

## ESTIMATION OF RED CLOVER (*Trifolium pratense* L.) FORAGE QUALITY PARAMETERS DEPENDING ON THE STAGE OF GROWTH

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Original scientific paper

**Abstract:** The objective of this study was to investigate the changes that take place in nutrient values of red clover (*Trifolium pratense* L.) cv K-27 at different stages of growth in the second and the third cut. The samples from different stages of growth: mid-bud stage, around 60% flowering and full flowering were investigated for crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), digestible dry matter (DDM), dry matter intake (DMI), relative feed value (RFV), total digestible nutrients (TDN), net energy-lactation (NEL), net energy-maintenance (NEm) and net energy-gain (NEg) content. Higher content of crude protein was found in the third cut (262.1 g kg<sup>-1</sup> of DM) than in the second cut (260.8 g kg<sup>-1</sup> of DM) in the first stage of development. TDN, DDM, DMI, RFV, NEL, NEm and NEg were calculated according to the appropriate equations adapted from common formulas for forages. The stage of plant development has a significant influence on the chemical composition and the relative feed value of red clover. The achieved results show that adequate maturity stage for cutting might be at mid-bloom stage, when crude protein content, NEL, NEm and NEg content are high.

**Key words:** red clover, forage quality, nutritive value, stage of growth

### Introduction

Red clover is a legume well adapted to poorly drained soils and more tolerant to lower soil pH and fertility than alfalfa. The yield potential of red clover is excellent and some red clover varieties can have higher fodder yields than alfalfa. Red clover is considered a short-lived legume, but new modern varieties are productive for three full seasons. Red clover is also of a very good quality as to its nutritive value and ensiling (*Hoffman and Broderick, 2001*).

The possibility to accurately predict the nutritive value of forage crops is a prerequisite for designing rations and directing forage crops breeding (*Kruse et al.*,

2008). Nutrient deficiencies in low quality roughages affect microbial growth and fermentation in the rumen and result in an overall low animal productivity. Quality forage promotes good health and better performance (Harris, 1992). Therefore, choosing adequate stage of growth of forages with good quality is very important for animal health as well as high milk and meat production.

Crude protein (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF), relative feed value (RFV) and total digestible nutrients (TDN) are important criteria for determining hay quality (Yolcu et al., 2008) in animal feeding. Therefore, CP, ADF, NDF, DDM, DMI, RFV, TDN and NEI (Lithourgidis et al., 2006) were examined as quality criteria in many studies.

This study was conducted in order to investigate the changes that take place in nutrient values (CP, NDF, ADF, DDM, DMI, RFV, TDN, NEI, NEm and NEg) of red clover, cv K-27 at different stages of growth in the second and the third cut.

## Materials and Methods

The experiment was carried out in the experimental field of Institute for forage crops in Kruševac in 2008 as a factorial trials, by randomized block system in three replicates. Three stages of growth of red clover (*Trifolium pratense* L.) cv K-27 were examined in the second and third cut. The first stage was cut after 22 days of the first cut, at mid-bud stage, another one after 29 days of the first cut, at around 60% flowering, and the third one in full flowering after 36 days of the first cut.

Crude protein (CP) were computed indirectly from the amount of total nitrogen, measured by the Kjeldahl method modified by Bremner, multiplied by factor 6,25. Acid detergent fiber (ADF) and neutral detergent fiber (NDF) analyses were determined by Van Soest (1963). Digestible dry matter (DDM), dry matter intake (DMI), relative feed value (RFV), total digestible nutrients (TDN), net energy-lactation (NEI), net energy-maintenance (NEm) and net energy-gain (NEg) were calculated according to the equations adapted from common formulas for forages (Schroeder, 1994).

Data were processed by the analysis of variance in a randomized block design. Effects were considered different based on significant ( $P < 0.01$ ) F ratio. The significance of differences between arithmetic means was tested by LSD test.

## Results and Discussion

The results of this investigation indicated that the crude protein content of red clover declined with advancing maturity in the second and third cut (Table 1). Crude protein content of plant tissue reflected the relative proportions of each tissue type, and was considered with other findings for red clover (*Hoffman et al., 1993*).

The NDF and ADF concentrations of forages provide useful information about quality. The results of this study show that contents of cell wall increased in both investigated cuts (Table 1). During maturation NDF content increased from 335.9 to 422.9 g kg<sup>-1</sup> of DM in the second and from 325.9 to 399.5 g kg<sup>-1</sup> of DM in the third cut ( $P < 0.01$ ). This results is in agreement with the report of *Kamalak et al. (2005)* who found that NDF and ADF content increased.

Forage quality declines with advancing maturity. Significant differences were determined among forage stages of growth in terms of DDM, DMI and RFV for animal feeding (Table 1). The highest DDM ratio was found at the first stage of plant development (70,1% in the second cut, and 75,7% in the third cut). The lowest DDM ratio was found at the third stage of plant development in both cuts. Maturity at harvest also influences forage consumption by animals. This is because NDF is more difficult to digest than the non-fiber components of forage. The RFV of red clover hay harvested at different maturity stages ranged from 194.3 to 140.9 in the second cut and from 216.0 to 153.7 in the third cut. This values are higher than values reported by *Stallings (2005)*. The decrease in DDM, DMI and RFV are possibly associated with increased NDF and ADF contents (*Wilson et al., 1991*) and increased lignification and decreased leaf:stem ratio (*Hides et al., 1983*) as the plant matures.

**Table1.** Forage quality of red clover (*Trifolium pratense* L.) cv K-27 at different maturity stages in the second and third cut

	CP g kg <sup>-1</sup>	NDF g kg <sup>-1</sup>	ADF g kg <sup>-1</sup>	DDM %	DMI %	RFV %
			II cut			
I stage	260.8 a	335.9 b	240.7 c	70.1 a	3.5 a	194.3 a
II stage	219.6 b	364.4 b	286.1 b	66.6 b	3.3 a	170.1 a
IIIstage	173.8 c	422.9 a	318.8 a	64.1 c	2.8 b	140.9 b
			III cut			
I stage	262.1 a	325.9 c	169.9 c	75.7 a	3.7 a	216.0 a
II stage	180.2 b	382.5 b	239.9 b	70.2 b	3.1 b	170.8 b
IIIstage	159.3 c	399.5 a	294.2 a	65.9 c	3.0 c	153.7 c

Different letters within a column denote significantly different means ( $P < 0.01$ )

TDN refers to the nutrients that can be utilized and available for livestock (*Lithourgidis et al., 2006*). Differences ( $P < 0.01$ ) in terms of TDN contents were determined among stages of growth in both cuts. Total digestible nutrients contents of red clover ranged from 68.7 to 59.7 % in the second cut, and from 76.8 to 62.5% in the third cut (Table 2). The highest total digestible nutrients contents were determined at the first stage of growth in the second and the third cut. It is important to provide cattle with adequate amounts of digestible energy for optimal animal performance. While many factors affect forage digestibility and ultimately TDN, the primary factor producers can control is forage maturity.

The energy concentration, expressed for dairy cows in MJ NEI kg<sup>-1</sup> DM is very important. The highest possible level of energy concentration is a prerequisite to feed highly performing cows successfully. But possibilities to increase cows energy intake by increasing the energy concentration are limited. The highest NEI content in the second cut was recorded at the first stage of development (6.54 MJ kg<sup>-1</sup> of DM), and the lowest was in the third stage of growth (5.62 MJ kg<sup>-1</sup> of DM). In the third cut NEI content ranged from 7.38 to 5.92 MJ kg<sup>-1</sup> of DM. The enhancement of NEI value is very important, as *Hrabe et al. (2003)* reported that an increase of NEI of clover-grass silage by 0.5 MJ kg<sup>-1</sup> of DM reduces the need for complementary feed by 2.5 kg per day.

The energy value of feed in our trial expressed by NEm values varied from 7.12 to 6.03 MJ kg<sup>-1</sup> of DM in the second and from 8.10 to 6.37 MJ kg<sup>-1</sup> of DM in the third cut. The differences between stages of growth were significant ( $P < 0.01$ ).

Significant differences between stages of growth were recorded for NEg in this study ( $P < 0.01$ ). The highest content of NEg was at the first stage of plant development (4.11 MJ kg<sup>-1</sup> of DM in the second cut, and 5.09 MJ kg<sup>-1</sup> of DM in the third cut). Higher content of NEg was determined in the third cut than in the second cut (Table 2).

**Table 2. Nutritive value of red clover (*Trifolium pratense* L.) cv K-27 at different maturity stages in the second and third cut**

	TDN %	NEI MJ kg <sup>-1</sup>	NEm MJ kg <sup>-1</sup>	NEg MJ kg <sup>-1</sup>
		II cut		
I stage	68.7 a	6.54 a	7.12 a	4.11 a
II stage	63.5 b	6.01 b	6.48 b	3.47 b
III stage	59.7 c	5.62 c	6.03 c	3.02 c
		III cut		
I stage	76.8 a	7.38 a	8.10 a	5.09 a
II stage	68.7 b	6.55 b	7.13 b	4.12 b
III stage	62.5 c	5.91 c	6.37 c	3.36 c

Different letters within a column denote significantly different means ( $P < 0.01$ )

Energy is often referred to digestible energy, net energy for maintenance (NEm), net energy for gain (NEg), net energy for lactation (NEl), and total digestible nutrients (TDN). When digestible energy becomes limiting in beef cattle diets, intake and animal performance can suffer. Signs of energy deficiency include lowered appetite, weight loss, poor growth, depressed reproductive performance, and reduced milk production. Providing adequate digestible energy in beef cattle diets is important for animal health and productivity as well as ranch profitability.

## Conclusion

The choice of harvesting moment is important insuring the forage crops quality and quantity. Relative feed value might be a relevant indicator supporting the decisions of cutting management, especially when red clover is a component of a legume-grass mixture. This experiment showed a decreasing RFV of the red clover, cv K-27 with plant development. In the second cut the grade of red clover hay at the first and the second stage of development was prime, but at the third maturity stage it was premium - 1. In the third cut, at all three maturity stages the grade of red clover hay was premium. Significant differences were determined in terms of CP, NDF, ADF, DDM, DMI, TDN, NEl, NEm and NEg. From our results we may conclude that adequate maturity stage for cutting might be mid-bloom stage, when crude protein content, NEl, NEm and NEg content are high.

## Acknowledgment

Research was financed by the Ministry of Education and Science, Republic of Serbia TR 31057.

## Procena parametara kvaliteta crvene deteline (*Trifolium pratense* L.) u zavisnosti od faze razvića

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## Rezime

Cilj ovog rada je bio ispitivanje promena hranljive vrednosti crvene deteline (*Trifolium pratense* L.) sorte K-27 u različitim fazama razvića u drugom i trećem otkosu. Uzorci iz različitih faza razvića: sredina butonizacije, oko 60% cvetanja i puno cvetanje su ispitivani na sadržaj sirovih proteina, neutralne

deterdžent celuloze, kisele deterdžent celuloze, svarljive suve materije, konzumiranja suve materije, relativne hranljive vrednosti, ukupnih svarljivih hranljivih materija, neto energije laktacije, neto energije za održavanje i neto energije prirasta. Veći sadržaj sirovih proteina ( $262,1 \text{ g kg}^{-1} \text{ SM}$ ) je ustanovljen u trećem, nego u drugom otkosu (od  $260,8 \text{ g kg}^{-1} \text{ SM}$ ) u prvoj fazi razvića. Sadržaj ukupnih svarljivih hranljivih materija, svarljivosti suve materije, konzumiranja suve materije, relativne hranljive vrednosti, neto energije laktacije, neto energije za održavanje i neto energije prirasta su izračunate prema odgovarajućim formulama za kabasta hraniva. Ustanovljeno je da faza razvića ima značajan uticaj na hemijski sastav i relativnu hranljivu vrednost crvene deteline. Dobijeni rezultati pokazuju da adekvatna faza razvića za košenje crvene deteline može biti sredina cvetanja kada je sadržaj sirovih proteina, NEI, NEM i NEg visok.

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Received 30 June 2011; accepted for publication 15 August 2011