Effect of fertilization on root growth in the wet submontane meadow

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ABSTRACT

Root net primary productivity (RNPP) was assessed in the wet submontane meadow in the highland region of the Czech Republic. RNPP was studied from 1992 to 1995 with help of the in-growth core technique. The effect of different intensities of fertilization on root growth was covered. In comparison with unfertilized stands, the application of fertilizers (90 kg N/ha) resulted, mostly significantly, in greater root productivity (297 g/m^2/year and 2.08 g/m^2/day, on average). This represented an increase by 72% and 71%, respectively. Variability in the fraction of RNPP to total net primary productivity was examined. Results indicate that this fraction varied from 0.18 to 0.25 across the compared treatments and decreased with increasing fertilization. Data show how different roles can grasslands play in accumulation of plant matter due to different levels of fertilization.

Keywords: biomass partitioning; grassland; effect of nutrients; net primary productivity; precipitation

Understanding of net primary productivity (NPP) is particularly important in grassland ecosystems. In most grasslands, a large proportion of biomass is below-ground (e.g. Rychnovská 1983, Hui and Jackson 2005). It is important to understand how different levels of fertilization affect soil carbon (C) pools/fluxes particularly, as these ecosystems store up to 30% of the world below-ground C (Risch et al. 2007). Grasslands therefore play a significant role in the global C cycle.

In grasslands, above-ground net primary production (ANPP) is positively correlated with mean annual precipitation (e.g. Kochy and Wilson 2004, Yahdjian and Sala 2006). Only a few studies related below-ground primary production (BNPP) with climate variables and, in addition, contradictory data on root growth and below-ground biomass were published (e.g. Ibrahim et al. 1997, Titlyanova et al. 1999, Gill et al. 2002, Hejduk and Hrabě 2003).

Summary of the observations of various authors together with data published by Fiala and Studený (1987) and Fiala (2010) show that increasing doses of fertilizers, above all nitrogen (N), decrease root relative to shoot growth. In addition, frequencies of cutting also decrease the weight of roots. Calculation of BNPP is mainly based on changes in total below-ground dry mass which is accompanied by many methodologist problems (Lauenroth et al. 2006). Another useful approach which enables to estimate root production is in-growth core technique (Milchunas 2009). Despite large data series gathered by this method (e.g. Tomaškin

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and Tomaškinová 2012), a large gap still exists in the knowledge of root increments in grasslands affected by different levels of fertilization.

The first aim of our study was to estimate and compare impacts of various fertilization and terms of cutting on changes in RNPP and in proportion of roots in total NPP (f_{RNPP} coefficient). In addition, we wanted to find differences in root increments in various parts of the year. We hypothesized that: (1) The effect of fertilization will be manifested in a greater amount of yearly root increments (RNPP), but proportion of roots (f_{RNPP}) in total NPP will be lower; (2) This effect will be more pronounced, due to the intensive root growth, in the period of favourable precipitation and temperature.

MATERIAL AND METHODS

Study site. The study was conducted in the meadow of the Moravian-Bohemian Highland during four years (1992–1995). This area is a part of Hercynicum (region build of granite and crystalic rocks with Ca-deficient soil). Brown acid gleyed soil occurred. Experimental stands were located near the village of Kameničky (49°43'N, 15°58'E, 530 m a.s.l.). The mean annual temperature and precipitation (for the period 1961–1990) in the region was 7°C and 762 mm (Kameničky, Svratouch). More detailed description of the area, plant community and its habitat are given by Hrabě et al. (2002). Data published by the Czech Institute of Hydrometeorology (1992–1995) for the region of the Moravian-Bohemian Highland indicates that, in comparison with 1993 and 1995, 1992 and 1994 growing seasons were drier and warmer (Figure 1). During growing seasons of both years the precipitation was about 46% and 27% below the long-term averages. These years were also substantially warmer (on the average by 2.2°C and 2.3°C, respectively). These characteristics were also reflected in Lang rain factors (sum of precipitation/mean temperature) calculated for all years. The factor attained 69, 90, 70 and 106 values in years 1992, 1993, 1994 and 1995, respectively.

Studied stands. The studied highland grassland was characterized as a species rich stand of semi-natural wet meadow. The species composition of the stand was characterized by herbs (*Sanguisorba officinalis*, *Ranunculus acer* and *Polygonum bistorta*) prevailed above grasses (primarily *Alopecurus pratensis* and *Festuca rubra*, Zelená V. unpubl.). Plant cover of herbs represented 49% of total stand, while grasses formed only 37% of the stand. Effects of mowing (three times per year) and NPK fertilization on root production at four nutrient levels (treatment 1 – unfertilized stand; treatment 2 – 30 kg/ha of P, 60 kg/ha of K; treatment 3 – 90 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K; treatment 4 – 180 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K) were examined (Hrabě et al. 2002). Five replications of each treatment were in Latin square design. Mean values of above-ground production ranged from 361 to 978 g/m^2 in unfertilized treatment and from 566 to 11 163 g/m in 180 kg N/ha + PK treatment in 1993 and 1994, respectively. Thus the proportion of roots represented about 80% of total below-ground dry mass and around 50% of total plant matter recorded in the autumn months. For these calculations we used ANPP data published by Hrabě et al. (2002) and below-ground plant values summarized by Holub et al. (2013).

Root analyses. In-growth core technique (Milchunas 2009) was used to assess RNPP in four years. Five plastic mesh tubes with river sand were inserted into holes (5 cm in diameter, 10 cm

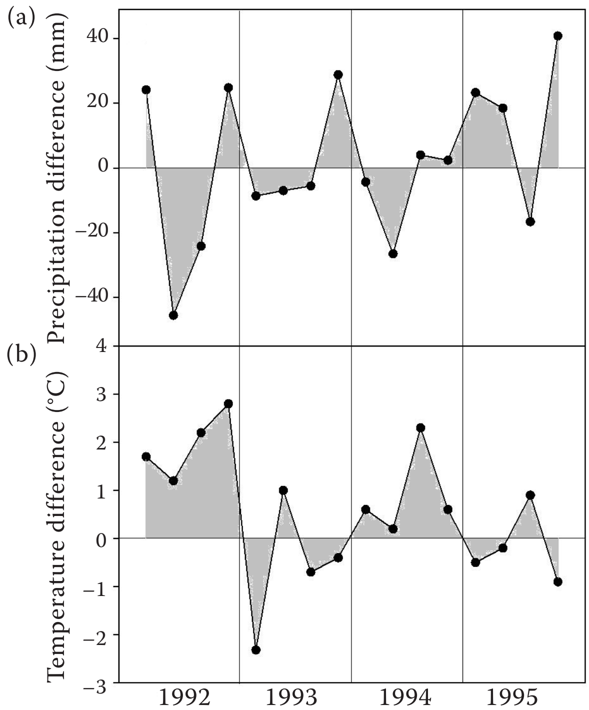

Figure 1. Differences in the precipitation (a) and air temperature (b) from the long-term values (1961–1990) recorded for the studied region of the Moravian-Bohemian Highland and calculated for quarters of the years 1992–1995

depth, $n = 5$) in experimental treatments at the beginning of the growing seasons. In the course of the growing seasons, cores were lifted usually before the first and second cuttings and in autumn months. After cutting, new soil cores were put in new holes. Root samples were washed and dried at 70°C to constant weight. Sum of produced root biomass in individual intervals represented only yearly root increments (not total below-ground plant parts). For comparison of the proportion of produced roots f_{RNPP} coefficient was used (Hui and Jackson 2005). The f_{RNPP} coefficient was calculated using the equation: f_{RNPP} = RNPP/(ANPP + RNPP), where RNPP was root net primary productivity and ANPP was above-ground net primary productivity. For these calculations we used ANPP data published by Hrabě et al. (2002).

Statistical analysis. Data were evaluated by an analysis of variance, using statistical package STATISTICA 7 (StatSoft, Tulsa, USA). Two-way ANOVA analysis was used to test the effects of fertilization and time period. Significant differences among means were tested (*LSD* test ($P < 0.05$) after ANOVA).

RESULTS AND DUSCUSSION

The ANOVA has shown that both the yearly and daily root increments were significantly affected by period of the year and grassland fertilization (Table 1). In unfertilized stands, the largest root increments (RNPP) attained 220 and 1.48 g/m^2 in yearly and daily periods of the 1992 and 1994 years, respectively (Table 2). However, 1.5 to 2.1 times higher values of RNPP and daily root increments reached in stands where the fertilization of 90 kg N/ha + PK was applied, although not significantly in all cases. A significant effect of period when roots penetrate into cores was recorded both in 1992 and 1993. Concerning interactions between fertilization and time period, yearly and daily root increments were significantly affected during three years studied (Table 1).

The effect of fertilization on roots showed that the application of 90 kg N/ha + PK resulted in a greater RNPP, i.e., 233–358 g/m^2 per yearly period and 1.58–2.75 g/m^2 per day, on average (Table 2). In comparison with unfertilized stands, it represents an increase of root production by 76% and 71% on average. However, application of N at the high rate of 180 kg/ha + PK caused rather a decrease in root increments (down to 216–275 g/m^2 and 1.33–1.94 g/m^2 in yearly periods and per day, on average). Increasing intensity of fertilization, and N doses particularly, generally resulted in the growth of above-ground plant parts rather than roots. Overview of literature data summarized by several authors (e.g. Gáborčík and Kohoutek 1999, Fiala 2010) indicates that contradictory data on root biomass affected by fertilization were often presented. Nevertheless, the results of present authors are supported by findings of several authors (e.g. Gáborčík 1985, Li and Redman 1992, Hrabě et al. 2002), who reported that total below-ground biomass decreased at the highest N level. Thus our results confirmed our both assumptions: fertilization supported root production, but root proportion was lowered.

Table 1. The effect of fertilization and time period on root increments in wet *Cirsium* meadow

	1992			1993		1994			1995	
	df	*F*	*P*	*F*	*P*	*df*	*F*	*P*	*F*	*P*
Yearly root increment (g/m^2/period)										
Fertilization (F)	3	4.4	**	2.7	ns	3	16.1	***	9.6	***
Period (P)	2	75.5	***	36.7	***	1	0.1	ns	2.9	ns
F × P	6	2.3	*	1.3	ns	3	3.7	*	4.1	*
Daily root increment (g/m^2/day)										
Fertilization	3	5.4	**	0.3	ns	3	8.8	***	9.6	***
Period	2	51.4	***	44.3	***	1	0.0	ns	2.9	ns
F × P	6	2.5	*	3.8	**	3	2.4	ns	4.1	*

Results of two-way ANOVA (*F*-test) analysis. ns – not significant; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$

Table 2. Mean values of root increments at different fertilization levels in wet *Cirsium* meadow in the course of four years (1992–1995)

	Fertilization			
	N0	N0-PK	N90-PK	N180-PK
Yearly root increments (g/m^2/period)				
1992 (29/4–4/11)	220^a (100%)	222^a (101%)	326^b (148%)	275^{ab} (125%)
1993 (10/5–22/9)	180^a (100%)	188^a (104%)	271^b (151%)	246^b (137%)
1994 (14/4–9/8)	172^a (100%)	173^a (101%)	358^b (208%)	225^a (131%)
1995 (21/4–3/10)	118^a (100%)	145^a (123%)	233^b (197%)	216^b (183%)
Daily root increments (g/m^2/day)				
1992 (29/4–4/11)	1.07^a (100%)	1.08^a (101%)	1.58^b (148%)	1.33^{ab} (124%)
1993 (10/5–22/9)	1.32^a (100%)	1.37^a (104%)	2.01^b (152%)	1.84^b (139%)
1994 (14/4–9/8)	1.48^a (100%)	1.49^a (101%)	2.75^b (186%)	1.94^a (131%)
1995 (21/4–3/10)	1.01^a (100%)	1.51^{ab} (150%)	1.99^b (197%)	1.84^b (182%)

Different letters denote significantly different values for four levels of fertilization separately for each year (*LSD* test ($P < 0.05$) after ANOVA). N0 – unfertilized; N0-PK – 30 kg/ha of P, 60 kg/ha of K; N90-PK – 90 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K; N180-PK – 180 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K

The fastest growth of below-ground organs can occur in different parts of growing season reflecting the annual cycle of vegetative activity (Titlyanova et al. 1999). In the present study, mostly significantly higher values of RNPP were assessed in the first half of the 1992 growing season through all treatments. At the beginning of the 1992 growing season, RNPP averaged 120, 155, 168 and 170 g/m^2 per period due to the first to fourth level of fertilization, respectively (Table 3). Intensive root growth, recorded from May until the middle of summer, was followed by lower amount of produced roots, usually during late summer months. In contrast, higher root growth in summer period and autumn was observed during the growing season 1993. Mostly significantly higher RNPP values were found in the last part of this growing season. This was documented by a larger amount of roots recorded in the autumn months in comparison with summer period and assessed nearly through all treatments (Table 3). No significant differences between two studied periods were found in the following years 1994 and 1995, except higher root growth in the treatment with the high N rate (180 kg/ha) during the second part of the growing season 1995. Thus, our findings confirmed our hypothesis and are in agreements with results of several authors that seasonal periodicity and higher percentage of active roots were usually characterized by two peaks. One at the beginning or in the middle of summer (often after flowering) and the second, lower, in autumn (e.g. Titlyanova et al. 1997, Fiala 2010).

Data on RNPP showed that the lowest amount of root production in all treatments can be associated with the lowest precipitation recorded for the region in 1992, but mostly not significant. The data on the increase in root growth due to the higher precipitation or irrigation were published recently (e.g. Li et al. 2011, Fiala et al. 2012). Our presented data were rather variable. Nevertheless, the highest value of daily root increment in different fertilization treatments was mostly recorded in 1994 or 1995 (Table 2). The pattern of biomass partitioning may be the result of plant species adaptation and responses to both internal and environmental stimuli (Hui and Jackson 2005). In the present study, application of the highest N rate (180 kg/ha) led to reduction of f_{RNPP} coefficient (0.18 in average) in comparison with other fertilization treatments (0.22–0.25 in averages). Data from 12 grassland types synthesized by Hui and Jackson (2005) showed that the fraction of BNPP to total NPP (f_{RNPP}) varied from 0.40 to 0.86 across sites. Thus the coefficient calculated for the studied highland meadow was lower. Although not statistically significant, the lowest values of the coefficient were found in the meadows where the highest doses of fertilizers were applied. Similarly, BNPP

Table 3. Mean values of root increments in various periods of year and under different levels of fertilization in wet *Cirsium* meadow in the different period of the four growing seasons (1992–1995)

	Year	Period	N0	N0-PK	N90-PK	N180-PK
Root increment (g/m^2/period)	1992	Apr 29–June18	120^{c}	155^{b}	168^{c}	170^{b}
		June 18–Aug 10	22^{a}	25^{a}	45^{a}	51^{a}
		Aug 10–Oct 23	78^{b}	42^{a}	113^{b}	53^{a}
	1993	May 10–June 16	19^{a}	30^{a}	27^{a}	21^{a}
		June 16–Aug 27	84^{b}	104^{b}	143^{b}	145^{c}
		Aug 27–Sept 22	77^{b}	53^{a}	101^{b}	83^{b}
	1994	Apr 14–June 13	97^{a}	66^{a}	162^{a}	135^{a}
		June 13–Aug 9	75^{a}	107^{a}	196^{a}	90^{a}
	1995	Apr 21–June 8	60^{a}	80^{a}	111^{a}	74^{a}
		June 8–Oct 3	58^{a}	65^{a}	122^{a}	141^{b}
Daily root increment (g/m^2/day)	1992	Apr 29–June18	1.52^{b}	1.96^{b}	2.13^{c}	2.16^{b}
		June 18–Aug 10	0.42^{a}	0.48^{a}	0.85^{a}	0.97^{a}
		Aug 10–Oct 23	1.05^{b}	0.56^{a}	1.54^{b}	0.72^{a}
	1993	May 10–June 16	0.53^{a}	0.81^{a}	0.74^{a}	0.56^{a}
		June 16–Aug 27	1.16^{a}	1.45^{ab}	1.99^{b}	1.98^{b}
		Aug 27–Sept 22	2.96^{b}	2.04^{b}	2.01^{b}	1.84^{b}
	1994	Apr 14–June 13	1.61^{a}	1.11^{a}	2.70^{a}	2.25^{a}
		June 13–Aug 9	1.35^{a}	1.91^{a}	2.80^{a}	1.61^{a}
	1995	Apr 21–June 8	1.24^{a}	1.67^{a}	2.30^{a}	1.55^{a}
		June 8–Oct 3	1.21^{a}	1.35^{a}	2.55^{a}	2.94^{b}

Different letters denote significantly different values among time periods within each year and fertilization treatment (*LSD* test ($P < 0.05$) after ANOVA). N0 – unfertilized; N0-PK – 30 kg/ha of P, 60 kg/ha of K; N90-PK – 90 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K; N180-PK – 180 kg/ha of N, 30 kg/ha of P, 60 kg/ha of K

in grasslands of the Inner Mongolia tended to decrease with N addition (Li et al. 2011). However, the fraction of below-ground to total biomass (f_{RNPP}) decreased with the addition of supplemental N. It is necessary to keep in mind that the effect of cutting also exists (Xu et al. 2012). Their data showed that clipping increased f_{RNPP} from 0.58 to 0.64 in tallgrass prairie.

Our new series of data indicate that fertilization of wet submontane meadows resulted in a greater RNPP. But the highest doses of fertilization applied (180 kg N/ha + PK) shown an opposite trend, i.e., a reduction of root growth parameters. This effect was more pronounced in the proportion of roots (f_{RNPP}) in total NPP due to the most intensive fertilization. Estimated values of RNPP affected by fertilization represent valuable data, which were not still assessed for wet and nutrient poor *Cirsium* meadows.

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