

Chemical Composition and Nutritional Properties of Landraces Alfalfa (*Medicago sativa* L.) Cultivars

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18 Landraces alfalfa cultivars were examined in a field experiment in year 2004. Adiguzel, Ahlat(2), Alaköy, Burcu, Çayirbasi(16), Dilburnu(6), Dönemeç(21), Ercis, Gülgören(23), Gülsinberk, Hidirkoy(13), Kasimoglu(11), Köprüler(12), Mahmudiye, Mollakasim(4), Otluca, Otluyazi(20). Crude protein (CP), dry matter (DM), K, Mg, Na, Cu, Mn and Zn, as a mineral concentration, neutral detergent fiber (NDF), acid detergent lignin (ADL) and acid detergent fiber (ADF) as a energy content were studied. A greater proportion of significant phenotypic variations were observed many genotypes. Considerable significant correlations in chemical concentrations between genotypes were measured. Amongst the characteristics examined in this experiment there are negative significant correlation observed between organic method (OM) with K ($r = -0.358^*$), highly negative significant between NDF with DM ($r = -0.881^{**}$), NEL ($r = -0.465^{**}$), NEG ($r = -0.449^{**}$) and TDN ($r = -0.451^{**}$), between ADF with NEL ($r = -0.997^{**}$) and NEG ($r = -0.994^{**}$). Highly significant positive relationships were determined between NDF with ADF ($r = 0.451^{**}$). There are highly positive significant correlation observed between OM with DM ($r = 0.883^{**}$), NEL ($r = 0.462^{**}$). This result suggest that Dilburnu (6), Mahmudiye, Otluca and Ovakisla cultivars should provide useful genetic material for enhancing mineral concentration in alfalfa forage. Variation could be exploited as an additional source of genetic variation in breeding programs for quality trials to achieve a higher genetic gain for breeding cycle.

Key Words: *Medicago sativa*, Correlation coefficient, Mineral content, Energy content.

INTRODUCTION

Turkey is rich regarding species, ecotype, genetic diversity and gene sources of plants¹. Genotypic variation in alfalfa forage has been documented and forage quality has been altered through selection. Juan *et al.*² found differences in crude protein, neutral detergent fiber and acid detergent fiber among multifoliate and trifoliate entries. Divergent selection for alfalfa lignin concentration resulted in higher leaf to stem ratio and lower NDF in

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low lignin than in high lignin selections³. The concentration of most nutrients is highest in leaves with the greater concentration of K in stems. Differences exist among leaves according to their position on the plant. Basal leaves have higher concentrations at Mg and decrease from the top to the bottom of the shoot. The concentration of K increases progressively to near the top of the plant then decreases slightly. The variation in environmental conditions will influence nutrient concentrations in forage, because of changes in rate of dry matter production, ion movement in soil, root activity and the uptake of nutrients by the plant^{4,5}.

Data on quality changes of leaves and stems of modern alfalfa cultivars subject to varying harvest regimes. Leaf NDF concentration and digestibility typically decline slowly with increasing maturity, while stem NDF and acid detergent fibre (ADF) concentration increase⁶.

Energy and protein are the most valuable components of alfalfa. The crude protein contents of alfalfa can be determined directly in a laboratory, but there is no direct chemical test to determine energy value. The energy value of alfalfa hay is closely related to its fiber content as the alfalfa plant matures, its fiber content increases and its energy value decreases. Several fiber tests are used in U.S. to estimate the energy value of alfalfa hay.

NDF reflects the bulkiness of forage, there is a limit to the amount of NDF that will fit into an animal rumen. When that limit is reached, she will stop eating. There is no more room until a significant portion of the fiber in the rumen is digested and/or passes on to the lower gut. The basic assumption is that high quality forage digests more completely and has higher energy values.

Forage quality can be defined as the relative performance of animals when herbage is fed to livestock. It is the product of nutrient concentration, intake potential, digestibility and partitioning of metabolized products within the animal. In addition to the direct response of animals to forage quality, because of limitations associated with cost and time in using animals. However, forage quality often is estimated by *in vitro* or chemical means⁷.

One of the most affected focuses on quality is the plant genus and species dependent on plant genotypes⁸. The contents of the mineral matter obtained from forage crops should be a level to need the animal feeding requirements. Okuyan *et al.*⁹, provided that forage crops should include for K 0.30-0.80 and Mg 0.10-0.20 %.

Fresh grasses usually exhibit low carbohydrate availability and high N content. At early maturity stages, a large of this protein is non-protein N. Some research suggests that a low ratio of soluble carbohydrates to available N reduces the efficiency of N utilization by the ruminant^{10,11}. Besides energy protein relationship, marginal mineral contents can also limit productivity. Thus, Ca and Mg content in small grain forages affected performance and health in pregnant or lactating cows^{12,13}.

The objective of this study was to measure within cultivars variances for alfalfa traits related to some mineral concentration, the energy value and the potential nutritive capacity of these native alfalfa ecotypes.

EXPERIMENTAL

18 Landraces alfalfa cultivars were used¹⁴ for this study Adiguzel, Ahlat(2), Alaköy, Burcu, Çayirbasi(16), Dilburnu(6), Dönemeç(21), Ercis, Gülgören(23), Gülsinberk, Hidirkoy(13), Kasimoglu(11), Köprüler(12), Mahmudiye, Mollakasim(4), Otluca, Otluyazi(20). 20 Plants (*i.e.*, genotypes) per cultivar were grown in plastic pots (number 8) in green house and these single plants transplanted to the field on 10 May 2004 at Field Crop Department, Faculty of Agriculture, Atatürk University, Erzurum, Turkey (39°55'N lat... 41°16'E long and 1950 m above sea level). An experiment field was in a deep clay silt soil in a randomized complete block with three replicates. The location is arid characterized by dry, cool temperate summers and 187 mm rainfall during April-August. 46.5 % of the annual average rainfall. Weeds were controlled with hand weeding when necessary. On ten randomly chosen plant were cut at early flowering period and forage was dried, weight ground to pass a 1 mm grid. On all samples with dry weight higher than 4 g near infrared spectra (NIRS) were collected (NIR systems 6500, NIR systems Inc. Silver Spring, MD) between 1100 and 2500 nm at every 2 nm. NDF, ADF and ADL by the Van Soest methods¹⁵. Total nitrogen was determined according to the Kjeldahl method and crude protein percentage was calculated using the factor $6.25 \times N$. Determination of Mg, Cu, Zn, Mn, contents of the samples were carried out by atomic absorption spectrometry and that of Na and K by flame emission using a Perkin-Elmer 2380 atomic absorption spectrophotometer. P was determined by a colorimetric method¹⁶. Ash content was determined by heating overnight at 550 °C Energy value calculated by methods of Kirchgessner and Kellner¹⁷ equation used as follow: Net energy lactation (NEL), Dry matter (DM), Net energy gain (NEG), Total digestible nitrogen (TDN).

$$\text{NEL} = 1.037 - 0.0124 \times \text{ADF}$$

$$\text{NEG} = \{2.54 - (2.42 / (\text{NEL} \times 2.2))\} \times 2.2$$

$$\text{TDN} = 8 + 86 \times \text{NEL}$$

Data obtained from the experiment were analyzed by using SPSS 11.0 (1998) statistical program.

RESULTS AND DISCUSSION

Generally, there were considerable variations in chemical composition between genotypes (Table-1). The dry matter (DM) content in alfalfa ranged from 58.28-82.46 g kg⁻¹ Otluca cultivar had significantly ($p < 0.001$) higher DM and OM content than for the other genotypes in contrary, Mahmudiye

TABLE-1
SHOOT TISSUE CHEMICAL MINERAL AND CELL-WALL ENERGY CONCENTRATION FOR ALFALFA CULTIVARS

	CP	OM (%)	DM	Ash (%)	Cu (ppm)	Mn (ppm)	Zn (ppm)	Na (%)	K (%)	Mg (%)	NDF	ADL	ADF	NEL	NEG	TDN
Adigüzel	15.40	62.20	73.20	11.00	4.25	22.55	36.50	0.48	2.46	0.23	37.82	6.94	32.01	0.64	1.76	63.05
Ahlat(2)	15.82	57.03	72.61	15.57	3.50	50.10	30.25	0.56	2.41	0.24	42.97	7.56	34.62	0.61	1.72	60.26
Alaköy	13.77	54.63	63.82	9.19	9.55	27.35	24.55	0.46	2.34	0.23	45.33	8.83	34.62	0.61	1.72	60.26
Burcu	17.15	57.52	67.05	9.53	3.75	35.75	40.50	0.43	2.29	0.37	42.53	7.22	34.91	0.60	1.71	59.95
Çayırbaşı(16)	12.71	57.78	69.06	11.28	2.90	22.90	28.75	0.49	2.36	0.26	42.23	7.39	35.55	0.60	1.70	59.27
Dilburmu(6)	13.14	64.10	73.63	9.53	2.15	25.65	14.35	0.48	2.34	0.32	35.93	5.94	29.71	0.67	1.79	65.51
Dönemeç(21)	15.59	57.92	69.17	11.25	4.00	31.95	27.50	0.53	2.28	0.32	42.08	7.22	34.46	0.61	1.72	60.44
Ercis	15.83	57.74	72.89	15.15	3.25	85.55	42.10	0.77	2.49	0.39	42.29	6.49	34.17	0.61	1.73	60.75
Güllören(23)	15.68	53.82	65.51	11.69	7.30	45.55	36.00	0.45	2.30	0.37	46.18	8.44	38.53	0.56	1.65	56.10
Gülşenberk	14.96	58.53	68.05	9.52	3.25	64.25	37.20	0.42	2.29	0.39	41.42	8.16	35.11	0.60	1.71	59.75
Hidinköy(13)	13.80	59.37	72.47	13.10	2.80	54.70	28.05	0.66	2.10	0.30	40.63	7.86	31.83	0.65	1.76	63.24
Kasımoglu(11)	15.69	59.56	72.90	13.34	3.75	25.45	18.65	0.63	2.40	0.41	40.44	8.04	32.07	0.64	1.76	62.99
Köprüler(12)	15.76	56.60	68.76	12.15	8.45	32.45	21.85	0.43	2.07	0.24	43.46	8.45	36.41	0.59	1.69	58.35
Mahmudiye	13.47	48.69	58.28	9.59	4.10	42.85	28.65	0.36	2.58	0.34	51.19	6.91	31.76	0.64	1.76	63.32
Mollakasım(4)	15.12	58.07	67.38	9.31	2.95	20.10	28.10	0.44	2.46	0.22	41.93	7.57	34.29	0.61	1.72	60.62
Otluca	15.97	73.71	82.46	8.75	3.75	56.15	31.35	0.29	2.18	0.45	26.29	7.58	33.74	0.62	1.73	61.21
Otluyazı(20)	13.91	56.85	68.90	12.05	5.45	38.50	32.75	0.63	2.49	0.25	43.15	8.08	36.93	0.58	1.67	57.81
Ovakisla	14.25	53.41	67.06	13.65	2.85	13.65	27.40	0.43	2.42	0.32	46.63	3.52	40.51	0.53	1.61	53.98
Mean	14.89	58.20	69.62	11.42	4.33	38.64	29.69	0.49	2.34	0.31	41.80	7.34	34.51	0.61	1.72	60.38
LSD(0.01)	0.86	2.21	1.82	2.05	1.03	9.80	5.89	0.127	0.123	0.028	2.21	0.49	2.09	0.026	0.037	2.23

ecotype had the lowest OM content (48.69 %). Genotypes had significant variation in ash content Ahlat(2) ecotype had the highest (15.57 %) and Otluca had the lowest (8.75 %). Mg, K and Na, generally were similar to or higher than previously published reports, while concentrations of Cu, Mn and Zn were similar or to slightly below other reports^{5,18,19}. Differences in germplasm growth stage sampling may have biased differences in mineral content because concentration of Mg, Mn, Fe, Cu and Zn decline with advancing alfalfa maturity^{20,21}. Mineral concentration among four alfalfa germplasm studied by Townsend *et al.*²² had similar result with present study.

NDF contents of cultivars were significantly differ ($p > 0.001$). Studied of NDF content varied with in the range of 26.29-46.18 %. Gulgoren(23) genotype had the higher NDF content (467.18 %) than that of the other genotypes. The average ADL of the variation was 7.34 % and highest lignin was measured at Alakoy genotypes (8.83 %), also Otluyazi, Kopruler(12) and Gulgoren(23) had higher lignin, respectively. ADF contents ranged from 29.71 to 40.51 %. Ovakisla genotype had the highest ADF contents. There were significant differences between alfalfa cultivars. Average NEL was 0.61 Mcal. NEG of cultivars ranged from 1.61 to 1.79 Mcal and TDN ranged 53.98-65.51 %. NDF reflects the bulkiness of a forage because forage fiber is bulky, there is a limit to amount of NDF that will fit into a animals rumen (first stomach) when that limit is reached she will stop eating. The proportion of NDF to body weight (BW) is an important fundamental relationship. We can estimate maximum footage dry matter intake. High NDF gives low intake forage. Energy content of forage often is estimated from ADF content. Energy can be expressed different energy sources as digestible energy, metabolic energy of lactation, net energy of gain. The basic assumption is that higher forage has low ADF and NDF compared with to low quality forage. High quality forage digested more completely and has higher energy values²³. But the identification and development of high yielding, highly digestible cultivars are complicated by the negative relationship between digestibility's and forage yield²⁴. Single linkage dendrogram indicated that Dilburnu(6), Mahmudiye, Otluca and Ovakisla cultivar differed than the other genotypes. On the other hand Dönemeç(21), Mollakasim (4) and Cayirbasi(16) much more differed as a single grouped than the remained genotypes (Fig. 1). A wide range of variation for digestibility could be found at the individual level, as for the others traits. NDF reflects the bulkiness of a forage because forage fiber is bulky, there is a limit to amount of NDF that will fit into an animals rumen (first stomach) when that limit is reached she will stop eating. High quality forage digested more completely and has higher energy values. The chemical composition of alfalfa (*Medicago sativa* L.) and two grasses, reed canary grass (*Phalaris arundinacea* L.) and brome grass (*Bromus inermis* Leys.), studied by

Thender and Westherlund²⁵. They stated that chemical composition of the whole plant consequently changes, as the chemical composition differs between various anatomical parts. In addition to these changes, there are also changes in the cell-wall composition during growth. These differences were larger in alfalfa leaf and stems. Data on quality changes of leaves and stems of modern alfalfa cultivars subject to varying harvest regimes. Leaf NDF concentration and digestibility typically decline slowly with increasing maturity, while stem NDF and ADF concentration increase⁶.

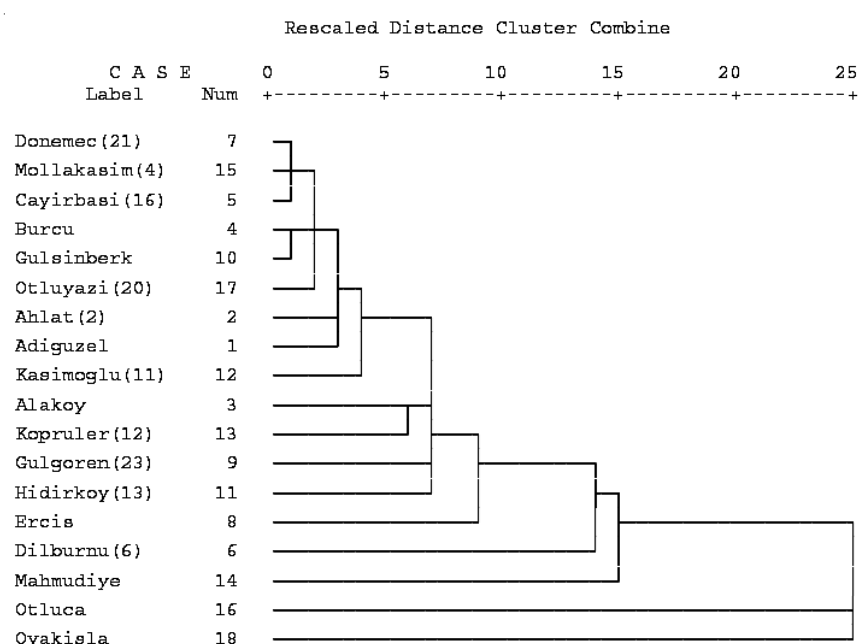


Fig. 1. Single linkage dendrogram of the alfalfa cultivars

There were significant phenotypic correlations between some of the mineral and cell wall properties (Table-2). There was not any significant correlation between CP and the observed properties. A significant negative correlation observed between Cu and DM (-0.361*) and positive correlation with ADL (0.526**). The Mn contents were positively correlated with Zn (0.409*) and Mg (0.395*). There were highly significant positive correlation between Na and crude ash (0.740**) and negative significance observed between K and OM (-0.358*), also crude ash and NEG (-0.330*). NDF was highly significantly correlated with ADF (0.452**) but negatively correlated with DM (-0.881**), NEL (-0.465**), NEG (-0.449**) and with TDN (-0.451**). There was not any significant correlation between ADL and the observed properties with exception of Cu. There were highly negative

TABLE-2
SIMPLE CORRELATION COEFFICIENT OF CHEMICAL MINERALS AND CELL-WALL
CONCENTRATION IN ALFALFA CULTIVARS

	CP (%)	Cu (ppm)	Mn (ppm)	Zn (ppm)	Na (%)	K (%)	Mg (%)	Ash (%)	NDF	ADL	ADF	OM (%)	DM	NEL	NEG
Cu (ppm)	0.059	1													
Mn (ppm)	0.287	-0.111	1												
Zn (ppm)	0.281	-0.013	0.409†	1											
Na (%)	0.106	-0.271	0.404	-0.100	1										
K (%)	-0.232	-0.195	-0.112	0.208	0.061	1									
Mg (%)	0.259	-0.248	0.395†	0.160	-0.130	-0.077	1								
Ash (%)	0.236	-0.202	0.352	-0.047	0.740‡	0.115	-0.095	1							
NDF	-0.163	0.254	-0.070	0.035	0.134	0.357	-0.257	0.299	1						
ADL	0.096	0.526‡	0.176	0.050	-0.061	-0.217	-0.057	-0.176	0.002	1					
ADF	0.102	0.269	-0.023	0.236	-0.081	0.097	0.006	0.321	0.451‡	-0.035	1				
OM (%)	0.166	-0.254	0.069	-0.035	-0.132	-0.358†	0.256	-0.297	-1.000	-0.004	-0.448	1			
DM	0.286	-0.361†	0.244	-0.059	0.228	-0.312	0.216	0.187	-0.881‡	-0.091	-0.303	0.883‡	1		
NEL	-0.104	-0.239	0.019	-0.251	0.079	-0.112	-0.019	-0.319	-0.465‡	0.065	-0.997‡	0.462‡	0.318	1	
NEG	-0.078	-0.236	0.055	-0.195	0.076	-0.127	-0.018	-0.330†	-0.449‡	0.075	-0.994‡	0.447‡	0.297	0.991‡	1
TDN	-0.102	-0.269	0.023	-0.236	0.081	-0.097	-0.006	-0.321	-0.451‡	0.035	-1.000	0.448‡	0.303	0.997‡	0.994‡

†Correlation is significant at the 0.05 level.

‡Correlation is significant at the 0.01 level.

significant correlation between ADF and NEG (-0.997**) and TDN (-0.994**). Highly significant positive correlation observed between OM and DM (0.883**), NEL (0.462**), NEG (0.447**) and TDN (0.448**). We observed a greater proportion of significant phenotypic correlation between minerals (Table-3) in this study than reported in earlier studies^{22,26}.

TABLE-3
RECOMMENDED MINERAL CONCENTRATION IN ALFALFA SHOOT
TISSUE FOR OPTIMUM ALFALFA GROWTH AND TO SATISFY
THE DAIRY REQUIREMENTS OF A MODERATELY
LACTATING DAIRY COW²²

Minerals	Alfalfa requirement (g kg ⁻¹)	Dairy cow requirement (g kg ⁻¹)
Potassium	20.0-35.0	9.00
Magnesium	3.0-10.0	2.00
Sodium	0.03-0.08	-
Copper	0.01-0.03	0.01
Manganese	0.03-0.25	0.04
Zinc	0.02-0.07	0.04

Mineral concentration reported in Table-1 were considered in relation to lactating daily cow requirements (Table-3) because their mineral requirements are generally representative of the mineral requirements for various classes of animals^{22,27}. Shoot tissue concentrations of Cu, Mn, Na, K and Mg in these populations generally were sufficient to meet dairy cow requirements (Table-3). K concentrations were sufficient in these populations; alfalfa often accumulates K in excess of plant and animal requirements²². K fertilizer is a common soil amendment used to increase forage yield to enhance winter hardness in colder climates and to promote rapid regrowth following cutting²⁸. It can also increase K and Mn accumulation and decrease Mg, Na, Cu, Zn concentration. High K levels in alfalfa forage may interfere with Mg utilization by cattle, thereby causing hypomagnesaemia²⁹.

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