, Hejcman et al. 2007a). Further research is necessary to determine why the cover of mosses increased with increase in N application.

Biomass production data collected over 40 years in the Černíkovice experiment is probably the longest continuous yield series for alluvial grassland in Central Europe. In the initial phase of the experiment, the decrease in biomass production was ascribed to the effect of experiment; the collection of all aboveground biomass resulted in depletion of other nutrients which were not applied (Velich 1986). Under the initial farm conditions approximately 35% of the biomass was left in the meadow after harvest.

High inter-annual variation in biomass production occurred despite the relatively stable water table level maintained by the water-course in the vicinity of the experiment. In 1981, 1989 and 2002, marked decreases in biomass production followed spring or summer flooding. Although many species of alluvial meadows are well adapted to flooding (Koutecký & Prach 2005), they may produce less biomass when flooded. In addition, part of the variability in biomass production is attributable to the mean temperature during the growth period before the first or second harvest, but not to precipitation (Daňhelka & Honsová 2006). High inter-annual fluctuations in biomass production under Czech conditions accord with the results of Pavlů et al. (2006). In the Černíkovice experiment, the yield fluctuations occurred on highly productive alluvial meadow at all levels of nutrient availability. Long-term studies on grassland ecology indicate, that differences in biomass production of control and PK treatment plots may not occur before the 25th year of the experiment. This was probably a consequence of nutrient depletion in the control, which occurred despite of the occasional flooding at this locality.

In the non-fertilized control, the height of the sward indicated suitable growth conditions even without the application of additional nutrients. This contrasts with the results of the Rengen Grassland Experiment where potential sward height was higher than actual in the control, but lower in treatments with complete fertilizer (Hejcman et al. 2007a). It is proposed that the difference between both heights in non-fertilized grassland can be used as an indicator of sward response to fertilizer application: if actual height is higher than potential, a relatively weak response of the plant community to fertilizer application is indicated. To verify this conclusion, other long-term fertilizer experiments have to be evaluated.

The Černíkovice experiment shows the importance of long-term studies in grassland research for separating inter-annual fluctuations from real trends. Further, some of the effects of fertilizers can be recognized many years after their application.

Acknowledgements

The authors are deeply indebted to Prof. Jiří Velich, the initiator of the experiment. The authors also thank Jiří Mrkvička for continuing the experiment over the past two decades. Thanks go to Jan W. Jongepier for linguistic correction and two anonymous referees for their useful comments. Tony Dixon kindly edited the English of the final version of the manuscript. The research was financially supported by grant NAZV QF 4062. M. H. was supported by project MSMT 2B06012 and V. P. by MA 0002700601.

Souhrn

Černíkovický pokus s dlouhodobým hnojením travního porostu byl založen v roce 1966 na aluviální louce s dominancí *Alopecurus pratensis*. Pokus se skládal z následujících šesti variant hnojení: nehnojená kontrola, PK, N_{50}PK, N_{100}PK, N_{150}PK a N_{200}PK. Pokusná louka byla v první etapě sečena třikrát ročně a od konce 80 let pouze dvakrát ročně. V polovině května 2005 před první sečí byla vizuálně odhadnuta pokryvnost přítomných druhů rostlin, změřena výška porostu a stanovena produkce nadzemní biomasy za účelem zjištění změn způsobených dlouhodobou aplikací hnojiv.

Počítáno pomocí RDA, vliv hnojení na strukturu porostu byl průkazný a vysvětlil 32% variability vegetačních dat. Leguminózy nebyly zjištěny ve variantě N_{200}PK a naopak dosahovaly nejvyšší pokryvnosti v kontrole a PK variantě. Trávy měly, na rozdíl od ostatních dvouděložných, nejnižší pokryvnost v PK variantě a v kontrole, které se průkazně lišily od všech variant s aplikací N. Druh *Alopecurus pratensis* převládal ve všech variantách s N_xPK hnojením. Byliny měly nejvyšší pokryvnost v kontrole následované PK variantou a obě varianty se průkazně lišily od všech variant s aplikací N. Druh *Achillea millefolium* byl zaznamenán ve všech variantách, nejvyšší pokryvnost však byla zjištěna v kontrole. Počet druhů cévnatých rostlin se pohyboval v rozmezí od 8 na 1 m^2 v N_{200}PK variantě do 24 v kontrole. Dále byl zjištěn průkazný pokles počtu druhů se vzrůstající výškou porostu. Pokryvnost mechorostů se pohybovala v rozmezí od 1 do 6 % a podobně jako výška porostu stoupala s úrovní hnojení. Produkce nadzemní biomasy byla nejnižší v kontrole, která se průkazně lišila od ostatních variant.

Na základě výsledků Černíkovického pokusu a jejich porovnání s ostatními dlouhodobými pokusy s hnojením bylo konstatováno, že nízkoprodukční travní porosty se specifickým druhovým složením jsou ohroženy nevhodným hnojením mnohem více než porosty přirozeně vysoce produktivní.

References

Botta-Dukát Z., Chytrý M., Hájková P. & Havlová M. (2005): Vegetation of lowland wet meadows along a climatic continentality gradient in Central Europe. – Preslia 77: 89–111.

Correll O., Isselstein J. & Pavlů V. (2003): Studying spatial and temporal dynamics of sward structure at low stocking densities: the use of an extended rising-plate-meter method. – Grass Forage Sci. 58: 450–454.

Crowley M. J., Johnston A. E., Silvertown J., Dodd, M., Mazancourt C., Herd M. S., Henman D. F. & Edwards G. R. (2005): Determinants of species richness in the Park Grass Experiment. – Am. Nat. 165: 179–192.

Daňhelka J. & Honsová D. (2006): Climate impact on yields of permanent grassland. – In: Sborník příspěvků VI. ročníku mezinárodní vědecké konference Agroregion 2006 [Proceedings of the 6th Agroregion conference 2006], p. 20–23, Zemědělská fakulta Jihočeské univerzity, České Budějovice.

De Kroon & Bobbink R. (1997): Clonal plant dominance under elevated nitrogen deposition, with species reference to *Brachypodium pinnatum* in chalk grassland. – In: De Kroon H. & van Groenendael J. (eds.), The ecology and evolution of clonal plants, p. 359–379, Backhuys, Leiden.

Di-Tommaso A. & Aarsen L. W. (1989): Resource manipulations in natural vegetation: a review. – Vegetatio 84: 9–29.

Dodd M. E., Silvertown J., McConway K., Potts J. & Crawley M. J. (1994): Application of the British national vegetation classification to the park grass experiment through time. – Folia Geobot. Phytotax. 29: 321–334.

Gaisler J., Fiala J. & Spoustová (1998): The changes of botanical composition and yield in dependence on the type of grassland and fertilization. – Rostlinná výroba 44: 39–44.

Galka A., Zarzynski J. & Kopeć M. (2005): Effect of different fertilization regimes on species composition and habitat in long-term grassland experiment. – Grassland Sci. Eur. 10:132–135.

Grime J. P., Hodgson J. G. & Hunt R. (1988): Comparative plant ecology. – Unwin Hyman, London.

Güsewell S., Koerselman W. & Verhoeven J. T. A. (2002): Time-dependent effects of fertilization on plant biomass in floating fens. – J. Veg. Sci. 13: 705–718.

Hejcman M., Klaudisová M., Schellberg J. & Honsová D. (2007a): The Rengen Grassland Experiment: plant species composition after 64 years of fertiliser application. – Agric. Ecosyst. Environ. 122: 259–266.

Hejcman M., Klaudisová M., Štursa J., Pavlů V., Schellberg J., Hejcmanová P., Hakl J., Rauch O. & Vacek S. (2007b): Revisiting a 37 years abandoned fertilizer experiment on *Nardus* grassland in the Czech Republic. – Agric. Ecosyst. Environ. 108: 231–236.

Klapp E. (1938): Wiesen und Weiden. – Paul Parey, Berlin & Hamburg.

Klečka A. & Fabian J. (1935): Praktický význam pokusnictví v lukařství [Practical importance of experimentation in grassland management]. – Zemědělský archiv 26 (7–8): 1–16.

Kopeć M. & Gondek K. (2002): The effect of long-term fertilization on the sulphur content in soil and in the mountain meadow sward (Czarny Potok). – Rostlinná výroba 48: 525–530.

Koutecký P. & Prach K. (2005): Recovery of alluvial meadows after extreme summer flood: a case study. – Ecohyd. Hydrob. 5: 32–38.

Královec J. & Prach K. (1997): Changes in botanical composition of a former intensity managed sub-montane grassland. – In: Management of grassland biodiversity. Proceedings of the International occasional symposium of European Grassland Federation, p. 139–142, European Grassland Federation, Warszawa – Lomza.

Kubát K., Hrouda L., Chrtek J., Kaplan Z., Kirschner J. & Štěpánek J. (eds.) (2002): Klíč ke květeně České republiky [Key to the flora of the Czech Republic]. – Academia, Praha.

Larin I. V. (1956): Prirodnyje senokosy i pastbišča [Natural meadows and pastures]. – Moscow.

Mrkvička J. & Veselá M. (2002): The influence of long-term fertilization on species diversity and yield potential of permanent meadow stand. – Rostlinná výroba 48: 69–75.

Mrkvička J., Veselá M., Klimeš F. & Kocourková D. (2006): The changes of species richness and diversity of foxtail type stand during long-term fertilization. – Sci. Agric. Boh. 37: 41–48.

Mrkvička J. & Veselá M. (2004): Vliv výživy na floristickou skladbu a výnosy lučního porostu [Effect of fertilization on plant species composition and on forage yield of grassland]. – Úroda 4: 16–17.

Niczyporuk A. & Jankowska-Huflejt H. (2000): Formation of plant cover during 50 years of differing utilization and fertilization of permanent grassland. – Zeszyty naukowe – Scientific papers of the Agricultural University of Cracow 368: 241–247.

Niinemets U. & Kull K. (2005): Co-limitation of plant primary productivity by nitrogen and phosphorus in a species-rich wooded meadow on calcareous soils. – Acta Oecol. 28: 345–356.

Pavlů V., Hejcman M., Pavlů L. & Gaisler J. (2003): Effect of rotational and continuous grazing on vegetation of an upland grassland in the Jizerské Hory Mts., Czech Republic. – Folia Geobot. 38: 21–34.

Pavlů V., Hejcman M., Pavlů L., Gaisler J. & Nežerková P. (2006): Effect of continuous grazing on forage quality, quantity and animal performance. – Agric. Ecosyst. Environ. 113: 349–355.
Prach (1993): Vegetation changes in a wet meadow complex, South Bohemia, Czech Republic. – Folia Geobot. Phytotax. 28: 1–13.
Pennings S. C., Clark C. M., Cleland E. E., Collins S. L., Gough L., Gross K. L., Milchunas D. G. & Suding K. N. (2005): Do individual plant species show predictable responses to nitrogen addition across multiple experiments? – Oikos 110: 547–555.
Regal V. & Krajčovič V. (1963): Pícninářství [Forage production]. – Státní zemědělské nakladatelství, Praha.
Sammul M., Kull K. & Tamm A. (2003): Clonal growth in species-rich grassland: result of a 20-year fertilization experiment. – Folia Geobot. 38: 1–20.
Schellberg J., Möseler B.M., Kühbauch W. & Rademacher I. F. (1999): Long-term effects of fertilizer on soil nutrient concentration, yield, forage quality and floristic composition of a hay meadow in the Eifel Mountains, Germany. – Grass Forage Sci. 54: 195–207.
Silvertown J. (1980): The dynamics of a grassland ecosystem: botanical equilibrium in the Park Grass Experiment. – J. Appl. Ecol. 17: 491–504.
Silvertown J., Poulton P., Johnston E., Grant E., Heard M. & Biss P. M. (2006): The Park Grass Experiment 1856–2006: its contribution to ecology. – J. Ecol. 94: 801–814.
Spiegelberger T., Hegg O., Matthies D., Hedlund K. & Schaffner U. (2006): Long-term effects of short-term perturbation in a subalpine grassland. – Ecology 87: 1939–1944.
StatSoft (1995): Statistica for Windows. – StatSoft, Tulsa.
ter Braak C. J. F. & Šmilauer P. (2002): CANOCO Reference manual and CanoDraw for Windows user's guide: Software for Canonical Community Ordination (version 4.5). – Microcomputer Power, Ithaca.
Tilman D. (1982): Resource competition and community structure. – Princeton University Press, Princeton.
Trnka M., Eitzinger J., Gruszczynski G., Buchgraber K., Resch R. & Schaumberger A. (2006): A simple statistical model for predicting herbage production from permanent grassland. – Grass Forage Sci. 61: 253–271.
Velich J. (1986): Studium vývoje produkční schopnosti trvalých lučních porostů a drnového procesu při dlouhodobém hnojení a jeho optimalizace [Study of development of yield capacity of grasslands under long-term fertilization and optimization of fertilization]. – Vysoká škola zemědělská v Praze, Agronomická fakulta, Praha.
Virtanen R., Johnston A.E., Crawley M. J. & Edwards G. R. (2000): Bryophyte biomass and species richness on the Park Grass Experiment, Rothamsted, UK. – Plant Ecol. 151: 129–141.

Received 8 November 2006
Revision received 22 April 2007
Accepted 16 May 2007

Appendix 1. – Mean cover of vascular plant species and mosses (in %), mean species richness per 1 m^2 and dry matter of the aboveground biomass yield in 2006 ($t \cdot ha^{-1}$) recorded in the Černíkovice experiment. FG – functional group (L – legumes, R – rhizomatous grasses, TG – tall graminoides, SG – short graminoides, SH – short herbs, TH – tall herbs). Treatment abbreviations are given in the Table 1. ▶▶

Species	FG	Treatment					
		Control	PK	$N_{50}PK$	$N_{100}PK$	$N_{150}PK$	$N_{200}PK$
Aegopodium podagraria	TH	1.4	0.5	0.4	0.9	1.5	1.9
Agrostis stolonifera	SG, R	0.3	2.4				
Achillea millefolium	TH	15.4	9.5	2.9	4.3	0.8	0.8
Alchemilla spp.	SH	0.7	1.5		0.3		
Alopecurus pratensis	TG, R	7.0	8.5	38.3	28.6	43.6	37.9
Angelica sylvestris	TH	0.1					
Anthoxanthum odoratum	SG	0.5	0.9		0.1		
Anthriscus sylvestris	TH	0.2	1.0	0.7	1.9	0.4	2.4
Arrhenatherum elatius	TG	0.1					0.6
Betonica officinalis	TH	0.3					
Cardamine pratensis	SH	0.8		0.1		0.1	
Cerastium holosteoides	SH	1.6	2.2	1.3	1.3	1.2	1.3
Cirsium arvense	TH	1.0	0.9	1.0	1.6	1.8	2.4
Cirsium palustre	TH				0.5	0.2	
Cynosurus cristatus	SG	0.1					
Dactylis glomerata	TG	0.6	0.6	1.1	1.1	0.4	0.8
Deschampsia cespitosa	TG	0.1	2.5	6.8	0.3	1.8	0.4
Elytrigia repens	H	1.0	0.5	0.5	0.5	0.5	2.5
Equisetum palustre	SH	1.5	3.0	0.9	0.5	0.9	0.6
Festuca pratensis	TG	2.3	1.9	2.8	0.6	3.8	0.4
Festuca rubra agg.	TG, R	3.9	1.1	2.5	2.9	1.3	0.8
Galium album	TH	0.9	0.5	0.1	1.3		
Glechoma hederacea	SH	0.5	0.3	1.1	1.8	3.9	1.2
Heracleum sphondylium	TH	0.1			0.1		
Holcus lanatus	TG, R	2.1	3.2	5.4	1.5	2.9	4.7
Lathyrus pratensis	L	6.0	5.9	1.6	2.5	0.5	
Leucanthemum vulgare	TH		0.5				
Linaria vugaris	SH			0.5	0.1		
Luzula campestris	SG	1.1	0.5		0.2	0.1	
Lychnis flos-cuculi	TH	0.1	0.9				
Lysimachia nummularia	SH	5.9	4.4	1.4	1.6	1.0	0.5
Plantago lanceolata	SH	1.3	1.1		0.1		
Poa pratensis	TG, R	11.8	17.0	18.4	25.4	26.9	27.1
Poa trivialis	TG, R		0.4	1.8	1.8	0.5	0.5
Polygala amarella	SH				0.1		
Ranunculus acris	TH	1.1	0.4	0.8	0.3	0.3	
Ranunculus repens	TH	1.3	2.3	0.8	0.6	0.8	
Rosa sp.	TH	0.1					
Rumex acetosa	TH	0.1					
Taraxacum spp.	SH	0.5	0.4		0.4	0.3	0.3
Trifolium campestre	L	0.4	0.4				
Trifolium dubium	L		0.1				
Trifolium pratense	L		0.6				
Trifolium repens	L	0.5	0.8		0.3	0.1	
Trisetum flavescens	TG	1.0	1.9		0.1	0.9	0.4
Urtica dioica	TH		0.6			0.2	0.3
Veronica arvensis	SH	0.6	0.8	0.5	2.9	0.5	
Veronica chamaedrys	SH	5.1	7.6	5.9	2.9	2.8	1.9
Veronica serpyllifolia	SH	0.4	0.9	0.2	0.7		
Vicia sp.	L				0.1	0.1	
Vicia tetrasperma	L	0.8					
Mean species richness per 1 m^2		18	17	15	15	14	10
Cover of mosses		1.0	2.0	2.5	4.0	5.0	5.5
Aboveground biomass yield in 2006		4.4	7.0	7.7	8.0	8.2	8.3