Properties of red clover in monoculture and in mixtures under the influence of nitrogen fertilization

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Abstract

The aim of this research was to determine the production properties of red clover cultivated in monoculture and in mixtures with perennial ryegrass during two years with application of fertilizer in different rates of N. The experimental factors were two mixtures of perennial ryegrass and red clover arranged in different proportions of 70:30 (30CP), 50:50 (50CP) and monoculture of red clover (100CP). N was applied as ammonium nitrate (34% N) in three different levels: 0, 50 and 100 kg N ha^{-1}. After each cut, the botanical composition was studied, and the herbage production parameters and crude protein content were analysed. The share of red clover in the crop had a significant effect on all of the studied properties except for the leaf:stem ratio and leaf surface. Higher proportion of red clover increased yield, plant height, specific leaf area (SLA) and leaf area index (LAI). The height of red clover plants and LAI were higher in monoculture than in mixtures by 12% and 46%, respectively. The crude protein content was 8% higher in mixtures than in pure crop. N fertilization reduced the yield of red clover, and increased other production parameters as well as the crude protein content.

Keywords: red clover, perennial ryegrass, morphology, mixture, yield

Introduction

Red clover is one of the most important perennial forage plants used in livestock nutrition. It has high nutritional value and gives fodder of very good quality suitable for all categories of domestic farm animals. It is a very valuable component of grass-leguminous mixtures. Due to its nitrogen-fixing properties, it reduces the need for the use of N mineral fertilizers, ensures higher content of proteins in the mixture as well as their better utilization, and it also ensures more uniform and higher production. According to Iepema *et al.* (2006), red clover in combination with perennial ryegrass and white clover can realize high yields with low amounts of N. The use of red clover in animal nutrition is limited by its short lifespan of 2-3 years. Factors influencing its persistence are improper management, disease and impaired competition. The cultivation of red clover with high-quality grasses and the application of N mineral fertilizers reduce its persistence (Oram *et al.,* 2014). So, there is a need for breeders to create varieties of red clover that will have greater competitiveness and persistence and enable its successful cultivation in mixtures and widespread use in meadows and pastures.

The aim of this research was to assess the effect of the share of red clover in perennial ryegrass-red clover mixtures and the effect of fertiliser N application rate on the yield of red clover, its morphological properties and the crude protein content.

Material and methods

A field study was conducted at the Institute for Animal Husbandry, Belgrade, Serbia (44° 49' 10" N, 20°, 18', 45" E; altitude 110 m.a.s.l.), in spring 2014. At the site, the mean annual precipitation was 714 mm and mean annual temperature 16.7 °C. The soil is a low carbonate chernozem with pH of 7.26, content of $CaCO_3$ of 3.30 g kg^{-1}, humus 43.50 g kg^{-1}, total N of 1.97 g kg^{-1}, P of 0.40 g kg^{-1} and K of 0.13 g kg^{-1}. The design of the experiment was split-plot with three replications and a plot size of 2×5 m. The red clover (*Trifolium pratense*; local variety K-39) was sown in monoculture (100%) and in mixtures with

perennial ryegrass (*Lolium perenne* cv. Calibra) to provide clover proportions of 30 and 50%. A seeding rate for red clover of 20 kg ha^{-1} was used and 25 kg ha^{-1} for perennial ryegrass. The amount of seed for sowing in mixtures was determined based on the planned sown proportions. N fertilization was supplied as ammonium nitrate with 0, 50 and 100 kg N ha^{-1}. Dry matter yield was recorded for three cuts in 2014 and 2015. For each cut the share of red clover was assessed by separation of species from 1 m^2 subplots. Morphological traits were evaluated on five randomly selected plants per plot. After measuring plant height, leaves were separated from the stem, the leaf area (cm^2) was recorded with ImageJ and the LAI was specified. Thereafter leaf and stem were oven dried, measured and used for calculation of leaf/stem ratio and SLA. Data were statistically processed using analysis of variance including the factors – mixture with different shares of red clover (30CP, 50CP, 100CP) and nitrogen fertilization (0N, 50N, 100N). Statistical analysis was performed in program STATISTICA 8.0 (StatSoft, Inc., 2007).

Results and discussion

The proportion of sown clover and N fertilisation level showed significant impact on DMY of red clover. The red clover DMY increased with increasing sown proportion in the mixtures, and was on average 6.0, 8.5 and 9.5 t ha^{-1} for the 30, 50 and 100% red clover treatment (Table 1). However, the yield of the 30CP mixture represents 63% of the yield of pure red clover and 89% for the 50CP mixtures, indicating higher productivity of red clover plants in mixtures with perennial ryegrass than in pure red clover crop as well as lower interspecific competition. The fertilization significantly reduced the yield of red clover by 24% (50N) and 29% (100N). Plant height is one of the most important traits of yield and persistence of red clover. There was a significant effect of mixture on plant height, which was highest in the pure red clover crop (69.80 cm) and lowest in red clover plants in the 30CP mixture (57.5 cm). According to Hoekstra *et al.* (2018), plant height is an indicator of persistence and competitive ability of plants. In monoculture there is high competition between plants for light, so the plant height is higher than in the mixtures. Measurement of leaf area is in direct connection with competition for light. The SLA value was the highest in plants of a pure crop and was lowest for plants in the 30CP mixture. The difference was significant only between plants in the 30CP mixture and plants in pure crop or in 50CP mixture. N fertilization increased values of SLA. The LAI was the highest in pure crop (4.11 cm^2) and lowest in the 30CP mixture (2.42 cm^2). N fertilization increased the LAI value from 2.74 to 3.57 cm^2. Knops

Table 1. Effect of mixture and N fertilization on red clover dry matter yield (DMY), plant height, specific leaf area (SLA), leaf area index (LAI), leaf/stem ratio, leaf area (LA), and content of crude protein (average over two years ± standard error of means).[1]

	DMY (t ha^{-1})	Plant height (cm)	SLA (cm^2 g DM^{-1})	LAI (m^2 m^2)	Leaf/stem ratio (g g^{-1})	LA (cm^2)	CP (g kg^{-1})
Effect of mixture							
30CP	6.0±0.3^b	57.5±1.6^c	265.82±7.81^b	2.42±0.16^c	0.39±0.01	141.27±5.97	16.27±0.24^a
50CP	8.5±0.7^a	62.4±1.8^b	287.47±9.49^a	3.35±0.38^b	0.40±0.02	159.35±5.91	16.61±0.13^a
100CP	9.5±0.9^a	67.0±0.9^a	291.42±6.27^a	4.11±0.43^a	0.38±0.01	154.24±9.78	15.21±0.29^b
Level of significance							
mixture	**	**	**	**	ns	ns	**
Effect of N fertilization							
0	9.7±0.9^a	62.8±2.6	287.02±10.73^a	2.74±0.10^b	0.36±0.01^c	137.89±5.51^b	16.03±0.39
50	7.4±0.7^b	61.1±1.3	264.65±4.73^b	3.57±0.47^a	0.39±0.01^b	165.98±8.94^a	15.71±0.25
100	6.8±0.5^b	63.0±1.8	293.06±6.65^a	3.56±0.50^a	0.41±0.01^a	150.98±7.00^b	16.35±0.23
Level of significance							
N fertilization	**	ns	**	*	**	**	ns
Interaction	ns	ns	**	**	ns	**	**

[1] ns = not significant, * significant at the 0.05 level, ** significant at the 0.01 level; Values with different letters are significantly different.

and Reinhart (2000) also confirm that nitrogen fertilization increases values of SLA and LAI by 82 and 202%, respectively. Fertilization increased leaf area with 9 and 20%. The N treatments had significant effect on leaf/stem ratio. It ranged from 0.36 g g^{-1} in 0N to 0.41 g g^{-1} in 100 N treatment. The CP content was higher in mixtures than in pure crop by 8%.

Proportion of red clover showed statistical differences between mixtures that were in line with the sowing density. N fertilization also had significant impact on red clover proportion (Figure 1). There were significant differences between treatment with 100 kg ha^{-1} and the other two treatments.

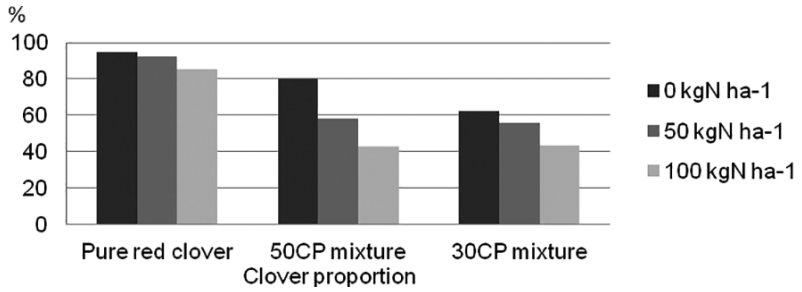

Figure 1. Mean red clover content (%) for three mixtures and three levels of N fertilization.

Conclusion

This study shows that perennial ryegrass is compatible for growing in mixtures with red clover. In pure red clover crop, there is greater competition for light than in red clover-perennial ryegrass mixtures. Red clover plants are more productive in the mixtures than in the pure crop. The N fertilization decreases the DMY of red clover but increases the parameters of leaf area and leaf/stem ratio. The content of CP in red clover increases when cultivated in mixture with perennial ryegrass.

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