

# The effect of different proportions of *Medicago media* Pers. in mixtures with *Festulolium braunii* (K. Richt.) A. Camus on the yield and feed value of green fodder

Marzenna Olszewska, Stefan Grzegorzczak and Anna Bałuch-Matecka

Department of Grassland and Green Space Management  
University of Warmia and Mazury in Olsztyn, 10–719 Olsztyn, Poland  
e-mail: marzenna.olszewska@uwm.edu.pl

A field experiment was performed in 2011–2013. The aim of the study was to determine the effect of different proportions of *Medicago media* in mixtures with *Festulolium braunii* on the yield and feed value of green fodder, including the content of crude protein (CP) and water-soluble carbohydrates (WSC), and the carbohydrate-to-protein ratio. It was found that alfalfa had a high competitive advantage over *Festulolium*. In the second and third year of full utilization, alfalfa was the predominant species in the tested mixtures regardless of the proportion of sown seeds. The mixtures with alfalfa were characterized by high and stable dry matter yields and high protein content, whereas *Festulolium* significantly increased the content of WSC and improved the WSC:CP ratio. The evaluated mixtures had a higher feed value than pure-sown components. The results of the study indicate that *Festulolium* mixtures with a lower proportion (30% and 50%) of alfalfa are characterized by a higher feed value.

*Key words:* chemical composition, forage biomass, grass, alfalfa

## Introduction

Protein content and energy value are critical factors in qualitative evaluations of roughage for cattle. These parameters vary considerably and are influenced by numerous factors, including the species composition of the sward, the growth stage of plants, weather conditions, season and time of day (Andrzejewska et al. 2013, Mäkinen et al. 2016, Purwin et al. 2016, Staniak 2016, Staniak and Harasim 2018). Roughage deficient in protein and energy has to be supplemented with concentrate and specialty feed additives, which significantly increases costs and decreases production profitability (Huhtanen and Broderick 2016). The content of protein and carbohydrates, the main sources of energy, in fodder crops per hectare is a very important consideration in cattle farming, and it has been widely discussed in the literature (Peyraud et al. 2014, Peyraud and Peeters 2016, Johansen et al. 2016, Jankowski et al. 2018).

Due to its high protein content, alfalfa is one of the major feed ingredients around the world (Seppänen et al. 2018). In many countries, alfalfa is the basic component of feed rations for beef and dairy cattle. The main limitation of alfalfa as a feed ingredient is that its protein is less available for cattle than the protein from other types of roughage. The above can be attributed to the fact that alfalfa protein is readily soluble in water and undergoes rapid degradation in the rumen. Rapid decomposition of protein decreases the efficiency of its bacterial synthesis, which makes it difficult to meet the nutrient requirements of cattle with regard to protein digested in the small intestine. Microbial protein synthesis is also negatively affected by low concentrations of carbohydrates (Broderick 1995, Jafari 2012, Andrzejewska et al. 2013). Therefore, alfalfa should be grown with grasses to minimize protein loss. The presence of grasses in mixtures increases the carbohydrate content of biomass and contributes to the production of feed with an optimal energy and protein balance (Sosnowski 2011, Simili da Silva et al. 2013, Luscher et al. 2014, Vasileva and Naydenova 2017). According to the literature, feeds with a high content of water-soluble carbohydrates (WSC) improve protein utilization, which increases milk yields and body weight gains, and decreases nitrogen loss (Miller et al. 2001, Evans et al. 2011).

Research has demonstrated that *Festulolium braunii* is a highly suitable component in mixtures with alfalfa due to its high carbohydrate content (Sosnowski and Jankowski 2010, Sosnowski 2011, Olszewska and Kobyliński 2016, Staniak 2016, Staniak and Harasim 2018). The optimal proportion of component crops in mixtures is an important consideration. The aim of this study was to determine the effect of different proportions of alfalfa in mixtures with *Festulolium* on the yield and feed value of green fodder, including the content of crude protein (CP) and WSC, and the carbohydrate-to-protein ratio (WSC:CP).

## Materials and methods

### Field experiment

The study was conducted in 2011–2013 in Poland. A small-area field experiment was performed at the Agricultural Experimental Station in Bałdy (53°72'N; 20°42'E) owned by the University of Warmia and Mazury in Olsztyn. The experiment was established on mineral soil of quality class IVa and very good rye complex according to the Polish soil classification system (Systematyka gleb Polski 1989). The arable layer was characterized by very low phosphorus content (4.6 mg P<sub>2</sub>O<sub>5</sub> in 100 g of soil) and moderate concentrations of potassium (14.9 mg K<sub>2</sub>O in 100 g of soil) and magnesium (5.0 mg in 100 g of soil). Soil pH was neutral (pH<sub>KCl</sub> 7.2). The experiment was established in the spring of 2010, and it had a randomized block design with four replications. The analyzed species were *Festulolium brauni* (Fb) cv. Sulino and *Medicago media* (Mm) cv. Radius. The experimental factor was the proportion of alfalfa seeds in the mixture: 30%, 50% and 70%. Pure-sown *Festulolium* and pure-sown alfalfa were the control treatments. Plot area was 10 m<sup>2</sup>. Before sowing, nitrogen, phosphorus and potassium fertilizers were applied to all plots at the following rates: 30 kg N, 80 kg P<sub>2</sub>O<sub>5</sub> and 60 kg K<sub>2</sub>O ha<sup>-1</sup>. In pure-sown treatments, the seeding rate was 40 kg ha<sup>-1</sup> for *Festulolium* and 15 kg ha<sup>-1</sup> for alfalfa. In the experimental treatments, the seeding rates corresponded to the percentages of the analyzed species in the mixture. Seeds were broadcast, and cover crops were not used. In the years of full utilization, pure-sown *Festulolium* was fertilized with nitrogen at 180 N ha<sup>-1</sup>, mixtures were fertilized with nitrogen at 90 kg N ha<sup>-1</sup>, whereas pure-sown was not fertilized with nitrogen. Nitrogen fertilizer was split into three equal doses and applied in early spring, after the first and second harvest. All plots were fertilized with phosphorus at 80 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> (one spring application) and potassium at 120 kg K<sub>2</sub>O ha<sup>-1</sup> (in spring and after the first harvest).

### Botanical and chemical analyses

The sward was mowed three times in every growing season. The first regrowth of mixtures was mowed at the beginning of *Festulolium* heading and the next one at the stage of lucerne budding. One-kilogram samples of green matter were collected after each harvest to determine the dry matter yield and the chemical composition of swards. The actual proportion of alfalfa in the mixtures was determined in botanical analyses on a weight basis. The content of WSC in plant material was determined with the anthrone reagent, and CP content was determined by the Kjeldahl method. The WSC:CP ratio were calculated.

### Statistical analysis

The results were processed statistically by analysis of variance (ANOVA) in the Statistica 12.0 program. The significance of differences was determined with Tukey's test at  $p < 0.05$ .

### Weather conditions

Weather conditions during 2011–2013 are presented in Table 1. The growing season of 2011 was characterized by favorable conditions for plant growth and development, and average monthly temperatures approximated the long-term average.

Table 1. Mean air temperature and rainfalls in the years 2011–2013

Years	The vegetation period						Mean
	IV	V	VI	VII	VIII	IX	
Mean air temperature (°C)							
2011	9.1	13.1	17.1	17.9	17.6	14.1	14.8
2012	7.8	13.4	15.0	19.0	17.7	13.5	14.4
2013	5.9	14.8	17.5	18.0	17.4	11.3	14.2
Multi-year	7.7	13.5	16.1	18.7	17.9	12.8	14.5
Rainfalls (mm)							
2011	22.5	51.1	81.7	202.8	82.1	67.5	84.6
2012	73.1	51.1	103.2	121.0	45.1	45.7	73.3
2013	28.5	54.5	61.2	121.9	37.6	101.1	67.5
Multi-year	33.3	58.5	80.4	74.2	59.4	56.9	60.5

With the exception of April and October, total precipitation in 2011 exceeded the long-term average, and rainfall levels in July were nearly four-times higher than the long-term average. The second year of full utilization was characterized by moderate average temperatures, high precipitation in April, June and July, and a minor water deficit in August and September. In 2013, weather conditions were less favorable for plant growth due to low temperatures in early spring and considerable rainfall deficiency in June and August.

## Results and discussion

### Proportion of alfalfa in sward of mixtures

The proportion of alfalfa plants in the analyzed sward of mixtures differed considerably from the proportion of sown seeds. In the first year of full utilization, alfalfa developed at a much slower rate than in successive years of the study. In the first spring cut, alfalfa accounted for 20% to 37% of the sward, and its proportion was lower relative to the percentage of alfalfa seeds sown in the tested mixtures (Fig. 1). The proportion of alfalfa continued to increase after every successive harvest and reached 54–57% in the third-cut herbage. The greatest increase was noted in the mixture sown with 30% Mm seeds and 70% Fb seeds. Alfalfa plants develop a deep and extensive root system in initial stages of growth, which slows down the development of aerial biomass and increases the competitive advantage of grasses. The production of aerial biomass is intensified after successive harvests when alfalfa plants draw upon the nutrient reserves accumulated in the root collar (Jelinowska and Staniak 2007, Vasileva and Naydenova 2017). Alfalfa developed rapidly in 2012 and 2013, and it was the predominant component of the analyzed mixtures in the second- and third-cut herbage when its proportions considerably exceeded the percentage of sown seeds. Sosnowski and Jankowski (2010) and Gawęł (2007, 2008) also reported on the high competitive advantage of alfalfa over *Festulolium* in successive years of utilization. In the present study, hybrid alfalfa cv. Radius was highly competitive over *Festulolium* cv. Sulino, in particular during dry spells in April, June and August 2013. Due to its shallow root system, *Festulolium* is less drought tolerant than alfalfa, and its development was strongly inhibited in the above months.

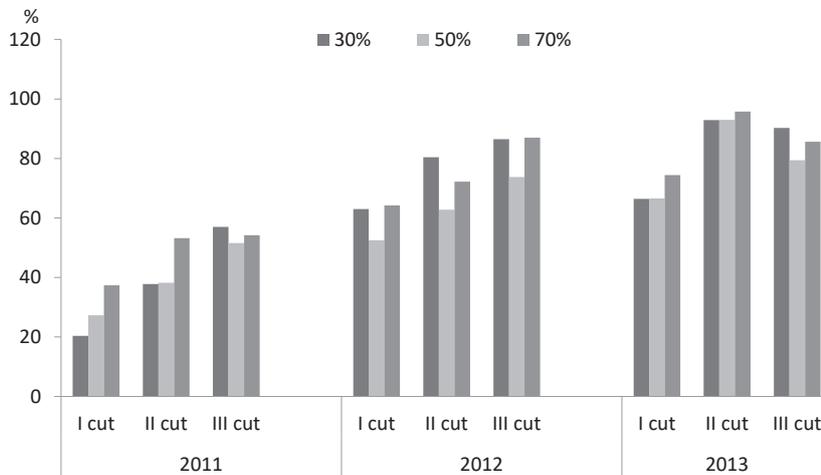


Fig. 1. The share of alfalfa in the yields of mixtures with various proportions of alfalfa (30%, 50%, 70%)

### Dry matter yield

In the first year of full utilization, mixtures produced significantly higher yields than pure-sown alfalfa and pure-sown *Festulolium* (Table 2). The highest yield was noted in the mixture composed of 30% alfalfa and 70% *Festulolium*. The total dry matter yield of the above mixture was 13.5% higher in comparison with pure-sown alfalfa and 8.2% higher in comparison with pure-sown *Festulolium*. The mixtures containing 50% and 70% alfalfa produced somewhat lower yields, but not significantly different. At the first harvest, the highest yield was noted in pure-sown *Festulolium*, and the lowest yield was observed in pure-sown alfalfa. Reverse relationships were noted in the second- and third-cut herbage. In the first- and second-cut herbage, the differences in the yields of mixtures with various proportions of alfalfa were not significant, whereas at the third harvest, the mixture composed of 30% Mm and 70% Fb was characterized by a significantly higher yield, than the mixture of 70% Mm and 30% Fb.

In the second year of full utilization, mixture yields were determined in the range of 10.78–11.58 Mg ha<sup>-1</sup> and were approximately 47% higher relative to the yield of pure-sown *Festulolium* (Table 2). The highest yield was noted in the 70% Mm+30% Fb mixture, and it was comparable to that of pure-sown alfalfa. Regardless of the botanical composition of the sward, the first-cut herbage had the highest share, and the third-cut herbage had the lowest share of total yield.

Table 2. Dry matter yield (Mg ha<sup>-1</sup>)

Treatment	I cut	II cut	III cut	Sum
2011				
Mm 100%	2.96 a	3.48 b	3.80 bc	10.24 a
Fb 100%	5.00 c	2.94 a	2.81 a	10.74 b
Mm70%+ Fb30%	4.30 b	3.01 ab	3.60 b	10.91 bc
Mm50%+ Fb50%	3.96 b	3.23 ab	3.97 cd	11.15 bc
Mm30%+ Fb70%	4.39 b	3.01 ab	4.22 d	11.62 c
2012				
Mm 100%	5.75 c	3.73 c	2.45 c	11.93 c
Fb 100%	3.70 a	2.58 a	1.08 a	7.35 a
Mm70%+ Fb30%	5.55 c	3.72 c	2.30 bc	11.58 c
Mm50%+ Fb50%	5.38 c	3.25 b	2.15 b	10.78 b
Mm30%+ Fb70%	4.98 b	3.65 c	2.18 bc	10.80 b
2013				
Mm 100%	4.83 b	3.60 b	2.88 d	11.30 c
Fb 100%	2.63 a	1.13 a	0.68 a	4.43 a
Mm70%+ Fb30%	4.53 b	3.43 b	2.35 c	10.30 bc
Mm50%+ Fb50%	4.45 b	3.18 b	1.88 b	9.50 b
Mm30%+ Fb70%	4.53 b	3.28 b	2.50 cd	10.30 bc
2011–2013				
Mm 100%	4.51 b	3.60 c	3.04 d	11.15 c
Fb 100%	3.77 a	2.21 a	1.52 a	7.51 a
Mm70%+ Fb30%	4.79 b	3.39 bc	2.75 bc	10.93 c
Mm50%+ Fb50%	4.59 b	3.22 b	2.66 b	10.47 b
Mm30%+ Fb70%	4.63 b	3.31 b	2.97 cd	10.91 c

Mm = *Medicago media*; Fb = *Festulolium braunii*

Values within a column marked with different letter are significantly different at  $p < 0.05$ .

In the last year of the study, the highest yield was noted in pure-sown alfalfa (11.30 Mg ha<sup>-1</sup>), whereas somewhat lower yields were observed in mixtures, in particular in the mixture with equal proportions (50%) of both components. In the 50% Mm+50% Fb mixture, yields were approximately 16% lower than in pure-sown alfalfa and more than twice higher than in pure-sown *Festulolium*. Similarly to the previous years of the study, the first-cut herbage had the highest share of total yield. In successive years, the greatest decrease in yield was observed in pure-sown *Festulolium*. The dry matter yield of *Festulolium* decreased by 32% in the second year and by 59% in the third year of the experiment, which suggests that alfalfa grown in mixtures stabilized yields in growing seasons with variable weather conditions. The yields of the analyzed mixtures were higher and more stable in comparison with the yields of pure-sown species due to better utilization of space, different growth and development rates of mixture components, and more complementary utilization of environmental resources (Vasileva and Vasilev 2012, Luscher et al. 2014, Vasileva and Ilieva 2016, Staniak and Harasim 2018).

An analysis of average annual dry matter yields over three years revealed that the yields of the mixtures containing 30% and 70% alfalfa were similar to the yields of pure-sown alfalfa and around 6% higher than the yield of the mixture containing 50% alfalfa. Pure-sown *Festulolium* was characterized by the lowest average annual dry matter yield.

Content and yield of crude protein

Protein is the most expensive component of ruminant feeds, and protein yields significantly affect production profitability. The average total protein yield during the 3-year studies was lowest in pure-sown *Festulolium* (0.81 Mg ha<sup>-1</sup>) and highest in pure-sown alfalfa (1.90 Mg ha<sup>-1</sup>). The mixtures containing alfalfa were characterized by significantly higher protein yield per unit area than pure sown Fb (Table 3). Subject to the proportion of alfalfa seeds in the seed mixture, the protein yield of mixtures was approximately 73–96% higher than in pure-sown *Festulolium* and approximately 16–26% lower than in pure-sown alfalfa. In the group of the analyzed mixtures, protein yield was lowest in the mixture containing 50% alfalfa (1.40 Mg ha<sup>-1</sup>). Protein yields were higher and similar in the mixtures containing 30% and 70% alfalfa. Minor differences in the protein yield of pure-sown alfalfa and mixtures and a clear decrease in the protein yield of pure-sown *Festulolium* were observed across the experimental years. An analysis of the average values for the 3-year experiment revealed that protein yields were highest in the first-cut herbage and continued to decrease after successive harvests (Table 3).

Table 3. Total protein yield (Mg ha<sup>-1</sup>)

Treatment	I cut	II cut	III cut	Sum
2011				
Mm 100%	0.53 ab	0.62 c	0.65 c	1.80 d
Fb 100%	0.49 a	0.26 a	0.28 a	1.03 a
Mm70%+ Fb30%	0.58 b	0.39 b	0.43 b	1.39 b
Mm50%+ Fb50%	0.51 ab	0.40 b	0.45 b	1.36 b
Mm30%+ Fb70%	0.54 ab	0.40 b	0.59 c	1.52 c
2012				
Mm 100%	0.88 d	0.63 d	0.42 c	1.93 e
Fb 100%	0.37 a	0.30 a	0.15 a	0.82 a
Mm70%+ Fb30%	0.75 c	0.54 c	0.41 bc	1.71 d
Mm50%+ Fb50%	0.56 b	0.39 b	0.37 b	1.31 b
Mm30%+ Fb70%	0.54 b	0.57 c	0.40 bc	1.51 c
2013				
Mm 100%	0.93 c	0.59 b	0.46 d	1.97 c
Fb 100%	0.31 a	0.16 a	0.11 a	0.58 a
Mm70%+ Fb30%	0.71 b	0.58 b	0.38 c	1.67 b
Mm50%+ Fb50%	0.72 b	0.52 b	0.29 b	1.53 b
Mm30%+ Fb70%	0.75 b	0.54 b	0.39 c	1.68 b
2011–2013				
Mm 100%	0.78 d	0.61 d	0.51 e	1.90 d
Fb 100%	0.39 a	0.24 a	0.18 a	0.81 a
Mm70%+ Fb30%	0.38 c	0.50 c	0.41 c	1.59 c
Mm50%+ Fb50%	0.60 b	0.44 b	0.37 b	1.40 b
Mm30%+ Fb70%	0.61 b	0.50 c	0.46 d	1.57 c

Mm = *Medicago media*, Fb = *Festulolium braunii*

Values within a column marked with different letter are significantly different at  $p < 0.05$ .

Protein yield was calculated based on the dry matter yield and the CP content of plants. Pure-sown alfalfa was characterized by the highest average protein content which was determined at 168.0 g kg<sup>-1</sup> on a dry matter basis and was 70% higher than in pure-sown *Festulolium* (Fig. 2). The protein content of the evaluated mixtures ranged from 138.3 to 148.0 g kg<sup>-1</sup> and was 17.5–25.7% higher than in pure-sown *Festulolium* and 11.9–17.7% lower than in pure-sown alfalfa, subject to the proportion of alfalfa seeds in the analyzed mixture. The mixture with equal proportions (50%) of both components was characterized by the lowest protein content. The remaining mixtures did not differ with regard to the above parameter. Only minor differences were found in the protein content of successive cuts. The protein content of pure-grown *Festulolium* increased at successive harvests, whereas the reverse was observed in pure-grown alfalfa.

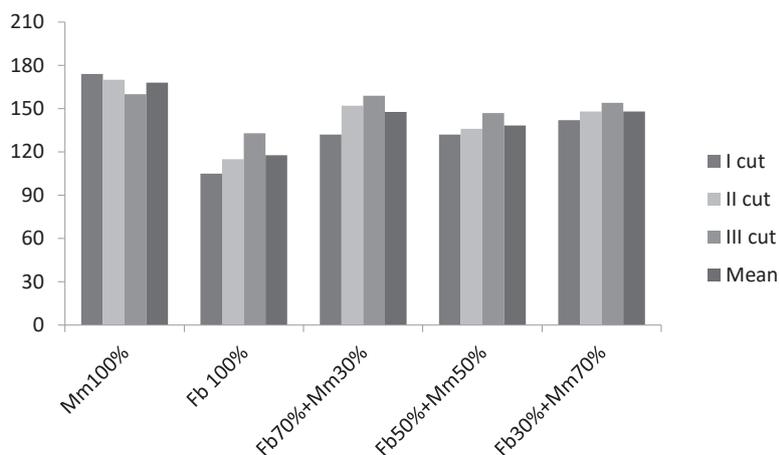


Fig. 2. Content of crude protein (average for years)

### Content and yield of water-soluble carbohydrates

The energy content of feeds determines the utilization of feed proteins. Water-soluble carbohydrates play an important role in cattle nutrition as the main source of highly available energy for microbiological processes in the rumen (Evans et al. 2011, Jafari 2012). In our study, pure-sown *Festulolium* was characterized by the highest average total WSC yield (1.10 Mg ha<sup>-1</sup>) which was approximately 156% higher relative to pure-sown alfalfa (0.44 Mg ha<sup>-1</sup>). As a result, the WSC yield of the mixtures containing *Festulolium* was higher. Depending on the composition of the analyzed mixtures, their WSC yields were around 72–100% higher relative to pure-sown alfalfa. The mixtures containing 30% and 50% alfalfa produced significantly higher WSC yields than the mixture containing 70% alfalfa (Table 4). The above trend was observed in the first and second year of full utilization. In pure-sown *Festulolium* and in the mixtures, the average WSC yields were highest in the first-cut herbage, whereas the WSC yields of pure-sown alfalfa did not differ significantly between harvests.

Table 4. Water soluble sugars yield (Mg ha<sup>-1</sup>)

Treatment	I cut	II cut	III cut	Sum
2011				
Mm 100%	0.16 a	0.09 a	0.18 a	0.43 a
Fb 100%	0.84 d	0.28 d	0.37 d	1.49 e
Mm70%+ Fb30%	0.44 b	0.12 b	0.29 c	0.85 b
Mm50%+ Fb50%	0.54 c	0.13 b	0.40 e	1.07 d
Mm30%+ Fb70%	0.51 c	0.16 c	0.26 b	0.93 c
2012				
Mm 100%	0.13 a	0.19 a	0.12 a	0.44 a
Fb 100%	0.31 e	0.29 c	0.11 a	0.71 d
Mm70%+ Fb30%	0.25 d	0.22 b	0.17 b	0.64 c
Mm50%+ Fb50%	0.17 b	0.20 b	0.20 c	0.57 b
Mm30%+ Fb70%	0.21 c	0.35 d	0.23 d	0.80 e
2011–2012				
Mm 100%	0.15 a	0.14 a	0.15 a	0.44 a
Fb 100%	0.57 c	0.29 d	0.24 b	1.10 d
Mm70%+ Fb30%	0.34 b	0.17 b	0.23 b	0.74 b
Mm50%+ Fb50%	0.35 b	0.17 b	0.30 c	0.82 c
Mm30%+ Fb70%	0.36 b	0.26 c	0.25 b	0.86 c

Mm = *Medicago media*, Fb = *Festulolium braunii*

Values within a column marked with different letter are significantly different at  $p < 0.05$ .

The WSC yield was determined by the WSC content of dry matter. The biomass of pure-sown alfalfa was characterized by very low WSC content (Fig. 3). This was estimated at 40 g kg<sup>-1</sup> and was more than 2.5-fold lower than in pure-sown *Festulolium*. The combination of alfalfa and *Festulolium* in the mixtures increased WSC concentrations and improved the feed value of green fodder. The average WSC content in the mixtures composed of 30% and 50% alfalfa reached 79.4 and 77.5 g kg<sup>-1</sup>, respectively, and it was much lower in the mixture containing 70% alfalfa (66.6 g kg<sup>-1</sup>). Differences in WSC content were also noted during the growing season. Carbohydrate concentrations were highest in the first-cut herbage of pure-sown *Festulolium*. In all treatments, the WSC content of dry matter was lowest in the second-cut herbage, in particular in the mixtures containing 50% and 70% alfalfa. Carbohydrate concentrations increased in the third-cut herbage. Similar results were reported by other authors who noted considerable variations in the WSC content during the growing season (Ciepiela 2014, Purwin et al. 2016). According to Griggs et al. (2005), the WSC content of grasses is highest in spring, lower in summer, and it increases again in autumn. The decrease in the WSC content of plants in summer can be attributed to high temperatures and high humidity. High air temperatures increase respiration and speed up sugar depletion in plants. Light exposure also plays a role in sugar accumulation in plant tissues. Sunlight increases the rate of photosynthesis, which directly influences sugar production and leads to variations in carbohydrate content during the growing season (Jafari 2012).

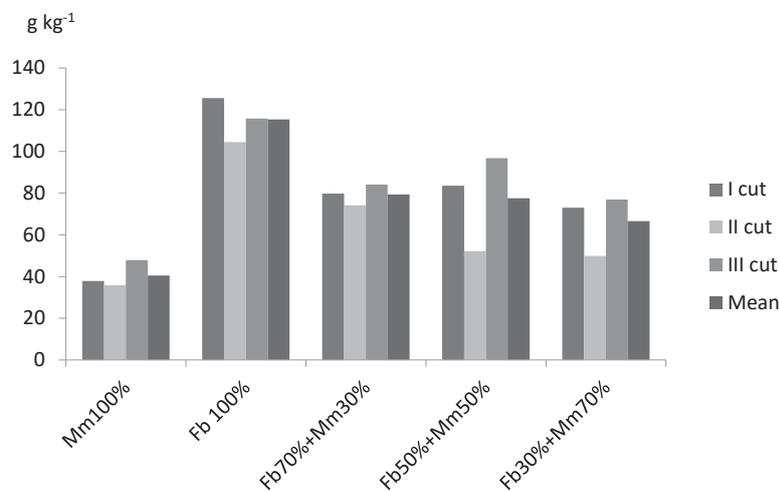


Fig. 3. Content of water-soluble carbohydrates (average for years)

### Carbohydrate-to-protein ratio

The WSC:CP ratio is an indicator of protein availability for animals (Jafari 2012, Simili da Silva et al. 2013, Huhtanen and Broderick 2016). For ruminants, the optimal value of the WSC:CP ratio is 0.8–1.5, and it should not be lower than 0.4. In our study, a desirable WSC:CP ratio was noted only in the biomass of pure-sown *Festulolium* (Fig. 4). In the biomass of pure-sown alfalfa, the analyzed parameter was determined at 0.25 on average, and it was below the minimum value. The evaluated mixtures were characterized by more favorable WSC:CP ratios (0.49–0.61), but the noted values did not meet the nutrient requirements of ruminants. The above can be attributed to the high proportions of alfalfa in the tested mixtures. Similar values of the WSC:CP ratio were reported by Simili da Silva et al. (2013) in legume and grass mixtures. In our study, the WSC:CP ratio was least favorable in the second-cut herbage, which can be attributed to its low carbohydrate content.

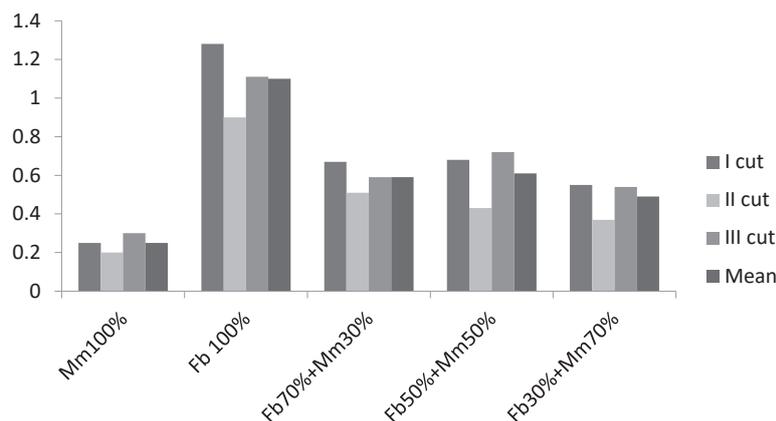


Fig. 4. Carbohydrate-to-protein ratio (average for years)

## Conclusions

The results of the study indicate that *Medicago media* has a high competitive advantage over *Festulolium braunii*. The proportion of alfalfa in the biomass of the tested mixtures increased in successive years of the experiment. In the second and third year of full utilization, alfalfa was the predominant sward species regardless of the proportion of alfalfa seeds in the mixture. During the growing season, the proportion of alfalfa was lowest in the first-cut herbage, and the species developed rapidly in the second and third harvest cycle. The analyzed mixtures were characterized by a higher feed value than pure-sown alfalfa and pure-sown *Festulolium*. The presence of alfalfa in the evaluated mixtures increased and stabilized dry matter and CP yields, whereas the presence of *Festulolium braunii* significantly increased the content of WSC and improved the WSC:CP ratio. Sward mixtures with equal proportions (50%) of both components were characterized by the lowest dry matter and CP yields. The above parameters were higher and similar in the mixtures containing 30% and 70% alfalfa. Carbohydrate yields and the WSC:CP ratio were higher in the mixtures with 30% and 50% alfalfa than in the mixture with 70% alfalfa. The results of this study indicate that *Festulolium* and alfalfa mixtures with a lower proportion of alfalfa (30% and 50%) have a higher feed value.

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