



## Evolution of Total Protein Content and the Ratio of Sugar-Protein in Dry Matter of *Dactylis glomerata* and *Festuca pratensis* Variations Depending on the Type of Soil

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### Authors' contributions

This work was carried out in collaboration between all authors. Author JS designed the study, wrote the protocol and wrote the first draft of the manuscript. Author KJ reviewed the experimental design and all drafts of the manuscript. Authors JS and PD managed the analyses of the study. Authors JS and KJ performed the statistical analysis. All authors read and approved the final manuscript.

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### ABSTRACT

The study was conducted to determine the effect of soil type on total protein content and the formation ratio of sugars to proteins in dry matter varieties of cocksfoot and tall fescue. The experiment was arranged and conducted according to Research Center for Cultivar Testing guidelines. The experimental plots were sown with varieties of *Dactylis glomerata*: Niva, Tukan, Amila, Crown Royale and with varieties of *Festuca pratensis*: Limosa, Pasja, Anturka, Amelka. The plots were randomly selected, 1.5 meters wide and 6.67 m long, with an area of 10 m<sup>2</sup>, grouped in blocks with four replications. They were separated by 1 meter pathways between blocks and with 0.5 meter pathways between sub-blocks. The pathways lay fallow. The experiment in Krzyżewo

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(organic soil) was set up on ploughed soil, with spring barley as the forecrop. In Uhnin (mineral soil) the experimental plots were located on peat meadow. The airy dry matter was shredded and ground. The obtained material was subjected to chemical analysis to determine dry matter (by determining moisture content), protein compounds and simple sugars. The method of determination was near-infrared spectroscopy (NIRS) using a NIRFlex N-500 spectrometer and readytouse INGOT calibration applications. Regardless of variety, cut and years of research, higher total protein content occurred in dry matter of cocksfoot grown on mineral soil ( $165 \text{ g}\cdot\text{kg}^{-1}$  DM) than organic ( $172 \text{ g}\cdot\text{kg}^{-1}$  DM). Higher values of sugar-protein ratios characterized biomass of varieties of meadow fescue (0.50) than cocksfoot (0.85). Regardless of the examined grass species, better nutritional value had the plant varieties located on mineral soils than organic.

*Keywords: Protein; sugar; grass; variety; soil type.*

## 1. INTRODUCTION

The value of feed obtained from grasslands largely is determined by total protein content, sugars, crude fiber and minerals as well as differently [1-3]. The chemical composition of grasses, which are the main components of meadow-pasture is greatly differentiated and largely depends on the species and variety [4,5]. The nutritional value of different varieties of grass species changes with they grow and develop - it depends significantly on phenological phases in which they are mowed or grazed [6,7]. On the value of grass species and varieties has also influence their ability to develop of generative shoots, which contain less protein and more fiber than leafy shoots of vegetative [8]. According to Frankow-Lindberg and Olsson [9], essential for the production of fodder on arable land has a first regrowth, because it is about 50% of annual yield. For feed quality it is very important an appropriate time of grasses harvest, which should be made no later than the phase of full heading stage of the plant. In spring the plants grow and develop very intensively, so they fast changes its structure. Delay mowing causes a decrease in protein content, while increasing the content of structural carbohydrates that affect forage quality deterioration and a decline its digestibility [10]. The best forage quality is obtained under frequent four-, and in favorable weather conditions and with abundant fertilization even fifty harvesting. For such use are very tolerate ryegrass, festulolium, fescue and orchard grass, but with less frequently we can cut timothy, due to the slow initial growth in and bromegrass, due to the small regrowth after cutting. In modern systems of animal nutrition a very important role fulfill silages [11]. For a good silage it is important to the dry matter content and the ratio of sugar to protein. With high protein content and low sugar one this process is

difficult and requires the use of silage additives [3].

The aim of the study was the analyze of protein content and the formation of sugars to proteins ratio in dry matter of cocksfoot and tall fescue varieties grown on mineral and organic soils.

## 2. MATERIALS AND METHODS

This paper has drawn on two field experiments set up and carried out between 2010 and 2013 by the Research Centre for Cultivar Testing in Słupia Wielka. The experiment was conducted in two experimental stations: one in the Research Centre for Cultivar Testing in Krzyżewo and the other in the Experimental Stations for Variety Testing in Uhnin, being a branch of the Research Centre for Cultivar Testing in Cicibor Duży. The stations are located in the Podlaskie Woivodeship, the Wysokie Mazowieckie county, in the commune of Sokoły. Uhnin is located in the Lublin Vovodeship, the Parczew county and the Dębowa Kłoda commune.

The experiment was arranged and conducted according to Research Center for Cultivar Testing guidelines. The experimental plots were sown with varieties of *Dactylis glomerata*: Niva, Tukan, Amila, Crown Royale and with varieties of *Festuca pratensis*: Limosa, Pasja, Anturka, Amelka (d. AND 1009).

The plots were randomly selected, 1.5 meters wide and 6.67 m long, with an area of  $10 \text{ m}^2$ , grouped in blocks with four replications. They were separated by 1 meter pathways between blocks and with 0.5 meter pathways between sub-blocks. The pathways lay fallow. The experiment in Krzyżewo was set up on ploughed soil, with spring barley as the forecrop. In Uhnin the experimental plots were located on peat

meadow. Tables 1 and 2 present soil characteristics and mineral fertilizers used.

In the research the amount of seeds of the grass sown varied depending on the variety and the location of the experiment. It was as follows (in kg·ha<sup>-1</sup>):

- *Dactylis glomerata* – Tukan: 16.3; Amila: 17.5; CR: 18.8 (Krzyżewo) and 17.6 (Uhnin); Niva: 18.3,
- *Festuca pratensis* – Pasja: 28.7; Limosa: 29.8 (Krzyżewo) and 27.1 (Uhnin); Anturka: 26.6; Amelka: 27.8 (Krzyżewo) and 27.9 (Uhnin).

The sowing dates for *Dactylis glomerata* were 22 April 2011 (Krzyżewo), 6 May 2011 (Uhnin) for *Festuca pratensis* 22 May 2010 (Krzyżewo) and 29 April 2010 (Uhnin).

In the year when the experiment was set up the grass was not harvested and only weeds were mowed. According to the guidelines of Research Centre for Cultivar Testing, the full exploitation of *Dactylis glomerata* varieties was due between 2012 and 2013, whereas for *Festuca pratensis* it was due between 2011 and 2012. In the experimental plots with the varieties of *Dactylis glomerata* the grass was harvested six times a year and chemical analysis of the biomass was

**Table 1. Soil conditions**

Grass	<i>Dactylis glomerata</i>		<i>Festuca pratensis</i>	
	Krzyżewo	Uhnin	Krzyżewo	Uhnin
Location experiment	Krzyżewo	Uhnin	Krzyżewo	Uhnin
Type soil	Mineral	Organic	Mineral	Organic
<b>Soil conditions</b>				
The value of soil according to IUNG	52	50	52	70
Agricultural value	5	1p	5	1z
Type	P	PS	P	PS
Texture	ls	-	ls	-
pH	6.7	5.5	6.7	5.5

Symbols: 1p – good and very good permanent meadow; 5 – good quality rye soil; P – podsollic soil; PS – peaty soil; ls – loamy sand; 1z – very good and good grassland

**Table 2. Mineral fertilizers used in the experiment with varieties of *Dactylis glomerata* and *Festuca pratensis***

	Fertilizers in kg·ha <sup>-1</sup>			
	Krzyżewo	Uhnin	Krzyżewo	Uhnin
Location experiment	Krzyżewo	Uhnin	Krzyżewo	Uhnin
Soil type	Mineral	Organic	Mineral	Organic
Nitrogen – N: before sowing,	270	80	80	80
in consecutive years (annual dose)	245	192	80	80
Phosphorus – P <sub>2</sub> O <sub>5</sub> : before sowing,	90	100	80	80
in consecutive years (annual dose)	80	100	80	80
Potassium – K <sub>2</sub> O: before sowing,	90	100	100	100
in consecutive years (annual dose)	130	110	100	100

**Table 3. Sielianinov's hydrothermal index (K) during the growing season in the years of the experiment in Krzyżewo and Uhnin**

Month	Location experiment/ Soil type					
	Krzyżewo/ Mineral			Uhnin/Organic		
	Year of experiment					
	2011	2012	2013	2011	2012	2013
IV	0.86 (s)	1.63 (dw)	2.50 (w)	1.39 (o)	1.06 (ds)	2.79 (bw)
V	1.64 (dw)	1.09 (ds)	1.80 (dw)	1.09 (ds)	0.84 (s)	2.87 (bw)
VI	1.06 (ds)	1.83 (dw)	1.53 (o)	2.12 (w)	1.92 (dw)	1.74 (dw)
VII	3.90 (sw)	1.55 (o)	1.08 (ds)	3.03 (sw)	0.81 (s)	0.92 (s)
VIII	1.15 (ds)	3.18 (sw)	0.89 (s)	0.79 (s)	1.25 (ds)	0.12 (ss)
IX	0.41 (bs)	0.40 (ss)	4.84 (sw)	0.21 (ss)	0.79 (s)	2.46 (w)
X	0.81 (s)	2.27 (w)	0.48 (bs)	1.27 (ds)	4.90 (sw)	0.46 (bs)

Note: (ss) – extremely dry; (bs) – very dry; (s) – dry; (ds) – quite dry; (o) – optimal; (dw) – quite wet; (w) – wet; (bw) – very wet; (sw) – extremely wet

done taking dry matter only from five cuts. The varieties of *Festuca pratensis* were harvested four times. Each year in the course of the experiment fresh and dry matter of each cut were weighed. In the phase of inflorescences the plants were harvested.

Research Centre for Cultivar Testing made those measurements available to be used in this paper.

The airy dry matter was shredded and ground. The obtained material was subjected to chemical analysis to determine dry matter (by determining moisture content), protein compounds and simple sugars. The method of determination was near-infrared spectroscopy (NIRS) using a NIRFlex N-500 spectrometer and ready to use INGOT calibration applications.

Tukey's test was used to find means that were significantly different from each other, at the significance level of  $LSD_{0.05}$ .

Climatic conditions of the area where the experiment was carried out are typical for the 9th agricultural and climatic eastern part of Poland. The average annual air temperature varies from 6.7 to 6.9°C and in the summer season the average 24 hour temperature is 15°C. The growing season usually starts on 28 March, lasts till 30 October and is 200 to 220 days long. The average climatic water balance during the time of the experiment varied considerably according to the period and location. Annual rainfall ranges from 550 to 650 mm, with not frequent but recurrent rain. Water stress was mainly observed in spring while water deficit occurred in July. During the time of the experiment weather data were provided by the Meteorological and Hydrological Stations in Krzyżewo and Uhnin. To determine temporal variation of meteorological parameters and their impact on plant growth Sielianinov's hydrothermal index was used with the month's classification according to Skowera and Puła [12]. As it can be seen from Table 3 space-time distribution of annual rainfall varied. April was a month of water stress only in 2011 in Krzyżewo ( $K = 0.86$ ), whereas in May water deficit was noted in Uhnin in 2012 ( $K = 0.84$ ). Every year in June there was enough rain both in Krzyżewo and Uhnin ( $K$  between 1.06 and 2.12). July was either extremely wet (Krzyżewo 2011  $K = 3.9$ , Uhnin  $K = 3.03$ ) or dry (Krzyżewo and Uhnin 2013). However, on the whole both July and August were rather dry whereas September and October happened to be extremely dry one year each with Sielianinov's hydrothermal index more than 4, (Krzyżewo 2013 and Uhnin 2012).

### 3. RESULTS AND DISCUSSION

The protein content in dry matter of cocksfoot depending on experimental factors (Table 4) ranged from 116 g kg<sup>-1</sup> DM in V-th regrowth of Niva variety to 204 g kg<sup>-1</sup> DM in I-th cut of CR variety on organic soils. The variability of traits affected the significance of the interaction soil type x cut x variety. According to Falkowski et al. [2], for the meadow grass optimum total protein content should be between 100 and 200 g·kg<sup>-1</sup> dry matter. Barszczewski et al. [13] reported that increase of protein content in the feed increase the digestibility, regardless of its quality [14].

**Table 4. The protein content in dry matter of cocksfoot varieties depending on the type of soil and cut**

Experimental factor		Total protein g·kg <sup>-1</sup> DM		
		Soil type (A)		Mean
		Mineral	Organic	
Niva	I	190	174	182
	II	159	173	166
	III	184	184	184
	IV	161	125	143
	V	173	116	145
Tukan	I	191	199	195
	II	174	165	169
	III	179	197	188
	IV	169	146	158
	V	184	148	166
Amila	I	181	190	189
	II	169	183	176
	III	168	167	167
	IV	166	133	149
	V	182	133	158
CR	I	167	204	185
	II	146	189	165
	III	167	179	173
	IV	149	145	147
	V	173	138	146
Cut (B)	I	184	192	188
	II	162	178	169
	III	174	182	178
	IV	162	137	149
	V	178	134	156
Variety (C)	Niva	173	154	164
	Tukan	179	170	175
	Amila	175	161	163
	CR	160	171	165
Year (D)	2012	158	180	169
	2013	186	149	168
Mean		172	165	169

$LSD_{p=0.05}$  for:  $AxBxC - 16.5$ ,  $AxB - 18.1$ ,  $AxC - 15.2$ ,  $AxD - 17.3$ ,  $BxC - 19.8$ ,  $A - r.n.$ ,  $B - 9.8$ ,  $C - 18.3$ ,  $D - r.n.$ ;  $r.n.$  - difference not significant

Levels of major factors such as soil type and variety, no significant influence on differences in total protein content in orchard grass. It confirmed relationship to widely described in the literature [5,7,15].

**Table 5. The protein content in dry matter of tall fescue cultivars depending on the type of soil and cut**

Experimental factor		Total protein g·kg <sup>-1</sup> DM		
		Soil type (A)		Mean
		Mineral	Organic	
Limosa	I	130	129	130
	II	104	99	102
	III	129	136	132
	IV	184	170	177
Pasja	I	162	174	168
	II	176	134	154
	III	140	159	150
	IV	148	155	152
Anturka	I	149	149	149
	II	162	160	161
	III	154	164	159
	IV	154	157	155
Amelka	I	157	137	147
	II	143	105	124
	III	124	102	113
	IV	126	134	130
Cut (B)	I	149	147	148
	II	146	125	135
	III	137	140	139
	IV	153	154	153
Variety (C)	Limosa	139	134	137
	Pasja	159	154	157
	Anturka	156	157	156
	Amelka	130	125	128
Year (D)	2011	142	140	141
	2012	149	144	147
Mean		146	142	144

LSD<sub>p=0.05</sub> for: AxBxC – 13.8, AxB – r.n.; AxC – r.n., AxD – r.n.; BxC – 20.1, A – r.n., B – r.n., C – 12.6, D – r.n.; r.n. - difference not significant

In the case of tall fescue was highlighted the importance of differences of protein content in the biomass of individual varieties (Table 5) (above). The highest protein content in dry matter were characterized a variety of Pasja (157 g kg<sup>-1</sup> DM) and Anturka (156 g kg<sup>-1</sup> DM), whereas the least Amelka variety (128 g kg<sup>-1</sup> DM). It should be emphasized that regardless of variety and soil type, the cut did not significantly affect on this trait, although the most of protein content was achieved in the harvesting of first and fourth regrowth (from 148 to 153 g kg<sup>-1</sup> DM). Soil type had no significant effect on the protein content in

harvested feed, also interact with the year studies did not affect on the differences of this means.

Barszczewski et al. [13] reported that an important element of chemical content, especially green fodder for silage is the sugar content soluble in water. They are a source of food for lactic acid bacteria, which are responsible for the fermentation processes [11,14].

**Table 6. The mean ratio of carbohydrates to protein in dry matter of cocksfoot variety depending on the type of soil and cut**

Experimental factor		Ratio of carbohydrates/protein		
		Soil type (A)		Mean
		Mineral	Organic	
Niva	I	0.48	0.36	0.42
	II	0.91	0.64	0.73
	III	0.31	0.29	0.30
	IV	0.30	0.70	0.50
	V	0.47	0.79	0.63
Tukan	I	0.46	0.34	0.40
	II	0.58	0.61	0.60
	III	0.33	0.52	0.43
	IV	0.26	0.66	0.46
	V	0.48	0.55	0.52
Amila	I	0.58	0.33	0.46
	II	0.70	0.59	0.64
	III	0.36	0.40	0.38
	IV	0.28	0.68	0.40
	V	0.42	0.50	0.46
CR	I	0.67	0.28	0.48
	II	0.73	0.53	0.62
	III	0.38	0.37	0.38
	IV	0.32	0.57	0.45
	V	0.39	0.55	0.47
Cut (B)	I	0.55	0.33	0.44
	II	0.71	0.52	0.62
	III	0.35	0.40	0.38
	IV	0.29	0.65	0.47
	V	0.44	0.60	0.52
Variety (C)	Niva	0.47	0.56	0.52
	Tukan	0.42	0.54	0.48
	Amila	0.47	0.50	0.49
	CR	0.50	0.46	0.48
Year (D)	2012	0.39	0.49	0.42
	2013	0.55	0.55	0.55
Mean		0.47	0.52	0.55

LSD<sub>p=0.05</sub> for: AxBxC – 0.12, AxB – 0.14, AxC – 0.07, AxD – 0.07, BxC – 0.12, A – 0.04, B – 0.07, C – 0.03, D – 0.11

In the analyzed plant material of cocksfoot (Table 6) the ratio of sugars to proteins ranged

from 0.26 for the plants of Toucan variety collected on mineral soils in the fourth cut to 0.91 for plants of Niva varieties also on those soils in the second cut. Regardless of the type of soil the highest value ratio of carbohydrate and protein in relation to the average value was characterized by Niva variety (0.52), while the lowest value (0.48) have Tukan and CR varieties. The big difference in sugar-protein relation also occurred within cuts. Regardless of variety, soil type and year of research, the widest ratio (0.62) had the plants collected in the second cut. Also relatively high value (0.52) was found in the biomass of the V-th cut. In contrast, the lowest value (0.38) was obtained in III-th cut. In addition it worth noting, that cultivation on organic soil regardless of the other experiment factors, contributed to the

growth of this ratio from the value of 0.47 (mineral soil) to 0.52 (organic soil). This ratio of sugars to proteins are typical for grasses, as indicated the results of the nutritional value of *Festulolium braunii* [16] and *Lolium multiflorum* [17]. It should be noted that the value of this parameter for the meadow sward, as demonstrated the research of Jankowska-Huflejt and Wróbel [3], were three times higher from 0.89 to 2.00 than those presented in work for cocksfoot. The plant material of meadow fescue was characterized by a wider ratio of carbohydrate and protein (Table 7). This values ranged from 0.53 for Anturka variety collected in III-th cut on mineral soils to 1.70 for Limosa variety collected in second cut also on these soils.

**Table 7. The mean ratio of carbohydrates to protein in dry matter of tall fescue cultivars depending on the type of soil and cut**

Experimental		Ratio of carbohydrates/protein		
		Soil type (A)		Mean
		Mineral	Organic	
Limosa	I	1.00	0.83	0.92
	II	1.70	1.30	1.50
	III	0.78	0.83	0.81
	IV	0.71	0.67	0.69
Pasja	I	1.10	0.74	0.92
	II	0.56	0.86	0.71
	III	0.81	0.75	0.78
	IV	0.79	0.77	0.78
Anturka	I	0.76	0.81	0.79
	II	0.74	0.74	0.74
	III	0.53	0.79	0.66
	IV	0.89	0.69	0.79
Amelka	I	0.70	1.07	0.89
	II	0.69	1.08	0.89
	III	1.12	1.19	1.16
	IV	1.00	0.92	0.96
Cut (B)	I	0.89	0.86	0.88
	II	0.92	0.99	0.96
	III	0.81	0.89	0.85
	IV	0.85	0.76	0.81
Variety (C)	Limosa	0.85	0.93	0.89
	Pasja	0.80	0.74	0.77
	Anturka	0.76	0.76	0.76
	Amelka	0.95	1.00	0.89
Year (D)	2011	0.81	0.79	0.80
	2012	0.87	0.93	0.90
Mean		0.84	0.86	0.85

LSD<sub>p=0.05</sub> for: AxBxC – 0.13, AxB – 0.08, AxC – 0.07, AxD – 0.09, BxC – 0.09, A – r.n., B – 0.09, C – 0.10, D – 0.09

According to Barszczewski et al. [13], on the ratio of carbohydrates to proteins in feed is mainly influenced by the participation of legumes. The authors reported that increasing of the share of protein plants (red clover) in the sward cause the decrease of this ratio value.

#### 4. CONCLUSIONS

Regardless of variety, cut and years of research, higher total protein content occurred in dry matter of cocksfoot grown on mineral soil than organic. Of the tested cultivars of cocksfoot the most preferred feed expressed with the high protein content characterized the feed of Tukan variety and from a variety of meadow fescue Pasja. Higher values of sugar-protein ratios characterized biomass of varieties of meadow fescue than cocksfoot. Regardless of the examined grass species, better nutritional value had the plant varieties located on mineral soils than organic.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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