

THE EFFECT OF UTILIZATION TERM ON THE BIOMASS PRODUCTION, ORGANIC MATTER DIGESTIBILITY AND ERGOSTEROL CONTENT OF SEMI-NATURAL GRASS STAND IN THE AUTUMN AND IN WINTER

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Abstract

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The biomass of dry matter (DM) and forage quality of a grass pasture in the Bohemian-Moravian Highlands (Czech Republic), situated at an altitude of 553 m a.s.l., was measured in late autumn and in winter in the period from 2000 to 2003. The semi-natural grass pasture was dominated by *Festuca rubra*, *Taraxacum officinale*, *Elytrigia repens*, *Dactylis glomerata*, *Trisetum flavescens*, *Poa* spp., *Agrostis tenuis* and *Phleum pratense*. Biomass production and forage quality were measured in November, December and January after one preparatory cut in June or two preparatory cuts in June and July, and in June and August. Biomass of DM decreased from November to January and ranged, depending on the year and the number of preparatory cuts, from 0.37–3.13 t ha⁻¹ in November to 0.15–1.36 t ha⁻¹ in January. The biomass of DM decreased the later the preparatory cut. Organic matter digestibility decreased from November to January, ranging from 0.448–0.606 in November to 0.352–0.578 in January. A delayed preparatory cut resulted in an increased digestibility. Ergosterol concentration increased with the progressing autumn and winter, ranging from 40–111 mg kg⁻¹ DM in November to 110–265 mg kg⁻¹ DM in January. Lower ergosterol concentrations were observed after a late preparatory cut. The results were statistically analysed by ANOVA and Tukey HSD test. The length of the main use was observed to have a significant effect on biomass of DM, digestibility of organic matter and ergosterol concentration ($P < 0.05$) in all three years of the investigation.

winter grazing, biomass, organic matter digestibility, ergosterol

The all-year-round outdoor raising of beef cattle and dry dairy cows represents an extensive form of land use and is of great importance particularly for peripheral areas (Opitz von Boberfeld, 2001). Livestock raised at pasture all-year round have green forage at their disposal in addition to preserved forage (Stählin and Tirtapradja, 1974). An extension of the grazing season into the winter can reduce the cost of feeding of dry dairy cows (D'Souza *et al.*, 1990; Adams

et al., 1994). In order that the pasture can be used also in winter, it is necessary either to perform the last mowing or grazing cycle in June or July or shift the last summer utilization period to the first week of August (Opitz von Boberfeld, 1997). The last summer utilization period influences winter forage yield and quality (Collins and Balasko, 1981; Gerrish *et al.*, 1994). An early summer harvest results usually in a lower forage quality in the autumn and winter (Opitz

von Boberfeld, 1997). Forage should not be infected by fungi in the autumn and winter because fungal attack can result in increased mycotoxin levels and consequently in health disorders in livestock (Opitz von Boberfeld, 1997). Ergosterol concentration can serve as an indicator of fungal attacks since it is a component of the fungal cell wall and hardly ever is present in tissues of higher plants (Seitz *et al.*, 1977; Martin *et al.*, 1990).

MATERIAL AND METHODS

Description of the site

The experiment was conducted in the Bohemian-Moravian Highlands (Czech Republic) at an altitude of 553 m. The average annual air temperature was 6.1 °C and the average annual total precipitation was 736 mm in years 1951–2000. The average annual air temperatures were 8.2 °C, 6.8 °C and 7.8 °C in years 2000, 2001 and 2003 respectively. Average annual total precipitation were 701 mm, 728 mm and 683 mm in years 2000, 2001 and 2002 respectively. The soil temperature dropped below 4 °C from December to March in 2000/2001, from November to March in 2001/2002 and from December to March in 2002/2003. Soil profile: A_p – E_N – B_M – B/C – C. The soil class is Loam over Silt Loam and the soil unit is Dystric Planosol. The species composition of the grass pasture under study in the first year of observation (expressed as proportions of the dominant species in the dry mat-

ter) was *Festuca rubra* (0.214), *Taraxacum officinale* (0.190), *Elytrigia repens* (0.148), *Dactylis glomerata* (0.124), *Trisetum flavescens* (0.09), *Poa spp.* (0.072), *Agrostis tenuis* (0.069) and *Phleum pratense* (0.064). All experimental plot between the last preparatory cut and the period of main use involved the application of 50 kg ha⁻¹ N. The plots were not grazed during this period.

Experimental design

A 3 × 3 split plot design was used with plots of 2 m × 10 m, with three replications. Individual plots were used in the same manner for a period of three years. Measure were accumulative effects.

The main effects evaluated and factor degrees are listed in Tab. I. If the entire pasture was covered with snow in a given month, forage was harvested immediately after the snow melting. Dates of harvest of the plots are listed in Tab. II.

I: Experimental factors evaluated and factor degrees

Dates of preparatory cuts (summer utilization)	June June + July June + August
Dates of winter cuts (winter utilization)	November December January

II: Harvest dates in the three years of the experiment

Year of observation	Summer utilization			Winter utilization		
2000/2001	8 June	11 July	8 August	1 Nov.	1 Dec.	15 March
2001/2002	5 June	11 July	6 August	5 Nov.	30 Jan.	19 Feb.
2002/2003	3 June	9 July	6 August	4 Nov.	2 Dec.	8 Jan.

Measurements

Plots were harvested with a finger cut machine MF 120 with the working width of 1.2 m and the harvested area was 12 m². The stubble height was 7 cm. All harvested biomass was weighed. Also, two samples (1 kg each) were taken away immediately after the harvest. These samples were dried at 103 °C and 60 °C, respectively. The stand up biomass of DM was determined from the sample dried at 103 °C. The sample dried at 60 °C was ground to particles of 1 mm size. This sample was used to estimate the *in vitro* organic matter digestibility (OMD). *In vitro* OMD was determined according to Hohenheimer Futterwerttest at

the Justus Liebig University. The analysed sample was mixed in a flask with the rumen liquid and incubated at 39 °C overnight. The amount of gasses produced was recorded for the period of 24 to estimate OMD (Menke and Steingass, 1987). Ergosterol concentration was analysed by HPLC. Measurements of ergosterol in an UV detector by means of HPLC were performed after the saponification of samples with ethanol and KOH and the subsequent extraction in petrolether. In this way it was possible to estimate total (i.e. both free and bond) concentration of ergosterol (Schwandorf and Müller, 1989).

Statistical analysis

The results were statistically analysed by ANOVA and Tukey HSD test. The data was analysed by means of the following statistical model:

$$x_{ijk} = \mu + \alpha_i + \beta_j + \gamma_k + \varepsilon_{ijk}$$

where, x = individual observation; μ = overall mean; α = effect of the harvest date; β = effect of the preparatory cut, γ = interaction between harvest date and preparatory cut, ε = residual error.

RESULTS

Biomass of DM in all three years was significantly ($P < 0.001$) influenced by the date of the preparatory cut (Tab. III). In November, biomass of DM with the preparatory cut in June ranged from 1.26–3.13 t ha⁻¹ in the three years (Tab. IV). In the case that the preparatory cut in June was followed by the preparatory cut in July, biomass of DM in November in the three years ranged from 0.73–2.07 t ha⁻¹. In the case that the second preparatory cut was delayed until August, biomass of DM in November ranged from 0.37–1.61 t ha⁻¹. There was a significant difference in biomass of DM ($P < 0.05$) in all three years between the preparatory cut in June and the preparatory cut in August, and in years 2001/2002 and 2002/2003 also between the preparatory cut in June and the preparatory cut in July (Tab. IV). In the three experimental years, there were no statistically significant differences between the preparatory cuts performed in July and August. The highest biomass of DM in December was with the preparatory cut performed in June (viz 0.68 t ha⁻¹–2.30 t ha⁻¹ depending on the year) and the lowest biomass of DM was recorded with the date of the last preparatory cut in August (0.19–0.65 t ha⁻¹). Differences between the harvest dates in Year 2 and Year 3 ($P < 0.05$) were found. Significant differences ($P < 0.05$) were found out between biomass of DM in November and December and November and January, respectively. There was no significant difference between biomass of DM production reached in December and January.

Sward qualities expressed as *in vitro* OMD (Tab. V) and by ergosterol concentration (Tab. VI) were not affected by the date of the preparatory cut during the three years as markedly as for biomass of DM. In Year 1 and Year 2, the *in vitro* OMD values were affected by the date of the preparatory cut ($P < 0.01$). In Year 3, the date of the preparatory cut was not observed to have any significant effect on *in vitro* OMD values. Significant differences in *in vitro* OMD ($P < 0.05$) were found out between preparatory cuts carried out in June and July (years 2000/2001) and in June

and August (years 2000/2001 and years 2001/2002). A statistically significant difference in *in vitro* OMD did not exist between the preparatory cut in June and the preparatory cut in August (Tab. V). The date of harvest exhibited a significant effect on *in vitro* OMD values in all three years ($P < 0.05$). Values of *in vitro* OMD were higher in November than in December and in January. When the date of the last preparatory cut was August, *in vitro* OMD values ranged in November from 0.524–0.606 in the three years. After the preparatory cut performed in July, *in vitro* OMD values ranged from 0.486 to 0.586 in the three years. The date of the preparatory cut in June resulted in *in vitro* OMD values decreasing to 0.448–0.569 in the three years. Values of *in vitro* OMD in December and January were lower than in November. The highest December values of *in vitro* OMD were recorded in 2002/2003; depending on the preparatory cut, they ranged from 0.516 to 0.558. In 2001/2002, they ranged (also depending on the preparatory cut) from 0.359 to 0.365. In January, the highest values of *in vitro* OMD were recorded in 2002/2003 (0.526 to 0.579) while the lowest ones were in 2001/2002 (0.352 to 0.358). There were significant differences ($P < 0.05$) between November and December as well as between November and January in all three years while between December and January this difference was significant only in the first experimental year. The ergosterol concentration in forage increased from November to January. The effect of harvest date on the concentration of ergosterol was significant ($P < 0.001$) in years 2001/2002 and years 2002/2003. A significant effect of the preparatory cut on the concentration of ergosterol was observed in the second and in the third experimental year too. When the harvest date in November was preceded by the preparatory cut in June, ergosterol concentration ranged from 71 mg kg⁻¹ DM to 111 mg kg⁻¹ DM in the three years. When the date of the preparatory cut was in August, ergosterol concentration ranged from 40–77 mg kg⁻¹ DM in the three years. The concentration of ergosterol increased with the ongoing winter. In December, it ranged from 68 to 169 mg kg⁻¹ DM and in January from 79 to 277 mg kg⁻¹ DM in the three years. The highest values were found out in December 2001 and in January 2002. The differences in ergosterol concentrations found between November and December were significant ($P < 0.05$) in 2001/2002 and 2002/2003. Differences between November and January were observed in 2001/2002 and 2002/2003 too. A statistically significant difference in ergosterol concentration was recorded also between the preparatory cut in July and the preparatory cut in August (Tab. VI). Ergosterol concentration is increasing with the lower digestibility of organic matter, with the correlation coefficient ranging from -0.47 to -0.87.

III: Sources of variation for biomass of DM, in vitro DOM and ergosterol during the three examination years

Source of variance	Degrees of freedom	2000/2001 Mean square F-test	2001/2002 Mean square F-test	2002/2003 Mean square F-test
Biomass of DM				
Block	2	0.77**	0.14	0.02
Harvest date (H)	2	0.19	1.50*	4.13*
Error of plots	4	0.04	0.17	0.34
Preparatory cut (P)	2	0.45***	3.10***	4.01***
P × H	4	0.04	0.54***	0.28**
Error of sub-plots	12	0.02	0.03	0.06
Total	26			
In vitro DOM				
Block	2	0.03	0.03	0.039
Harvest date (H)	2	4.85***	4.32**	0.75*
Error of plots	4	0.02	0.14	0.08
Preparatory cut (P)	2	0.19**	0.28**	0.24
P × H	4	0.13*	0.27***	0.13
Error of sub-plots	12	0.02	0.25	0.06
Total	26			
Ergosterol				
Block	2	7369.44	65515	219.62
Harvest date (H)	2	24754.78	60887.26***	23994.23**
Error of plots	4	5260.39	592.09	590.21
Preparatory cut (P)	2	32.33	12516.59***	9823.42***
P × H	4	500.44	4500.70***	4155.80**
Error of sub-plots	12	460.69	429.00	557.04
Total	26			

IV: Biomass of DM ($t\ ha^{-1}$) in the autumn/winter as influenced by the date of the preparatory cut and the harvest date in the years 2000–2003

Harvest date	Preparatory cut						
	June		June + July		June + August		
	Mean	s.e. of mean	Mean	s.e. of mean	Mean	s.e. of mean	
2000/2001							
November	1.26	0.27	0.89	0.16	0.72	0.17	0.96 ^a
December	0.87	0.22	0.58	0.08	0.54	0.28	0.66 ^a
January	0.98	0.19	0.96	0.14	0.52	0.11	0.82 ^a
	1.04 ^a		0.81 ^{ab}		0.59 ^b		
2001/2002							
November	2.35	0.38	0.73	0.10	0.37	0.03	1.15 ^a
December	0.68	0.18	0.32	0.09	0.19	0.03	0.40 ^b
January	1.06	0.15	0.36	0.01	0.15	0.01	0.52 ^b
	1.36 ^a		0.47 ^b		0.24 ^b		

Harvest date	Preparatory cut						
	June		June + July		June + August		
	Mean	s.e. of mean	Mean	s.e. of mean	Mean	s.e. of mean	
2002/2003							
November	3.13	0.18	2.07	0.20	1.61	0.27	2.27 ^a
December	2.30	0.14	0.89	0.16	0.65	0.08	1.28 ^b
January	1.36	0.32	0.82	0.13	0.75	0.20	0.98 ^b
	2.26 ^a		1.26 ^b		1.00 ^b		

V: OMD values in the autumn/winter as influenced by the date of the preparatory cut and the harvest date in years 2000–2003

Harvest date	Preparatory cut						
	June		June + July		June + August		
	Mean	s.e. of mean	Mean	s.e. of mean	Mean	s.e. of mean	
2000/2001							
November	0.530	0.01	0.539	0.008	0.541	0.011	0.537 ^a
December	0.447	0.023	0.445	0.022	0.415	0.01	0.436 ^b
January	0.460	0.005	0.368	0.003	0.384	0.009	0.404 ^c
	0.479 ^a		0.451 ^b		0.447 ^b		
2001/2002							
November	0.448	0.012	0.486	0.008	0.524	0.009	0.486 ^a
December	0.359	0.006	0.365	0.011	0.362	0.005	0.362 ^b
January	0.352	0.002	0.358	0.01	0.358	0.013	0.356 ^b
	0.386 ^a		0.403 ^{ab}		0.415 ^b		
2002/2003							
November	0.569	0.017	0.586	0.007	0.606	0.019	0.587 ^a
December	0.516	0.010	0.558	0.015	0.517	0.025	0.530 ^b
January	0.526	0.011	0.543	0.013	0.579	0.009	0.549 ^b
	0.537 ^a		0.562 ^a		0.567 ^a		

VI: Ergosterol concentration (mg kg^{-1} dry weight) in the autumn/winter as influenced by the date of the preparatory cut and the harvest date in years 2000–2003

Harvest date	Preparatory cut						
	June		June + July		June + August		
	Mean	s.e. of mean	Mean	s.e. of mean	Mean	s.e. of mean	
2000/2001							
November	93	9.71	86	28.62	77	23.84	85 ^a
December	79	32.84	68	15.17	90	24.83	79 ^a
January	159	16.46	185	35.47	175	44.14	173 ^a
	110 ^a		113 ^a		114 ^a		
2001/2002							
November	111	7.84	95	10.53	40	8.39	82 ^a
December	277	8.50	169	12.12	231	8.65	226 ^b
January	265	26.08	241	13.86	163	7.88	223 ^b
	218 ^a		168 ^b		145 ^b		

Harvest date	Preparatory cut						
	June		June + July		June + August		
	Mean	s.e. of mean	Mean	s.e. of mean	Mean	s.e. of mean	
2002/2003							
November	71	9.44	52	14.74	42	8.86	55 ^a
December	196	16.13	121	11.45	76	8.89	131 ^b
January	149	14.21	202	22.55	110	3.89	154 ^b
	139 ^a		125 ^a		76 ^b		

DISCUSSION

In the Czech Republic (Central Europe), the possibility of reducing feeding costs by means of a prolongation of the grazing season is in a very early stage of investigation. There are no research studies concerning the quality and production capacity of grass stands in the autumn and winter. Many authors have studied the quality and yield of herbage in the autumn and winter; they also investigated the possibilities of a prolongation of the grazing season but mostly under conditions of Atlantic climate and overseas (Baker *et al.*, 1961; Frame, 1970; Opitz von Boberfeld and Wolf, 2002). The territory of the Czech Republic is influenced by Atlantic climate only during the summer months while in winter the influx of cold air from Siberia predominates. When comparing our results with data published by other authors (Nerušíl and Kohoutek, 2002), it was found out that the production capacity decreased as the winter progressed. Archer and Decker (1977) published similar results. This decrease is caused by the ageing of stands, which is associated with the dying-off and decay of plant biomass. As far as the digestibility was concerned, Baker *et al.* (1965) reported an OMD value 0.710 in November in grassland stand in England with *Festuca arundinacea* dominating. This stand had been studied since the second half of August. Wolf and Opitz von Boberfeld (2003) reported that OMD values by a *Festuca arundinacea* stand in Hessen (Germany) from June to December ranged from 0.555 to 0.605 during a period of three years. In our three-year experiment comparable results were obtained only in winter 2002/2003. On the other hand, markedly lower OMD values were recorded in 2001/2002. Lower biomass of DM and decreased OMD values recorded in winter 2001/2002 could be explained also on the basis of reduced growth capacity of stands at low temperatures. As compared with the winter 2002/2003, average air and soil temperatures in November 2001 were 0.6 °C and 3.4 °C, respectively, while in 2002 the corresponding values were 4.3 °C and 5.1 °C. In November 2002, climatic conditions enabled a greater growth of grasses. *Dactylis glomerata* and/or *Poa pratensis* can grow

also at low temperatures: *Dactylis glomerata* begins to grow at temperatures of 4.4 °C (Leasure, 1952). The process of dying-off could be delayed and compensated by the growth of new shoots.

The concentration of ergosterol increased with the winter. There were also fluctuations in individual years. In winter 2000/2001, the sum of precipitation was lower and the temperatures were higher; the occurrence of mulds and, thus, the concentration of ergosterol were lower than in the subsequent two years especially in December. On the other hand, a wetter and colder autumn of 2001/2002 was reflected in an increased concentration of ergosterol. Concentration of ergosterol decreased levels after delayed preparatory cuts were mentioned by Opitz von Boberfeld (1996). Opitz von Boberfeld and Wolf (2002) found by *Festuca arundinacea* lower concentration of ergosterol in December (48–129 mg.kg⁻¹ DM) than in February (157–313 mg.kg⁻¹ DM). They monitored increasing concentration of ergosterol as the winter progressed.

These results indicate the importance of winter grazing not only in countries influenced by Atlantic maritime climate but also under conditions of Central Europe, which is influenced by influx of cold Siberian air.

CONCLUSION

Biomass of DM and organic matter digestibility decreased as the winter proceeded due to biomass ageing. Production and quality of forage used in the autumn and in winter is also influenced by the effect of the date of the preparatory cut in the summer period. Delaying the date of the preparatory cut to July or August reduces the biomass of dry matter but increases the digestibility of the herbage. Ergosterol concentration, as an indicator of sward infestation by fungi increased from November to January. Climatic conditions of Central Europe show a greater fluctuation of temperatures than the Atlantic maritime climate. This is reflected the annual fluctuations in grassland herbage quality and production. It follows from the research results that, with respect to

sward quality and production, the grazing period can be extended under climatic conditions of the Czech Republic until November and under more favourable weather conditions even until the beginning of

December. It appears to be optimal to use the sward in the summer period in July at the latest.

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SOUHRN

Vliv termínu využití na produkci biomasy, stravitelnost organické hmoty a obsah ergosterolu u polopřirozeného travního porostu na podzim a v zimě

Na Českomoravské vrchovině v nadmořské výšce 553 m byla v letech 2000–2003 hodnocena produkce a kvalita travního porostu využívaného pozdě na podzim a v zimě (listopad, prosinec a leden). U polopřirozeného travního porostu s dominancí *Festuca rubra*, *Taraxacum officinale*, *Elytrigia repens*, *Dactylis glomerata*, *Trisetum flavescens*, *Poa* ssp., *Agrostis tenuis* a *Phleum pratense* byly zjišťovány produkce nadzemní biomasy a stanovena stravitelnost organické hmoty (SOH) a obsah ergosterolu. Hlavnímu využití předcházela buď jedna přípravná seč v červnu nebo dvě přípravné seče v červnu a červenci, případně červnu a srpnu. Výnosy sušiny se snižovaly od listopadu do ledna. V závislosti na sledovaném roce a termínu přípravné seče byly v listopadu výnosy sušiny od 0,37 do 3,13 t.ha⁻¹ a v lednu od 0,15 t.ha⁻¹ do 1,36 t.ha⁻¹. Od listopadu do ledna klesala také stravitelnost organické hmoty. V listopadu byla od 44,8 % do 60,6 % a v lednu od 35,2 % do 57,8 %. Opožděný termín přípravné seče v létě se odrazil ve zvýšení stravitelnosti a snížení produkce nadzemní biomasy. Obsah ergosterolu v průběhu podzimu a zimy stoupal. V listopadu byl od 40 mg.kg⁻¹ sušiny do 111 mg.kg⁻¹ sušiny a v lednu od 110 mg.kg⁻¹ sušiny do 265 mg.kg⁻¹ sušiny. Nižší obsah ergosterolu byl při pozdějším termínu přípravné seče. Statisticky průkazný vliv ($P < 0,05$) na výnosy a kvalitu píce měl nejen termín hlavního využití (listopad, prosinec nebo leden), tak také poslední termín přípravné seče v létě (červen, červenec nebo srpen).

zimní pastva, biomasa, stravitelnost organické hmoty, ergosterol

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