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Determination of Relative Feed Value, Energy and Nutrient Contents of Annual Ryegrass (*Lolium multiflorum* Lam.)**

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ABSTRACT

This study was carried out to determine the effect of the number of harvests on the feed value of annual ryegrass (*Lolium multiflorum* Lam.). The annual ryegrass grown in a private family farm in Hürrempalangası Village, Erzincan was used as the research material. The ryegrass was harvested twice in the experimental period. The plants harvested in the flowering period were baled, and samples were collected using a drill and probe. The samples were analyzed according to the harvest time in terms of chemical content, metabolizable energy (ME), net energy lactation (NEL), total digestible nutrients (TDN), and relative feed value (RFV). The obtained data were subjected to analysis of variance, and the differences between the means were determined by T comparison test. The difference between the plants harvested in different periods was significant in terms of nutrient contents ($p<0.05$). The NEL, ME, and TDN contents were calculated using the nutrient composition of the ryegrass. The difference between the harvests in terms of TDN and RFV was found significant ($p<0.01$). The means related to CP, RFV, NEL, ME, and TDN contents of the annual ryegrass were found to be 12.74% and 18.72%, 111. 56 and 98.79, 1593.9 and 1589.4 Kcal/kg DM, 2352.2 and 2345.5 Kcal/kg DM, and 59.19% and 57.02% in the first and second harvest, respectively. In conclusion, chemical composition and RFV parameters of the annual ryegrass, it can be asserted that the annual ryegrass has a high potential as a forage source and may be used in ruminant rations.

Tek Yıllık Çim (*Lolium multiflorum* Lam.) Yem Bitkisinin Nispi Yem Değeri, Enerji ve Besin İçeriğinin Belirlenmesi

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ÖZ

Bu çalışma, hasat sayısının tek yıllık çim (*Lolium multiflorum* Lam.) yem değerine etkisini belirlemek amacıyla yapılmıştır. Araştırma materyali olarak Erzincan ili Hürrempalangası Köyü'ndeki özel bir aile çiftliğinde yetiştirilen tek yıllık çim kullanılmıştır. Tek yıllık çim deneme döneminde iki kez hasat edilmiştir. Bitkiler çiçeklenme döneminde hasat edildikten ve balyalandıktan sonra matkap ve sonda kullanılarak numuneler alınmıştır. Numunelerin hasat zamanına göre kimyasal içerikleri ile metabolik enerji (ME), net enerji laktasyon (NE_L), toplam sindirilebilir besin maddeleri (TSBM) ve nispi yem değeri (NYD) parametreleri belirlenmiştir. Elde edilen veriler varyans analizine tabi tutulmuş ve ortalamalar arasındaki farklar T testi ile tespit edilmiştir. Farklı dönemlerde hasat edilen bitkiler besin madde içerikleri açısından meydana gelen farklılıklar istatistiksel olarak önemli bulunmuştur ($P<0.05$). NE_L, ME ve TSBM içerikleri, çim otunun besin madde bileşimleri kullanılarak hesaplanmıştır. TSBM ve NYD açısından hasatlar arasındaki fark önemli bulunmuştur ($P<0.01$). Tek yıllık çimin birinci ve ikinci hasatta ortalama HP, NDF, NEL, ME ve TSBM içerikleri sırasıyla; %12,74 ve %18,72, 111,56 ve 98,79, 1593,9 ve 1589,4 Kcal/kg DM, 2352,2 ve 2345,5 Kcal/kg DM ve %59,19 ve %57,02 olmuştur. Sonuç olarak, kimyasal bileşimi ve NYD parametreleri dikkate alındığında yüksek bir yem kaynağı potansiyeline sahip olan tek yıllık çimin kaba yem kaynağı olarak ruminant rasyonlarında kullanılabileceği söylenebilir.

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1. Introduction

Forages account for a significant part of the feeds of the animals which are the main source of protein in the nutrition of humans. Forages are produced from the natural or artificial meadows and the forage plants.

The economic value of the culture of forage plants reveals itself especially in the animal production materials. The products produced from the animals fed with the forages with high nutritive value have a high quality, and they also have a great importance for the human life and the economy of a country.

Forage production is necessary and very important for feeding ruminants. Diets for productive ruminants usually comprise two different types of feed: forages and concentrates. Forages partly meet the nutrient needs of the animals and protect them against certain diseases. Necessary importance should be given to the cultivation of forage plants for an efficient and profitable animal production, and the cultivated areas should be extended. One of the easiest ways for meeting the feed needs of the animals is the agriculture of forage plants across the country and in Erzincan. Therefore, it is of great significance for the forage production facilities to meet the needs of the animal production facilities in Erzincan (Gürsoy, 2013). Production of forage crops is too low and insufficient to meet the needs of ruminants. The cultivation area of forage plants has a size of 207.178 decares in the center of Erzincan and 556.638 decares in its districts (TUİK, 2017a). Erzincan has an important potential in the animal sector with 93,076 big ruminants and 417,074 small ruminants (TUİK, 2017b).

There are many problems waiting for a solution in the ruminant livestock of Turkey today. The most urgent and important one is the inability to provide sufficient forages for ruminant feeding. It is highly necessary that good quality feed and mixed feed resources be used in the rations and the environmental factors be improved to be able to get efficiency from the ruminants in line with their genetic capacities (Baylan, 2007). For these reasons, the field agriculture should include new quality roughages.

Annual ryegrass (*Lolium multiflorum* Lam.) is one of the important species that have recently been cultivated for forage (Özdemir et al., 2019). Annual species are taller (40-100 cm) and more productive than the perennial ones. Ryegrass, a forage plant with a short life, has a good grass quality.

It is unique in that it can grow rapidly and be re-harvested. It adapts to all kinds of soils, leaves the intensive organic substances to the soil, and is preferred in alternate plantation. It can grow well in both heavy and sandy soils. However, it shows its best growth in the soils very well processed and supported with animal fertilizers. It is widely used in the production of silage and dry grass. In addition, it is so common as the 2nd product after cereals. It has very high protein and energy values (Aktar et al., 2021). It is sugary and easy-digestible; therefore, animals eat it pleasurably and hungrily. It grows in fall and spring in the regions where it snows and hibernates in winter (Soya et al., 1997; Lale, 2020). In Turkey, the production of this plant species has recently become the new favorite of the producers with the agricultural incentives provided by the state in order to increase the production of feed plants (Baytekin et al. 2009). Katiç et al. (2009) examined the differences between 4 different alfalfa varieties in terms of crude protein, ADF, and NDF in Serbia in 2005 and 2006. They reported that there were significant differences between the harvests, but not between the varieties. It has been reported in the literature that the metabolizable energy (ME) content of Italian grass varies between 7.15 and 11.8 MJ kg-1 DM and this range changes depending on the factors such as season, soil type, and planting time (Colf, 2010). In their study on three different forms of Caramba plant (fresh, silage, and hay), Özelçam et al. (2015) reported that the HP content was between 12.83% and 8.91%, the NDF content between 57.41% and 63.70%, the ADF content between 35.32% and 43.29%, and the ADL content between 5.55% and 8.86%. They also reported that the DM and organic matter (OM) digestibility of three different forms were within the ranges of 73.01-79.58% and 74.44-81.37%, respectively, and there was no significant difference between three different forms in terms of these two parameters. As a result of the study, they asserted that the Caramba could be used as a source of roughage in the forms of fresh, silage, and hay in ruminant feeding.

In their research, McCormick et al. (2008) mowed forkgrass, bermuda grass, ryegrass, and sorghum grass and fed them to dairy cows and calves. They reported that the best yield increase was achieved with ryegrass. However, it lagged behind others in terms of milk fat. In order to grow quality plants, rations should be prepared in a balanced way considering the feed values depending on the harvest times. Thus, the desired level can be obtained in animal products, and metabolic diseases caused by unbalanced rations can be prevented.

This study was conducted to determine the chemical composition, relative feed value, net energy lactation, metabolizable energy parameters, and total digestible nutrients (*Lolium multiflorum* Lam.) of annual ryegrass depending on the number of harvests.

2. Material and Methods

Annual ryegrass (*Lolium multiflorum* Lam.), the feed material of the study, was harvested in a 5-decare agricultural field in Hurrempalangası Village (39 475832N, 39 215618E), Erzincan. The seeds of the annual ryegrass were sown using a sowing machine with a seed density of 5 kg seeds per decare at a row spacing of 15 cm with a depth of 1.5-2 cm, and then a roller passed over them. 25 kg of compound fertilizer containing nitrogen, phosphorus, and potassium in equal proportions (15:15:15) was applied to the base. After the first harvest, nitrate fertilizer was applied at 15 kg/da. The seeds were sown on October 28, 2016 and their growth was monitored during the study. When their height reached 70-80 cm in the flowering period, the annual ryegrass plants were harvested two times using a drummed mower on June 23, 2017 and August 03, 2017. Surface irrigation was performed 2 weeks after the sowing and after every harvest. After each harvest, the plants were baled (rectangular) under suitable conditions. 0.5 kg feed sample was collected from 10 roughage bales using a drill and probe, and then the samples were put in the cloth bags tagged with the harvest date and sample number. The feed samples collected for each harvest were brought to the Feed Analysis Laboratory, Department of Field Crops, Faculty of Agriculture, Atatürk University. They were ground to pass through a 1 mm sieve, put in transparent plastic bags, tagged, and left for further analysis (Canbolat and Karaman, 2009; Gürsoy and Macit, 2017; Ayaşan et al., 2020a).

In the present study, the feed samples were burnt by keeping in an ash furnace at 550 °C for 4 hours after drying in an oven at 105 °C. Then, they were cooled down, and their crude ash (CA) contents were determined. Firstly, the nitrogen (N) contents of the feed samples were determined using Kjeldahl method, and then, the determined nitrogen values were multiplied by 6.25 and the crude protein (CP) contents were calculated. Ether extract (EE) analysis was conducted in line with the method specified in AOAC (1990). Crude fiber (CF), Neutral Detergent Fiber (NDF), and acid detergent fiber (ADF) contents were determined using an ANKOM 200 Fiber Analyzer in line with the method reported by Van Soest et al. (1991). Chemical analyses (DM, CP, EE, CA) and the analyses of cell wall structural elements (NDF and ADF) of the feed samples were carried out in five replications. RFV, ME, and NE_L values of the feeds were calculated using the following equations. The total digestible nutrient (TDN) content was calculated according to the methods of Moore and Undersander (2002).

$$DMD\% = 88.9 - (0.779 \times ADF\ \%),\ DMI\ \% = 120 / NDF,$$

$$RFV = DMD\ \% \times DMI\% \times 0.775 \text{ (Van Dyke and Anderson, 2000)},$$

$$NE_L,\ ME\ (Mcal/Kg\ DM) = 0.82\ DE \text{ and } NE_L\ (Mcal/Kg\ DM) = 0.5557\ DE - 0.12$$

$$ME = 3303 + 96.08\ EE\% - 37.05\ CF\ \% - 32.27\ CA\ \% \text{ (Abaş et al., 2005)}.$$

Statistical analyses

The obtained data were subjected to analysis of variance using SPSS 12.0 package program and the differences between the means were determined by T comparison test (Petrie ve Watson, 1999).

3. Results and Discussion

3.1. Chemical composition of the annual ryegrass

The nutrient composition of the annual ryegrass was determined depending on the number of harvests. Table 1 shows the DM, CA, EE, CP, CF, NDF, and ADF contents of the annual ryegrass.

Table 1. Chemical composition of the annual ryegrass by the number of harvests, % (in DM).

Harvest Number	Dry Matter	Crude Ash	Ether Extract	Crude Protein	ADF	NDF	Crude Fiber	NFE	Hemicellulose
First harvest	93.43	17.64	4.91	12.74	30.86	54.05	23.04	35.08	23.09
Second harvest	92.69	13.93	4.49	18.72	33.75	58.96	25.12	30.53	25.21
SEM	1.05	0.14	0.16	0.32	0.25	0.41	0.16	1.18	0.27
P	NS	**	*	* *	**	**	**	*	**

NDF: Neutral Detergent Fiber, ADF: Acid Detergent Fiber, NFE: N-free Extract, SEM: Standard Error of Mean, *: P<0.05, **: P<0.01, NS: Not Significant

The DM content of the annual ryegrass was found to be 93.43% in the first harvest and 92.69% in the second harvest. No significant difference was found to exist between the first and second harvest in terms of DM content (P>0.05). The DM contents found in the present study were higher than those reported by Karakurt and Ekiz (1996), Göktepe (2015), and Özelçam et al. (2015), but similar to those reported by Gürsoy and Macit (2017).

The CA content of the annual ryegrass was found to be 17.64% in the first harvest and 13.93% in the second harvest. The difference between the first and second harvest was significant in terms of CA content (P<0.01). This result was similar to those reported in the literature (Akgün et al., 2008; Barchiesi-Ferrari et al., 2011; Kuşvuran, 2011; Göktepe, 2015; Gürsoy and Macit, 2017).

The EE content of the annual ryegrass was found to be 4.91% in the first harvest and 4.49% in the second harvest. The difference between the first and second harvest was significant in terms of EE content (P<0.05). The EE contents found in the present study were higher than those reported in the literature (Göktepe, 2015; Özelçam et al., 2015; Gürsoy and Macit, 2017). We are of the opinion that this was caused by the differences between the present study and other studies in terms of soil structure, climate, and plant variety.

The CP content was found to be 12.74% in the first harvest and 18.72% in the second harvest. There was a significant difference between the first and second harvest in terms of CP content (P<0.01). The CP contents found in the present study were similar to those reported by Serin et al. (1996), Barchiesi-Ferrari et al. (2011), Kuşvuran (2011), Kuşvuran and Tansı (2011), Göktepe (2015), and Kara (2016); lower than those reported by Dovel et al. (1999), Meeske et al. (2009), and Teutsch and Smith (2001); and higher than those reported by Akgün et al. (2008), Çarpıcı and Çelik (2014), Kuşvuran et al. (2014), Şimşek (2015), Kavut and Geren (2018), Özdemir et al. (2019), and Türk et al. (2019). These differences may be due to the differences in environmental conditions and fertilizers.

As for the cell wall structural elements, the ADF content of the annual ryegrass samples was found as 30.96% and 33.75%, NDF content as 54.5% and 58.96%, CF content as 23.04% and 25.12%, NFE content as 35.08% and 30.53%, and hemicellulose content as 23.09% and 25.21% in the first and second harvest, respectively. While a decrease was observed in the DM, CA, EE, and NFE contents in the second harvest compared to the first harvest, an increase was observed in the CP, ADF, NDF, and hemicellulose contents. Kavut and Geren (2018) reported that ADF and NDF contents increased due to delaying harvest dates.

The difference between the first and second harvest of the annual ryegrass was significant in terms of ADF content (P<0.01). The ADF contents found in the present study were in agreement with those reported by Dovel et al. (1999), Meeske et al. (2009), Kara (2016), and Gürsoy and Macit (2017); lower than those reported by Barchiesi-Ferrari et al. (2011), Çarpıcı and Çelik (2014), Kuşvuran et al. (2014), Özelçam et al. (2015), and Gemalmaz and Bilal (2016); and higher than those reported by Teutsch and Smith (2001).

It was found that the NDF content of the annual ryegrass was 58.96% in the first harvest and 54.05% in the second harvest. There was a significant difference between the harvests in terms of NDF content (P<0.01). The ADF contents found in the present study were similar to those reported by Kuşvuran et al. (2014) and Gürsoy and Macit (2017); lower than those reported by Dovel et al. (1999), Teutsch and Smith (2001), Meeske et al. (2009), Özelçam et al. (2015), and Kara (2016); and higher than those reported by Barchiesi-Ferrari et al. (2011), Çarpıcı and Çelik (2014), and Gemalmaz and Bilal (2016). Şimşek (2015) examined the yield and quality characteristics of Italian ryegrass (*Lolium multiflorum* Lam.) under the conditions of Kırşehir and reported that the ADF and NDF contents were 39.66% and 59.67%, respectively.

Göktepe (2015) examined the annual ryegrass grown in the Black Sea climate at 5 harvest times in one season and reported that the mean contents of DM, CP, CS, EE, ADF, NDF, ADL, and ME were 91.52%, 17.39, 28.30, 2.28, 30.26, 49.95, 3.96, and 10.32 MJ/kg, respectively. Kavut and Geren (2018) investigated the yield and quality characteristics of different mixtures of ryegrass and hairy vetch at different harvest times under the conditions of Izmir and reported that the ADF and NDF contents increased with the delay in harvest time, and the CP content increased as the legume ratio increased in the mixture. Özdemir et al. (2019) investigated the effects of different nitrogen doses on the yield and quality of ryegrass under the conditions of Bursa and reported that nitrogen fertilization in irrigated conditions increased the yield and quality of the ryegrass. The difference between the present study and other studies in terms of chemical contents may be due to the differences in harvest times, vegetation periods, climates, soils, and plant diversities.

3.2. Digestible dry matter, dry matter intake, and relative feed value of the annual ryegrass

Relative feed value is an important tool for marketing the forages and determining their quality (Rohweder et al., 1978; Moore, 1994). Relative Feed Value is based on the assumption that alfalfa hay in full bloom (41% ADF and 53% NDF) has an RFV of 100. As the RFV falls below 100, the quality of the feed decreases, and vice versa (Redfearn et al., 2006). RFVs below 75 refer to the 5th quality, 75-86 to the 4th quality, 87-102 to the 3rd quality, 103-124 to the 2nd quality, 125-150 to the 1st quality (Rohweder et al., 1978).

Table 2 shows the DDM, DMI, and RFVs of the annual ryegrass in the first and second harvest.

Table 2. The digestible dry matter, dry matter intake, and relative feed value of the annual ryegrass.

Harvest number	DMI (%)	DDM (%)	RFV
First harvest	2.22	64.79	111.56
Second harvest	2.04	62.62	98.79
SEM	0.02	0.19	1.09
P	**	**	**

DMI: Dry Matter Intake, DDM: Digestible Dry Matter, RFV: Relative Feed Value, SEM: Standard Error of Mean, **: P<0.01

DMD, DMI, and RFV of the feed samples were calculated using the cell wall structural elements. It was found that there were significant differences between the first and second harvest in terms of DMI, DDM, and RFV (P<0.01). The DDM, DMI, and RFV of the annual ryegrass were found to be lower in the second harvest than in the first harvest.

The DMI of the annual ryegrass was calculated as 2.22% in the first harvest and 2.04% in the second harvest. The DDM of the annual ryegrass was found to be 64.79% in the first harvest and 62.62% in the second harvest. In a previous study, the DDM and DMI of the caramba variety of ryegrass harvested in five different periods were reported as 65.32% and 2.42% on average, respectively (Göktepe, 2015). Kara (2016) reported the DDM as 64.18% and Çetin (2017) as 61.6%. Lale (2020) examined the yield and quality characteristics of Italian ryegrass varieties grown under the ecological conditions of Bingöl and reported that the average DDM and DMI were 58.32% and 2.02%, respectively. The differences between similar studies are due to the different ADF and NDF values.

RFV of the annual ryegrass was calculated as 111.56 in the first harvest and 98.79 in the second harvest. These values were in agreement with those reported by Çolak (2015) and Gürsoy and Macit (2017); lower than those reported by Kuşvuran et al. (2014); and higher than those reported by Dovel et al. (1999), Göktepe (2015), Kara (2016), and Lale (2020). In the present study, when the RFV (111,56) calculated for the first harvest of the annual ryegrass was compared with the RFV of 100, the value accepted as the normal for *Medicago sativa;* it can be classified as high quality. On the other hand, the RFV (98.79) found for the second harvest was similar to the normal RFV for *Medicago sativa*. Çolak (2015) reported that 8 kg/da of nitrogen dose should be used in the ryegrass cultivation under the conditions of Ankara in order to have a high yield and RFV. Lale (2020) investigated the feed value of 6 varieties of ryegrass under the ecological conditions of Bingöl and reported that the highest RFV was found in the variety Vallinert. The differences between similar studies may be due to the differences in the cultivars used and the ecologies in which the studies were conducted. Moreover, growing conditions and genotypic differences create differences in ADF, NDF, and RFV values.

3.3. Net energy lactation, metabolizable energy, and total digestible nutrients of the annual ryegrass

Net energy lactation (NE_L) is an energy evaluation system that is calculated using the metabolizable energy of the forages and the regression equations between the metabolizable energy value and the digestible nutrients, especially based on the studies on dairy cows. Because forages can be digested by ruminants at a certain level, their ME contents are very important. The energy lost with the methane gas is taken into consideration in calculating ME in ruminants (Kutlu, 2008). TDN, which is related to ADF concentration, refers to the nutrients available for livestock (Yavuz et al., 2017).

Table 3 shows the net energy lactation, metabolizable energy, and total digestible nutrients of the annual ryegrass.

Table 3. Net Energy Lactation (Kcal/Kg DM), Metabolizable Energy (Kcal/Kg DM), and Total Digestible Nutrients of the Annual Ryegrass.

Harvest number	NE_L	ME	TDN
First harvest	1593.9	2352.2	59.19
Second harvest	1589.4	2345.5	57.02
SEM	10.63	15.68	0.19
P	NS	NS	*

NE_L: Net Energy Lactation, ME: Metabolizable Energy, TDN: Total Digestible Nutrients, SEM: Standard Error of Mean, *: $P<0.05$, NS: Not Significant

No difference was found to exist between the first and second harvest of the annual ryegrass in terms of NE_L and ME ($P>0.05$), but TDN was affected by the harvest number ($P<0.01$). TDN values of the annual ryegrass were calculated as 59.19% in the first harvest and 57.02% in the second harvest. Yavuz et al. (2017) reported the TDN of annual ryegrass lines as 57.97% on average, which is similar to the result found in the present study.

NE_L and ME values found in the current study were lower than those reported by Özelçam et al. (2015) and Gürsoy and Macit (2017). This difference may be due to the differences between the studies in terms of harvest time, vegetation period, climate, soil, and plant species (Eseceli et al., 2020; Ayaşan et al., 2020b).

4. Conclusion

As a result, it was observed that the first harvest of the ryegrass yielded higher values than the second harvest in all parameters except for the cell wall elements. When the chemical composition, RFV, NE_L, ME, and TDN values of the annual ryegrass are evaluated together, it can be asserted that the ryegrass can be grown without hesitation under the climate conditions of Erzincan and be an alternative forage crop. While preparing the rations, it is necessary to take into account the harvest time of ryegrass. Widespread cultivation of ryegrass can help eliminate the deficit in quality roughage for the nutrition of ruminants.

Statement of Conflict of Interest

The authors of the article declare that there is no conflict of interest.

Author's Contributions

The authors declare that they have contributed equally to the article.

** Part of the data in this article was held in the IV. It was presented as a paper at the International Conference on Developments in Basic and Application Sciences

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