

## Evaluation of the Nutrient Content and Protein Fractions of Four Different Common Vetch Varieties

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**Abstract:** The objectives of this study were to determine the chemical composition, digestibility, in situ N kinetics, and by-pass protein contents of 4 different common vetch varieties grown under irrigation. Four different common vetch varieties (Emir 20/1, Nilüfer 17/1, 28/1, and Uludağ 31/4) with different characteristics were selected. The vetch plots were planted on June 15 2002 and each variety was randomly assigned to 3 replications. The vetches were harvested by hand using a clipper on September 11. All samples were analyzed for dry matter (DM), ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent insoluble nitrogen (ADIN) concentrations. In vitro dry matter digestibility, and in situ degradability of samples at given times were also determined. While the concentrations of DM and ADIN-N did not differ, concentrations of organic matter (OM), CP, NDF, and ADF varied significantly among different vetch varieties ( $P < 0.05$ ). The water soluble DM content of Nilüfer 17/1 was significantly greater than that of 28/1 ( $P < 0.05$ ), but in situ DM degradabilities after 48-h incubation, in vitro dry matter digestibilities, and metabolizable energy (ME) and net energy for lactation (NEL) values were statistically similar among the vetch varieties ( $P > 0.05$ ). While the water soluble protein (WSP) concentration was lower, concentrations of potentially degradable protein (PDP), and by-pass CP as a percentage of total digestible CP or DM were significantly greater in 28/1 than in Nilüfer 17/1 and Uludağ 31/4 ( $P < 0.05$ ). However, concentrations of non-degradable protein (NDP) and in situ CP degradabilities after 48-h incubation did not differ among the vetch varieties ( $P > 0.05$ ). It can be concluded that if high by-pass protein content is desired, 28/1 and Emir 20/1 should be preferred over the other varieties. Otherwise, Nilüfer 17/1 and Uludağ 31/4 should be first choice, based on DM digestibility and CP contents, because the energy contents and percentage of non-digestible CP did not differ among the vetches.

**Key Words:** Common vetch, digestibility, by-pass protein

### Dört Farklı Adi Fiğ Varyetesinin Besin Madde İçeriği ve Protein Fraksiyonlarının Değerlendirilmesi

**Özet:** Bu çalışmanın amacı, dört farklı adi fiğ varyetesinin besin madde kompozisyonu, sindirilebilirliği, in situ N kinetiği ve by-pass protein içeriğini belirlemektir. Farklı özelliklerde dört farklı adi fiğ varyetesi (Emir 20/1, Nilüfer 17/1, 28/1 ve Uludağ 31/4) seçilmiştir. Fiğ parselleri 15 Haziran 2002'de, her varyete tesadüfi olarak seçilmiş üç tekrerrü şeklinde ekilmiştir. Fiğler 11 Eylül'de çim makası kullanılarak elle hasat edilmiştir. Tüm örnekler kuru madde (KM), ham kül (HK), ham protein (HP), nötral deterjan fiber (NDF), asit deterjan fiber (ADF) ve asit deterjan solüsyonunda çözünmeyen azot (ADIN) içeriklerini belirlemek için analiz edilmiştir. Örneklerin in vitro KM sindirimi ve belli saatlerde in situ yıkılmaları da belirlenmiştir. Örneklerin KM ve ADIN içerikleri benzer, organik madde, HP, NDF ve ADF içerikleri fiğ varyeteleri arasında anlamlı derecede farklı bulunmuştur ( $P < 0,05$ ). Nilüfer 17/1 varyetesinin suda çözünen KM içeriği 28/1 varyetesine oranla anlamlı derecede yüksek ( $P < 0,05$ ), ancak 48 saat inkubasyon sonrası in situ KM yıkılmaları, in vitro KM sindirim, metabolik enerji ve net enerji laktasyon değerleri istatistiksel olarak varyeteler arasında benzer bulunmuştur ( $P > 0,05$ ). 28/1 varyetesinin suda çözünen protein içeriği düşük, potansiyel olarak yıkılabilir protein, total sindirilebilir HP ve KM içerisindeki by-pass protein oranları Nilüfer 17/1 ve Uludağ 31/4'e oranla daha yüksek bulunmuştur ( $P < 0,05$ ). Ancak, yıkılmayan protein oranı, 48 saat inkubasyon sonrası HP yıkılmaları fiğ varyeteleri arasında değişmemiştir ( $P > 0,05$ ). Eğer yüksek by-pass protein içeriği arzulaniyorsa, 28/1 ve Emir 20/1 varyeteleri diğer varyetelere tercih edilebilir, aksi takdirde, enerji içerikleri ve sindirilmeyen HP içerikleri varyeteler arasında değişmediği için, KM sindirimi ve HP içerikleri baz alınarak Nilüfer 17/1 ve Uludağ 31/4 varyetelerinin ilk tercih olabileceği sonucuna varılmıştır.

**Anahtar Sözcükler:** Adi fiğ, sindirim, by-pass protein

## Introduction

Feed cost comprises from 50% to 70% of total farming expenses in Turkey (1). In order to reduce feed costs and create more sustainable management systems for moderate-sized, family operations, value-added livestock enterprises must be integrated with existing cropping enterprises.

One of the most important factors affecting the Turkish farming system is the lack of cheap, abundant and high quality feedstuff. The feeding of low-quality forages such as crop residues (wheat, barley straw) and low-quality hays with protein (meal) or energy supplementation (grain barley) to wintering ruminants is a common practice in Turkey. However, these low-quality forages may limit the performance of dairy and fast growing beef cows due to their high gut-filling capacity (2). Dairy cows can only produce high milk yields and beef cows can only reach their maximum potential if their intermediary metabolism is supplied with sufficient nutrients (1). Thus, high-quality forages have to be produced.

Vetch is grown in some parts of Turkey as a rotation plant and utilized as grain or hay in animal production. Vetch hay can contain up to 20% crude protein (CP). It can supply a considerable amount of the CP requirement of ruminant animals. Because ruminant animals possess rumen microbes, which can digest cellulose and use non-protein nitrogen to produce high quality microbial protein, ruminants are the predominant forage utilizer among animals (3). Forages provide 83% of the protein requirement of beef cattle and 90% of the protein requirement of sheep (4).

Forage protein serves as a source of metabolizable protein for ruminants by providing both ruminally degradable protein for microbial growth and some ruminally undegradable protein for intestinal digestion (5). Because of the rapid and extensive degradation of forages in the rumen (6), escape protein concentrations of forages are usually low (7). However, degradation rates and thus escape protein concentrations vary among legumes and grasses, and even within legume and grass species (6). In addition to forage species, factors such as temperature and drought have been reported to affect protein in forages (8). Therefore, protein fractions of similar forage species may differ from one location to another.

The objectives of this study were to determine the chemical composition, digestibility, in situ N kinetics, and by-pass protein contents of 4 different common vetch varieties grown under irrigation.

## Materials and Methods

Four different common vetch varieties (Emir 20/1, Nilüfer 17/1, 28/1, and Uludağ 31/4) with different characteristics were selected. All of the vetches were grown under irrigation. The vetch plots were planted on June 15 2002 and each variety was randomly assigned to 3 replications. Rows were 5 m long and with 20 cm spacing. Each plot consisted of 6 rows. The vetches were seeded at 48 g per m<sup>2</sup>. The vetch fields were watered every 10 days and were fertilized with 40 kg of nitrogen and 70 kg of phosphorus per hectare.

Vetches were randomly harvested from a 1 m<sup>2</sup> area in duplicate by hand using a clipper on September 11 and air-dried. While Nilüfer 17/1 and Uludağ 31/4 were in the late blooming stage, Emir 20/1 and 28/1 were in the early seedling stage of maturity when the vetches were harvested. All of the samples obtained from the 1 m<sup>2</sup> area were weighed and first air-dried. Then, the dry matter (DM) of samples was determined by oven drying of triplicate sub-samples at 65 °C for 72 h, after air-drying (9). DM yields were calculated by multiplying the DM content of the vetches by fresh hay yields.

All of the analyses were performed on dried samples. Dried samples were ground to pass through a 1 mm screen before analysis. Ash concentrations of samples were determined in a muffle furnace at 550 °C for 8 h. All samples were analyzed for CP by Kjeldahl procedure (9), neutral detergent fiber (NDF) (10), acid detergent fiber (ADF) (11) and acid detergent insoluble nitrogen (ADIN) (10) concentrations. In vitro dry matter digestibilities (IVDMD) of samples were determined by the procedure described by Tilley and Terry (12), as modified by Marten and Barnes (13). Ruminal ingesta from an alfalfa-fed ruminally fistulated ram was hand-collected and strained through 4 layers of cheesecloth to provide the inocula for the IVDMD determination. Metabolizable energy (ME, Mcal/kg) and net energy for lactation (NEL, Mcal/kg) values were calculated using the following equations (14):

$$\text{ME, (Mcal/kg)} = \text{Digestible energy} \times 0.82$$

$$\text{NEL, (Mcal/kg)} = 0.00245 \times \text{digestible energy, kg} - 0.12$$

To estimate the in situ degradation kinetics and fractions of CP, oven-dried samples of vetch hay were ground through a 2-mm screen. Approximately 3.5 g of each vetch sample was weighed into a Dacron bag. The bags used were constructed of Dacron polyester having an average pore size of 50  $\mu$  and internal dimensions of 15 x 7 cm. The suspension of the bags in the rumen was accomplished by tying them onto tygon tubing with nylon string. Six bags were affixed to each tygon tubing for each incubation time.

Three mature fistulated Morkaraman x Kıvrıkcık lambs (averaging 40 kg) fed chopped alfalfa hay were used for the incubation of samples in Dacron bags in this study. Samples in Dacron bags were placed in the rumen and incubated for 0, 12, and 48 h. Two bags of sample for each variety and each plot were inserted into the rumen of each ram for each incubation time. After the removal of the bags from the rumen, they were washed under running water in a small washing machine for about 15 min. Then all bags were dried for 24 h at 65 °C in a drying oven and DM recovery was determined. Undigested residues were analyzed for nitrogen by the micro-Kjeldahl procedure (8).

Loss of DM from the bags caused by exposure of substrates to the digestive action of the rumen and the washing process that followed resulted in the partitioning of CP in each of the varieties into 3 fractions: 1) soluble fractions of CP (WSP) were determined as the differences between initial CP content and amounts of CP recovered in 0 time-incubation; 2) potentially digestible fractions of CP (PDP) were determined as 100 - (non-digestible fraction and water soluble fractions of CP); 3) non-digestible fractions of N (NDP) were determined as the differences between initial CP content and amount of CP recovered after 48 - h incubation of samples in the rumen (15).

A modified technique reported by Mullahey et al. (16) was used to determine the percentage of vetch protein that escaped ruminal degradation.

The proportion of total protein that would escape ruminal digestion was calculated as total residual N remaining after 12 - h incubation, adjusted for the indigestible N (ADIN) using the following equations (16):

By-pass Protein Percentage, % of total protein = (Total residual N - ADIN of total residue) / (Total plant-N-ADIN of total plant) x 100.

By-pass Protein Percentage, % DM = (by-pass protein percentage) x (% Total plant CP).

### Statistical analysis

All data were subjected to analysis of variance for completely randomized design using the GLM procedure of SAS and means were separated by Duncan's t-test (17).

## Results

Table 1 shows the chemical composition of the different vetch varieties conserved as hay. DM concentrations of the vetches were significantly different when harvested. While concentrations of DM and ADIN-N did not differ, concentrations of OM, CP, NDF, and ADF were significantly different among the different vetch varieties conserved as hay. OM concentrations of Emir 20/1 and Uludağ 31/4 were significantly greater than that of Nilüfer 17/1 ( $P < 0.05$ ). CP concentrations of the vetch hays ranged between 17.75 and 20.30 and were the highest in Uludağ 31/4 ( $P < 0.05$ ). Nilüfer 17/1 had the lowest NDF (40.76%) and 28/1 had the lowest ADF (28.14%) concentrations among the vetch varieties ( $P < 0.05$ ).

DM yields of the vetches were significantly different and the highest yields were obtained with 28/1 variety (Figure,  $P < 0.05$ ).

The water soluble DM (WSDM) content of Nilüfer 17/1 was significantly greater than that of 28/1 vetch ( $P < 0.05$ ), but in situ DM degradabilities after 48-h incubation, in vitro dry matter digestibilities, ME, Mcal/kg and NEL, Mcal/kg values were statistically similar among the vetch varieties (Table 2;  $P > 0.05$ ).

While the WSP concentration was lower, concentrations of PDP, by-pass CP as a percentage of total digestible CP and DM were significantly greater in 28/1 than in Nilüfer 17/1 and Uludağ 31/4 (Table 3;  $P < 0.05$ ). However, concentrations of NDP and in situ CP degradabilities after 48-h incubation did not differ among the vetch varieties ( $P > 0.05$ ).

## Discussion

The aim of this study was to determine the chemical composition, digestibility, in situ N kinetics, and by-pass protein contents of 4 different vetch varieties newly adapted to eastern Anatolian conditions.

Table 1. Chemical compositions of different vetch varieties conserved as hay, % DM.

Varieties	DM as fresh	DM as hay	Ash	OM	CP	ADIN-N, %CP	NDF	ADF
Replication (n)	6	6	6	6	6	6	6	6
Emir 20/1	22.46 <sup>a</sup>	94.75	10.40 <sup>b</sup>	89.60 <sup>a</sup>	17.87 <sup>b</sup>	15.86	49.36 <sup>a</sup>	29.16 <sup>b</sup>
28/1	24.46 <sup>a</sup>	94.05	11.17 <sup>a-b</sup>	88.83 <sup>a-b</sup>	17.75 <sup>b</sup>	15.24	44.17 <sup>a-b</sup>	28.14 <sup>b</sup>
Nilüfer 17/1	14.91 <sup>b</sup>	94.02	12.64 <sup>a</sup>	87.36 <sup>b</sup>	19.79 <sup>a</sup>	17.10	40.76 <sup>b</sup>	30.26 <sup>b</sup>
Uludağ 31/4	15.25 <sup>b</sup>	93.98	10.20 <sup>b</sup>	89.80 <sup>a</sup>	20.30 <sup>a</sup>	14.79	49.29 <sup>a-b</sup>	32.91 <sup>a</sup>
SEM	1.01	0.76	0.87	0.87	1.27	1.63	3.69	1.40

<sup>a-b</sup> ...Means with different superscripts within a column are significantly different, (P < 0.05). DM, dry matter; OM, organic matter; CP, crude protein; ADIN-N, non-digestible fractions of N, %CP; NDF, neutral detergent fiber; ADF, acid detergent fiber.

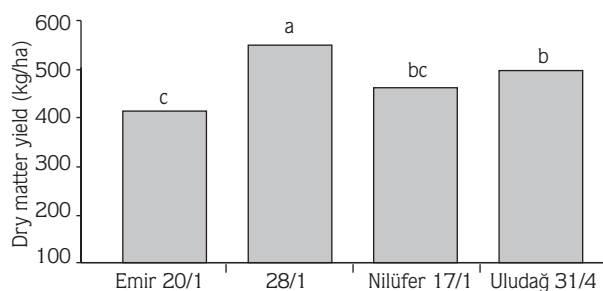


Figure. Dry matter yields of 4 vetch varieties.

Table 2. In situ degradability and in vitro DM digestibility (IVDMD) and energy values of different vetch varieties conserved as hay.

Varieties	WSDM, % DM	Degradability of DM *	IVDMD, % DM	ME, Mcal/kg	NEL, Mcal/kg
Replication (n)	18	18	6	6	6
Emir 20/1	43.96 <sup>ab</sup>	81.13	64.67	2.338	1.464
28/1	42.22 <sup>b</sup>	81.26	63.83	2.301	1.443
Nilüfer 17/1	46.20 <sup>a</sup>	81.56	60.96	2.156	1.373
Uludağ 31/4	43.77 <sup>a</sup>	81.67	63.85	2.308	1.445
SEM	1.93	1.08	3.21	0.104	0.06

<sup>a-b</sup> ...Means with different superscripts within a column are significantly different, (P < 0.05).

\* DM degradability after 48-h incubation. WSDM, water soluble dry matter, IVDMD, in vitro dry matter digestibilities; ME, metabolizable energy, Mcal/kg; NEL, net energy for lactation, Mcal/ kg.

The chemical compositions of the 4 varieties are presented in Table 1. Even though all of the vetches were planted and harvested on the same days, DM contents were different, indicating that Emir 20/1 and 28/1 matured faster than Nilufer 17/1 and Uludağ 31/4. It is

well known that cultivators often differ on physiological maturity, which results in differences in the concentration of DM among varieties. However, DM concentrations of vetch hays were similar and above 93.0%, indicating good conservation as hay. The OM content of Emir 20/1

Table 3. In situ degradability and fractions of CP in different vetch varieties conserved as hay.

Varieties	WSP, % CP	PDP, % CP	NDP, % CP	Degradability of CP*	By-pass CP, % CP	By-pass CP, % DM
Replication (n)	18	18	18	18	18	18
Emir 20/1	47.31 <sup>ab</sup>	45.13 <sup>ab</sup>	7.55	92.45	23.02 <sup>a</sup>	4.11 <sup>a</sup>
28/1	42.07 <sup>b</sup>	50.86 <sup>a</sup>	7.08	92.92	29.43 <sup>a</sup>	5.12 <sup>a</sup>
Nilüfer 17/1	56.63 <sup>a</sup>	36.26 <sup>b</sup>	7.11	92.89	11.73 <sup>b</sup>	2.01 <sup>b</sup>
Uludağ 31/4	54.11 <sup>a</sup>	38.67 <sup>b</sup>	7.22	92.78	13.87 <sup>b</sup>	2.84 <sup>b</sup>
SEM	5.93	5.68	0.98	0.98	4.01	0.64

<sup>a-b</sup> ...Means with different superscripts within a column are significantly different, ( $P < 0.05$ ). CP degradability after 48-h incubation. WSP: soluble fractions of CP, PDP: potentially digestible fractions of CP, NDP: non-digestible fractions of CP.

and Uludağ 31/4 was significantly greater ( $P < 0.05$ ) than that of Nilüfer 31/4 and ranged from 87.36% to 89.80%. OM levels in the current study were similar to those reported by Caballero et al. (18), but lower than those reported by the NRC (7), and Smith (19). The lower OM levels observed in this study could be due to soil contamination of vetch samples caused by irrigation. Uludağ 31/4 and Nilüfer 17/1 had significantly greater CP concentrations than Emir 20/1 and 28/1 ( $P < 0.05$ ). It is well known that the CP content of the same plant varieties can significantly differ (20). CP concentrations of the vetch varieties were between 17.75% and 20.30%, which are in agreement with the values reported in the literature (7,18,19). The concentrations of ADIN-N as a percentage of total CP were similar among the vetch varieties ( $P > 0.05$ ), and higher than the values reported in the literature (7). While Emir 20/1 and Uludağ 31/4 had significantly greater NDF concentrations than Nilüfer 17/1, ADF concentration was highest in Uludağ 31/4 ( $P < 0.05$ ).

Nilüfer 28/1 had a significantly greater DM yield (5613.0 kg/ha) than the other groups ( $P < 0.05$ ). Some experiments have shown cultivar differences in quality and quantity when harvested on common dates, but it is difficult to determine if the differences are confounded with maturity, which often differs among entries (21).

Water soluble DM contents were significantly different among the vetch varieties ( $P < 0.05$ ). Water soluble DM contents of forage are associated with forage intake. There is a strong positive correlation between WSDM and forage intake in ruminant animals. As WSDM increases, forage intake also increases (22,23). Water

soluble DM contents of the vetch varieties used in this study were above 42.00%, which are considerably high values, suggesting a good forage intake when fed to ruminant animals.

In situ ruminal DM degradabilities, after 48-h incubation, were similar among the vetch varieties. Similarly, in vitro DM digestibility values did not differ significantly among the vetch varieties ( $P > 0.05$ ). Because energy values were calculated from in vitro digestibility values, both ME and NEL values were statistically similar among the 4 vetch varieties ( $P > 0.05$ ). In vitro DM digestibility and ME values ranged from 60.96% to 64.67% DM; and from 2.156 to 2.338 Mcal/kg, respectively. Both digestibility and ME values are at the upper edge of values reported in the literature (7,19).

The vetch variety affected the relative proportion of CP fraction within the same maturity stage (Table 3). Nilüfer 17/1 and Uludağ 31/4 contained a higher concentration of WSP fraction than 28/1 ( $P < 0.05$ ), and ranged from 42.07% to 56.63% of total CP. Caballero et al. (18) indicated that CP fraction A increased from fresh to field-cured forage and CP fraction B<sub>1</sub> increased with maturity and decreased from fresh to dried. Both soluble fractions, the instantaneously fermented (CP fraction A) and the rapidly fermented (CP fraction B<sub>1</sub>), represented together between 410 and 530 g kg<sup>-1</sup> of total CP, which is in agreement with the results of the current study. The WSP fraction combines these fractions A and B<sub>1</sub>. The relative proportion of CP fractionated as PDP was higher in 28/1 than in Nilüfer 17/1 and Uludağ 31/4. The PDP fraction represented the second greatest

proportion of total CP, with values ranging from 36.26% to 50.86% of total CP. The undegraded and indigestible CP fraction, named NDP, made up some approximately 7% of total CP, and did not change among the varieties. The NDP fraction values observed in the current study were a little higher than those reported by Çelik (6.0%) (23) and Caballero et al. (5.0%) (18). These results illustrate that vetch CP is likely to be largely degraded in the rumen.

By-pass protein as a percentage of both total digestible CP and DM was significantly greater in 28/1 and Emir 20/1 than in Nilüfer 17/1 and Uludağ 31/4 ( $P < 0.05$ ). By-pass protein as a percentage of total digestible CP was 23.02, 29.43, 11.73, and 13.87 for Emir 20/1, 28/1, Nilüfer 17/1, and Uludağ 31/4, respectively. By-pass protein percentages of Nilüfer 17/1 and Uludağ 31/4 are similar to the value reported by the NRC (7), but by-pass protein percentages of Emir 20/1 and 28/1 are well above the value reported by the NRC (7). Similarly, Brown and Pittman (6) have reported that degradation rates and thus escape protein concentrations

vary among legumes and grasses, and even within legume and grass species.

In conclusion, while CP contents were significantly greater, by-pass protein contents were significantly lower in Nilüfer 17/1 and Uludağ 31/4 than in Emir 20/1 and 28/1. However, energy contents and percentage of non-digestible CP did not differ among the vetches. If high by-pass protein content is desired, 28/1 and Emir 20/1 should be preferred over the other varieties. Otherwise, Nilüfer 17/1 and Uludağ 31/4 should be first choice, based on DM digestibility and CP contents. 28/1 had the highest digestible DM yield among the vetch varieties. Even though Nilüfer 17/1 and Uludağ 31/4 had greater CP contents than the others, these differences were most likely due to differences in maturities.

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