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Araştırma Makalesi/*Research Article (Original Paper)*

The Change in The Forage Quality of Alfalfa (*Medicago Sativa* L.) in Grazing and Non-Grazing Artificially Established Pastures

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Abstract: This research was conducted to determine the forage quality of alfalfa during grazing season in artificial pasture from the years 2010 to 2012 in Isparta. The mixture of the pasture used were *Medicago sativa* L. (15%) + *Onobrychis sativa* Lam. (15%) + *Agropyron cristatum* L. (35%) + *Bromus inermis* L. (35%). Forage samples were collected from grazing and non-grazing areas once every 15 days during the grazing seasons. The crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF) contents, *in vitro* dry matter digestibility (IVDMD), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg) ratios were determined on the alfalfa forage samples. According to results of this research, the CP, IVDMD, Mg, P and K contents decreased throughout the grazing season, while ADF, NDF and Ca contents increased in grazing and non-grazing areas. The ADF, NDF, Ca, Mg contents of alfalfa in non-grazed areas were higher than the grazed areas, while CP, IVDMD, P and K contents of grazed areas were higher than non-grazed areas. It can be concluded that the harvesting at the late stages caused a reduction in forage quality of alfalfa in grazing and non-grazing areas.

Key words: Artificial pasture, ADF, CP, forage quality, *Medicago sativa*

Yapay Meranın Otlanan ve Korunan Alanlarında Yoncanın (*Medicago sativa* L.) Ot Kalitesindeki Değişim

Özet: Bu araştırma otlatma sezonu boyunca yapay merada yoncanın ot kalitesindeki değişimi belirlemek amacıyla 2010-2012 yılları arasında Isparta'da yürütülmüştür. Merada kullanılan karışım *Medicago sativa* L. (%15) + *Onobrychis sativa* Lam. (%15) + *Agropyron cristatum* L. (%35) + *Bromus inermis* L. (%35)'dır. Otlatma sezonu boyunca otlanan ve korunan alanlardan her 15 günde bir bitki örnekleri toplanmıştır. Yonca örneklerinde ham protein, ADF, NDF, in vitro kuru madde sindirilebilirliği (IVDMD), fosfor, potasyum, kalsiyum ve magnezyum içerikleri belirlenmiştir. Araştırma sonuçlarına göre hem korunan hem otlatılan alanlarda, yoncanın ham protein, IVDMD, Mg, P ve K içerikleri otlatma mevsimi boyunca azalırken, ADF, NDF ve Ca içerikleri artmıştır. Korunan alanlardaki yoncanın ADF, NDF, Ca ve Mg içeriği otlanan alanlara göre daha yüksek bulunurken, ham protein, IVDMD, P ve K içerikleri otlanan alanlarda daha yüksek bulunmuştur. Sonuç olarak, hasat zamanının gecikmesi otlanan ve korunan alanlarda yoncanın ot kalitesinde azalmaya neden olmuştur.

Anahtar kelimeler: Yapay mera, ADF, ham protein, ot kalitesi, *Medicago sativa*

Introduction

The deep root system of alfalfa makes it more drought tolerant than cool-season legumes and grasses. Although alfalfa does not make maximum growth during summer droughts, it usually provides good summer pastures. During extreme drought, this aspect is even more important since cool-season grasses become dormant. Combining grass with alfalfa in pastures to be grazed provides several benefits, but it also can create additional challenges with fertilizer, irrigation and grazing management. Including grass and non-bloating legumes like birdsfoot trefoil and cicer milkvetch in a mixture with alfalfa can lower the bloat potential compared to pure alfalfa. Energy of pure alfalfa also can be low relative to the high protein levels. By adding grass, the overall diet mixture may provide a better energy-to-protein ratio for the grazing animal than alfalfa alone (Volesky 2010).

Establishment of artificial pasture plays an important role in producing sufficient and good quality feed for damaged vegetation. In most situations, dryland pastures best comprised a simple mixture containing two and three species having similar palatability, season of growth, grazing tolerance, drought tolerance and rare cases of regrowth (Holzworth et al. 2003). Alfalfa, sainfoin or other legumes planted in mixtures with grasses, provide nitrogen to increase yield and nutritive values of the entire mixture. However, it is sometimes difficult to keep legumes in the mixture because of their high palatability (Holzworth and Weisher 2010). Alfalfa, crested wheatgrass and smooth brome cultivar would be suited for use in binary grass-alfalfa mixtures for dryland hay production (Berdahl et al. 2001).

The relative performance of animals is generally associated with the forage quality. Higher nutritive quality of feeds are dependent on higher levels of cell-soluble, crude protein and mineral contents. These components of forage decline substantially with the advanced plant growth and reach the lowest level when plants become quality (Koc and Gokkus 1994) as in all steppe vegetation. The changing trend of nutritive component of forage shows great differences among range types because the timing and length of growing season differ among them due to climate (Holechek et al. 2004). Most plants show a similarity in declining nutrient composition with advancing development towards maturation (Rama et al. 1973; Stubbendieck and Foster 1978; Rebole et al. 2004).

Therefore, in this research, it was aimed to determine chemical composition of the alfalfa during the grazing season in artificial pastures established in the Mediterranean Region of Turkey.

Materials and Methods

This research was conducted at Suleyman Demirel University Research Farm in Isparta Province (37°45′N, 30°33′E, elevation 1035 m) located in the Mediterranean region of Turkey on three consecutive years of 2010 and 2012. Total precipitation was 623.5 mm in 2010, 400 mm in 2011, 436 mm in 2012 (Jan-July). The long-term average is 506 mm. Average temperature was 13.39 ^{o}C in 2010, 12.08 ^{o}C in 2011, 10.89 ^{o}C in 2012 (Jan-July). The long- term average is 12.01 ^{o}C (Figures 1 and 2). The major soil characteristics of the research area, determined based on the method described by Rowell (1994), were as follows: The soil texture was clay loam, the organic matter was 1.3% as determined using the Walkley–Black method, the lime was 7.1% as determined using a Scheibler calcimeter, the total salt was 0.29%, the exchangeable K was 122 mg kg^{-1} by 1 N NH_4OAc, the extractable P was 3.3 mg kg^{-1} by 0.4 N $NaHCO_3$ extraction, and the pH of a soil-saturated extract was 7.7. The soil type was calcareous fluvisol.
In March 2010, two artificial grazing lands, covering 1.5 ha pasture each land were established at university farm. Pasture was composed of alfalfa (*Medicago sativa* L., 15%) + sainfoin (*Onobrychis sativa* Lam., 15%) + crested wheatgrass (*Agropyron cristatum* L., 35%) + smooth bromegrass (*Bromus inermis* L., 35%). Cutting and maintenance applications were made in the first year. There are no irrigation in the pasture. Nitrogen (ammonium nitrate) and phosphorus (triple super phosphate) were applied in March and October in second and third years. Pastures were harvested twice during the end of June and beginning of October in 2010. Animal grazing applications were performed in the second and the third year of the study since the first year covered only the establishment of the artificial pastures. The animals were turned out to pasture for grazing on the 1st of May and the grazing was terminated on the 1st of August each year. 10 Holstein male calves with an average 6 months old were included and allocated evenly to artificially established pasture in the experiment which lasted for 90 days in 2011 and 2012. The number of animals is determined according to carrying capacity of artificial pasture. The animals had a free access to the water during all experimental periods.

Four non-grazed areas within pasture were established in order to determine chemical composition changes of alfalfa and fenced with wires by 4×3 m size and forage samples were collected by using 0.5 m^2 (0.5×1 m) quadrats fortnightly from May to August each year. The crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF) contents, *in vitro* dry matter digestibility (IVDMD), phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg), ratios were determined as well.

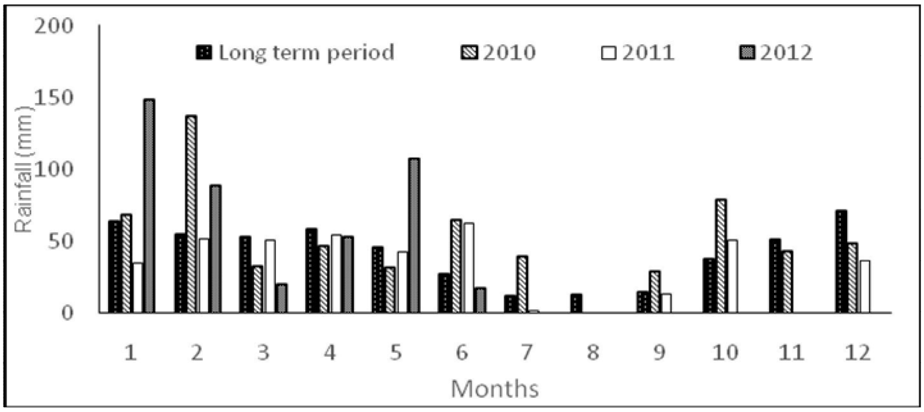

Figure 1. Rainfall values for individual experimental years and over the long term

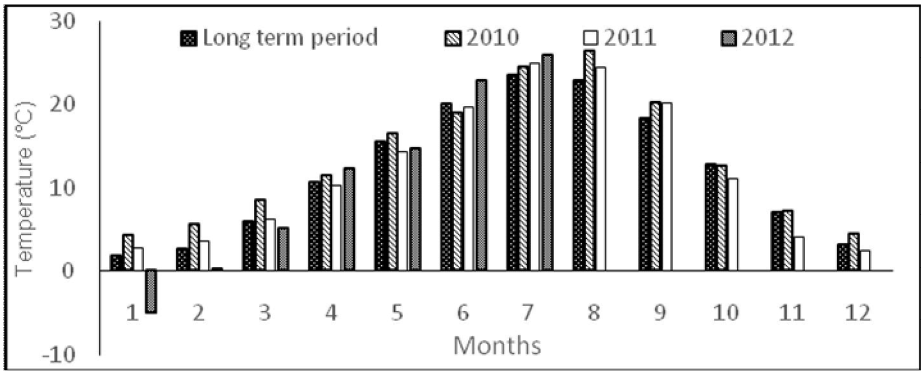

Figure 2. Temperature values for individual experimental years and over the long term

The collected samples after the harvest were weighed and dried at 70 °C for 48 h. The dried samples were reassembled and ground to pass through a 1-mm screen. The crude protein (CP) content was calculated by multiplying the Kjeldahl nitrogen concentration by 6.25 (Kacar and Inal 2008); K, Ca and Mg contents of samples was determined using an atomic spectrophotometer after digesting the samples with $HClO_4$:HNO_3 (1:4); P content was determined by vanadomolybdophosphoric yellow colour method (Kacar and Kovancı 1982). The acid detergent fiber (ADF) and neutral detergent fiber (NDF) concentrations were measured according to methods from Ankom Technology. Tilley and Terry's (1963) methods were used to determine *in vitro* dry matter digestibility (IVDMD) of samples.

The data were subjected the analysis of variance using GLM procedure (MINITAB 2010). The means were compared by pairwise comparison test by Duncan at the 5 % level of significance.

Results and Discussion

The effects of the grazing and sampling times on all investigated traits were significant as shown in Table 1. The effects of the grazing and sampling times on CP contents were significant. The CP contents decreased in grazed and non-grazed areas. The highest CP contents were obtained from beginning of the grazing season while the lowest CP contents were determined at end of the grazing season (Figure 3). Statistically significant interactions between grazing × sampling times were found for CP ratios in alfalfa (Table 1). These interactions indicated that harvesting stage affected CP ratios differently according to the grazing. The CP contents were decreased linearly throughout the grazing season in grazed and non-grazed areas (Figure 3). Maturity stage at harvest is the most important factor determining forage quality. Besides N, and hence protein, most minerals decline with advancing plant development. Other reports also support that the CP contents decreases by advancing stage of maturity (Koc et al. 2000; Rebole et al. 2004; Turk et al. 2011), suggesting that animals should be supplemented with protein sources, especially towards the end of the grazing season. Forage quality lessens substantially towards the end of growing season. The CP ratios of the grazed areas were higher than that of non-grazed areas in the present study. This could be associated with the continued re-growth of the plants in grazed areas because young plant tissues are more nutritious than dead or mature plant (Lyons et al. 1996).

Table 1. Results of analysis of variance and mean squares of the traits determined

Sources of variations	df	CP	NDF	ADF	IVDMD	P	K	Ca	Mg
Block (year)	6	0.014 ns	3.21**	1.78**	0.81 ns	0.0002	0.0030*	0.0011	0.0001
Year	1	75.24**	278.1**	559.44**	98.11**	0.0019	0.4161**	0.6246**	0.0443**
Grazing (G)	1	3.45**	22.12**	2.78**	201.62**	0.0091**	0.4891**	0.1735**	0.1747**
Sampling Times (ST)	6	282.18**	1259.62**	763.71**	1403.1**	0.0588**	1.9124**	4.5678**	0.1438**
G x ST intr	6	0.41**	5.81**	2.58**	8.69**	0.0003	0.0142**	0.0064**	0.0156**
G x Year	1	1.30	12.91	0.10 ns	1.61 ns	0.00002	0.0089	0.0217	0.0031
ST x Year	6	0.49	0.21 ns	0.24 ns	0.11 ns	0.00001	0.0017	0.0053	0.0004
G x ST x Year intr.	6	0.033 ns	0.48 ns	0.15 ns	0.05 ns	0.00000	0.0037	0.0009	0.0001
Error	78	0.03	0.71	0.31	0.74 ns	0.00008	0.0010	0.0007	0.0002

df: degrees of freedom, ns: not significant, *$p<0.05$ and **$p<0.01$

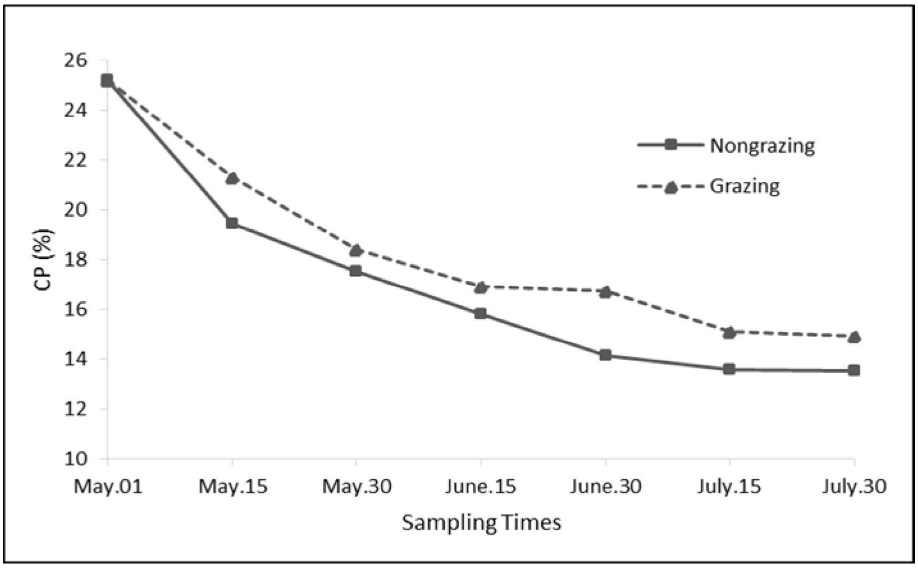

Figure 3. Seasonal variation of CP content of alfalfa in artificial pastures

As shown in Table 1, ADF and NDF contents of alfalfa were significantly affected by both grazing and sampling times. The NDF and ADF contents increased linearly in grazed and non-grazed areas. The effects of grazing × sampling times interactions on ADF and NDF contents were found statistically significant ($p< 0.05$). The reason for this, differences are the changes in the ADF and NDF rates. Acid detergent fiber and NDF contents were increased during the grazing season (Figures 4 and 5). This could be explained by the decrease in proportion of leaves and the increase of the stems proportion with advanced maturity. Because, ADF and NDF contents of stems are higher than the leaves. Similar results were reported by Karslı et al. (2003), Kaya et al. (2004), Erkovan et al. (2009), Turk et al. (2014), Albayrak et al. (2009). The trends in ADF and NDF contents with increasing maturity are normally the reverse of protein (Rebole et al. 2004; Turk et al. 2009). Young plant cells has the primary cell wall, but also the secondary cell wall occurs with maturing. This causes being the more fibrous of mature plants (Arzani et al. 2004). ADF and NDF contents of non-grazed areas were higher than those of grazed areas in the present study. This could be explained by the continued re-growth of the plants in the grazed areas.
The grazing and sampling times showed significant effects on IVDMD of alfalfa (Table 1). The IVDMD was decreased throughout the grazing season in grazed and non-grazed areas (Figure 6). Similar results were reported by Hitz and Russell (1998), Karsli et al. (2003). The reason for this decrease in digestibility of plants is the increase in the lignin content (Jung and Vogel, 1992). The decrease in IVDMD resulted from the increase structural tissues in stems (Arzani et al. 2004). Pinkerton (1996) stated that there is a close relationship between digestibility and cell wall structure. Fiber content is increased as the plants grow, the digestibility decreases (Erfanzadeh 2001). Overall, IVDMD varies depending on crude cellulose levels in its structure. Digestibility of plenty of leafy green forage is very high, whereas

the increase of the stems proportion with advanced maturity causes a decrease digestibility (Aksoy and Yilmaz 2003).

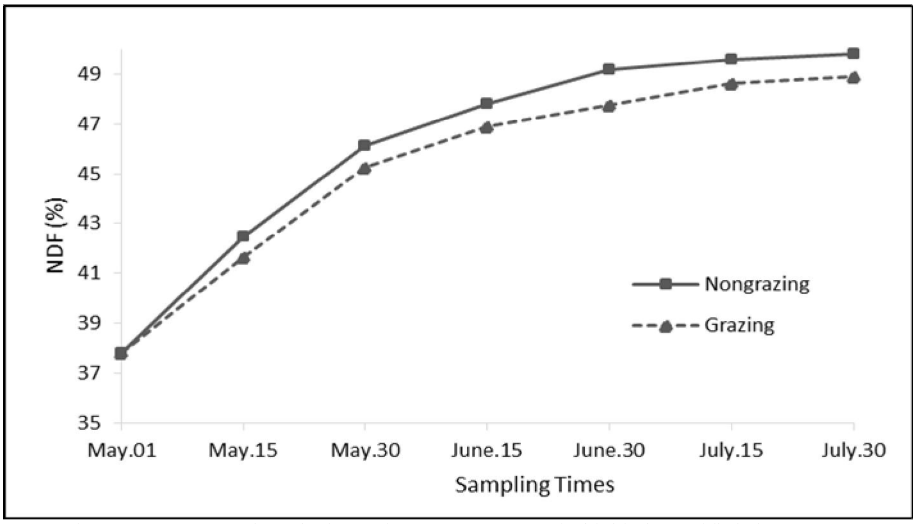

Figure 4. Seasonal variation of NDF content of alfalfa in artificial pastures

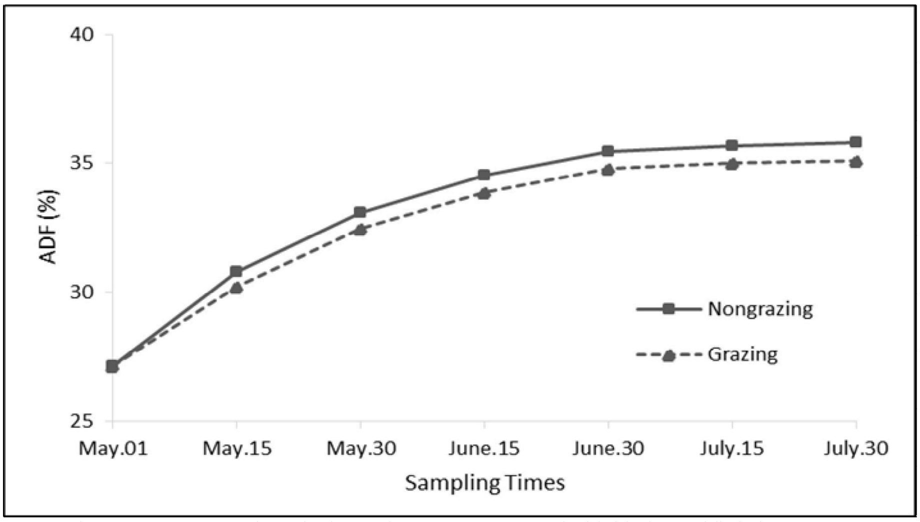

Figure 5. Seasonal variation of ADF content of alfalfa in artificial pastures

The P, K and Mg contents were decreased during the grazing season, while Ca contents increased in the present study (Figures 7, 8, 9, and 10). The changing trend of nutritive component of forage shows great differences among range types because the timing and length of growing season differ among them due to climate (Holechek et al. 2004). Most plants show a similarity in declining nutrient composition with advancing development towards maturation (Rama et al. 1973; Stubbendieck and Foster 1978; Rebole et al. 2004).

Forage quality is determined by maturity stage at harvest as the most important factor. Because P, Ca, Mg and K contents of forage decreased with delayed cutting, forage quality declines with advancing maturity (Blaser et al. 1986; Tan and Serin 1996; Turk et al. 2007). Tan et al. (2003) reported that the contents of K, Mg, Ca and P decreased as plant growth advanced. These results are in agreement with our results, except for Ca content. There is a rapid uptake of minerals during early growth and a gradual dilution as the plant matures (Lanyasunya et al. 2007). The changes in element content with maturity are related to the increasing stem to leaf ratio. Leaves are richer in mineral nutrients than stems (Tan et al. 1997), and the proportion of leaves declines as plant growth advances because of senescence of the lower leaves or damage by diseases (Albrecht and Marvin 1995). Changes in P content normally parallel those of protein in regard to seasonal changes. Phosphorus and Mg contents both decreased significantly with advancing season (Oelberg 1956). In contrast, Ca content generally increases as the season advances (Savage and Heller 1947). The increase with maturity was explained on the basis of the increased amount of cellular material which is composed principally of this element.

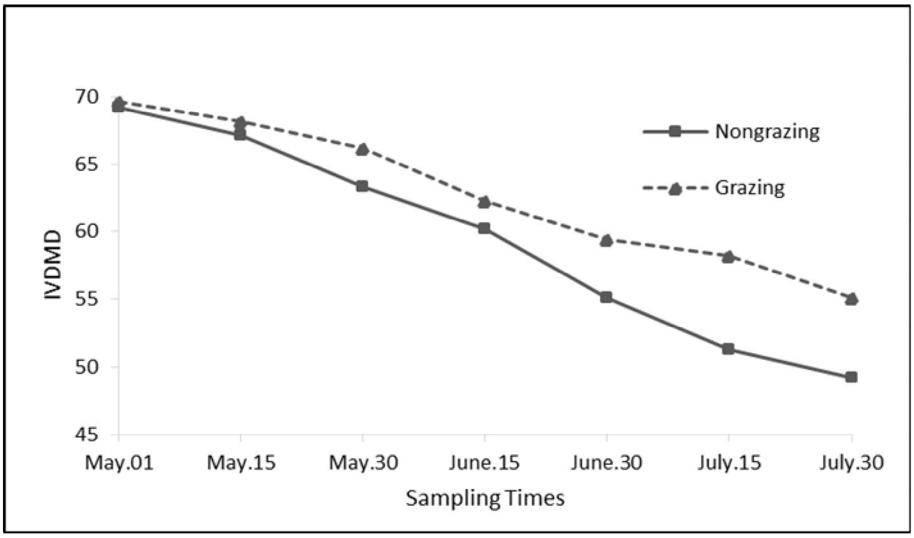

Figure 6. Seasonal variation of IVDMD of alfalfa in artificial pastures

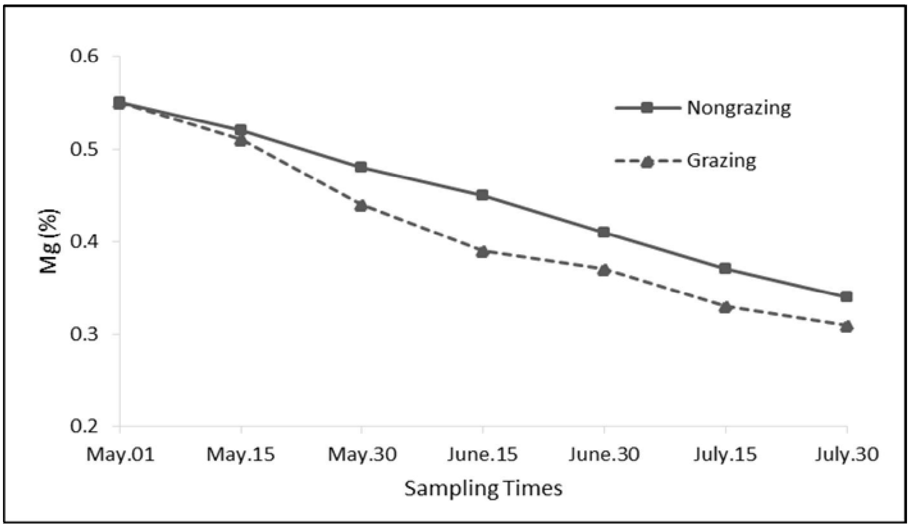

Figure 7. Seasonal variation of Mg content of alfalfa in artificial pastures

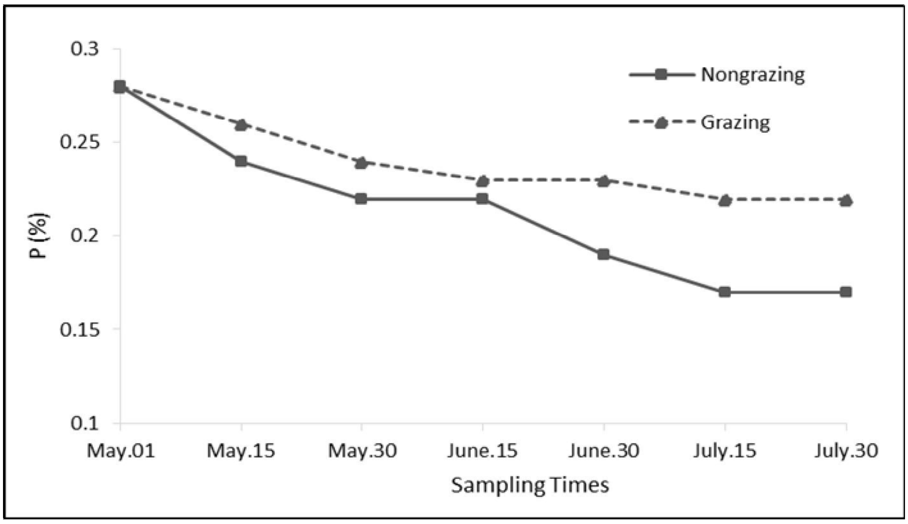

Figure 8. Seasonal variation of P content of alfalfa in artificial pastures

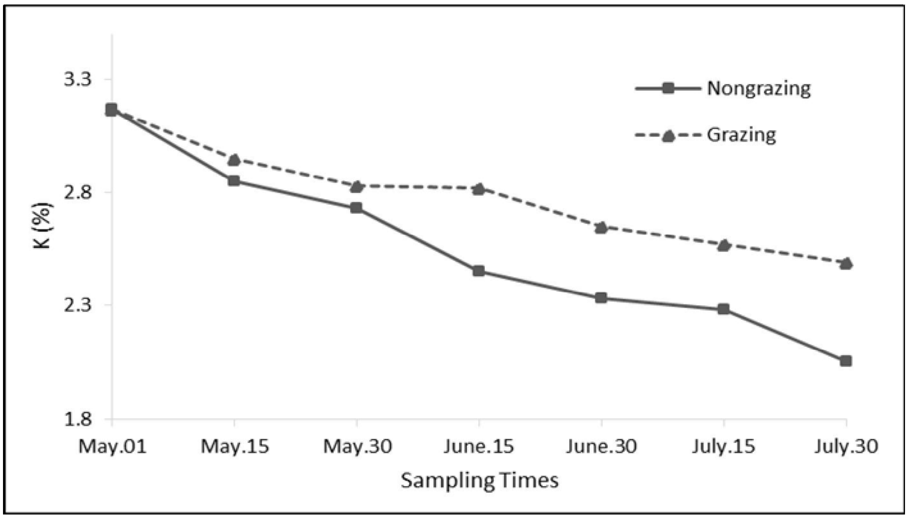

Figure 9. Seasonal variation of K content of alfalfa in artificial pastures

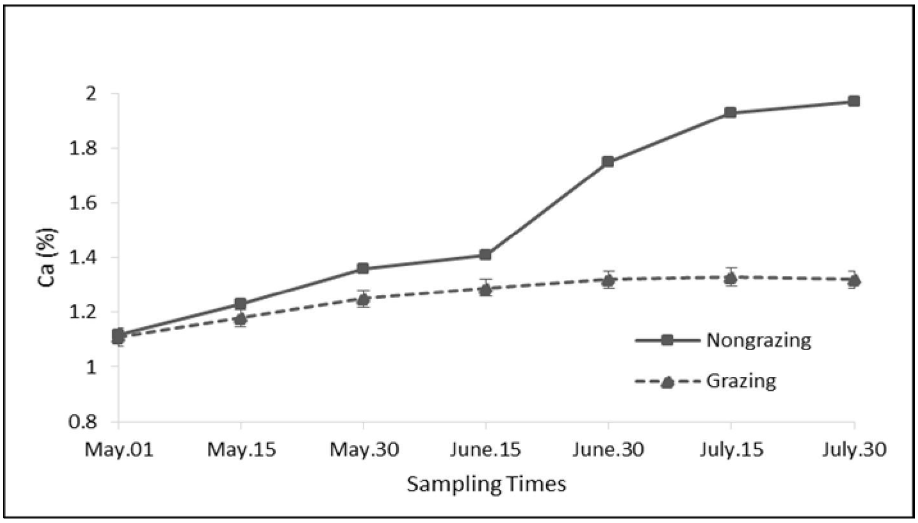

Figure 10. Seasonal variation of Ca content of alfalfa in artificial pastures

In the present study the Ca and Mg ratios of the non-grazed areas were higher than that of grazed areas The P and K ratios of the grazed areas were also higher than that of non-grazed areas. The American

National Research Council (NRC 1996) recommends that forage crops should contain 3.1 g kg^{-1} Ca, 6.5 g kg^{-1} K concentration for beef cattle. Tajeda et al. (1985) reported that forage crops should contain at least 0.3% of Ca, 0.2% of Mg, 0.8% of K for ruminants. The chemical properties found in this research indicate that the nutritional quality values of artificial pastures were higher than all the recommended standard values for ruminants.

Conclusions

ADF, NDF and Ca contents were increased in grazing and non-grazing areas while the CP, IVDMD, Mg, P and K contents were decreased. The ADF, NDF, Ca, and Mg contents of alfalfa in non-grazed areas were higher than the grazed areas, while CP, IVDMD, P and K contents of grazed areas were higher than non-grazed areas. In conclusion, it can be emphasized that forage quality of alfalfa in grazing and non-grazing areas was decreased by harvesting at the late stages.

Acknowledgments

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