

DETERMINATION OF CHEMICAL COMPOSITION AND RUMEN DEGRADABILITY OF DRY MATTER AND CELL WALL OF *VICIA CANESCENS* AT DIFFERENT PHENOLOGICAL STAGES IN NEOR Rangelands OF ARDABIL PROVINCE

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ABSTRACT: This study was carried out in order to determine the chemical composition and rumen degradability of dry matter and cell wall of *Vicia canescens* at different phenological stages in Neor rangelands of Ardabil province. The samples of *Vicia canescens* was collected from two sites and three phenological stages of growth (vegetative, flowering and seed ripening). Rumen degradability of dry matter and cell wall of *Vicia canescens*.L at different phenological stages using nylon bag method and two castrated and ruminally fistulated male Moghani sheep were applied. There were significant differences between the amount of DM, NDF, ADF, Ash contents and DM degradability and also between the amount of degradability parameters of DM, NDF and ADF including rapid degradable portion(a), slow degradable portion (b), constant rate of degradability (c)and effective degradability (ED)in different phenological stages ($P<0.01$). The results showed that with herb maturity ruminal digestibility of dry matter and degradability parameters of DM, NDF and ADF were significant decreased and DM, NDF and ADF values increased significant ($P<0.01$). The Highest and lowest amount “a” and “b” parameters of DM were obtained at vegetative stage and seed ripening respectively. Also, altitude effect on chemical compositions, dry matter degradability and studied degradability parameters were significant ($P<0.01$). Generally, it is concluded that maturity of plant led to decrease in the amount of degradability consequently diminish nutritive value of plants.

Key words: Degradability, Nutritional value, Neor rangelands, Phenological stages.

INTRODUCTION

Forage plants are an important component of agricultural systems. The plants rangelands play importance role in animal productions, soil conservation and local climate balance. Due to shortage of animal protein and necessity of increasing animal productions by this available local source is essential that present adequate information of nutritional value this feed sources [3]. In order to achieve to this purpose the determination of nutritive value and dry matter, cell wall rumen degradability of rangeland plants is more important. In generally, the nutritional value of range plants is expressed on the chemical composition, digestibility and availability of nutrient [26], that influenced by different factors such as variety, species, phenological stages, amount of leaf, soil and climate. The phenological stages effect on hay quality in cutting time more of than other factors [17, 22]. Chen et al [7] and Grant et al [13] showed that the phenological stage was induced the most of different in the acid detergent fibre (ADF) content plants. By increasing age plant its requirements for skeletal tissue reach the highest point as a result the amounts of structural carbohydrates such as cellulose, hemi-cellulose and lignin construction should be greater[13].The digestibility of neutral detergent fibre (DNDF) of leguminous plants is high in vegetable stage but decrease as linear and moderate by progressing maturity [15]. Also, the lowest digestibility of dry matter (DMD) for seed ripening stage and the highest DMD for vegetative stage is observed in plants rangelands [8]. There are about 166 *Vicia* species in the world that account of subspecies it achieve more than 300 species [23].

Vicia canescens is belonging to vicia gregaria groups and Fabaceae family [9]. This study was carried out in order to determine the chemical composition and rumen degradability of dry matter and cell wall of *Vicia canescens* at different phenological stages in Neor rangelands of Ardabil province.

METHODS AND MATERIALS

Sampling and Chemical Composition Analysis

The sampling of *Viciacanescens* was carried out by systematic (shape of M letter) as Ghorbani et al [11] from two site and three phenological stages of growth (vegetative, flowering and seed ripening) in Neor altitudes rangelands of Ardabil cityenvirons. The first and second sites were nominated at the altitude range of 1500-1700 meter and 1900-2100 from sea level. The samples were dried in the air room temperature, then milled and passed a sieve 2 mm pore. The air-dried intact and digesta samples were analyzed for NDF and ADF using ANKOM apparatus and recommended method of Van Soest et al [32] and DM, EE, Ash and OM were determined according to the method of AOAC [4].

In situ degradability

Rumen degradability of dry matter and cell wall of *Viciacanescens*.L at different phenological stages using nylon bag method and two castrated and ruminally fistulated male Moghani sheep were applied. The air-dried intact samples were passed through a sieve 50 μm pore in order to remove soil and dust particles and to reduce in particle losses of samples. Then, amount of 4-5 grams samples were placed in each nylon bag (10 \times 5cm, 50 μm pore and 5 mg surface bag/cm²) and appended through the fistula into rumen. For obtain digestibility data of samples used 0, 2, 4, 8, 16, 24, 48 and 72 hour of ruminal incubation. All bags after removing from rumen at certain hours were washed and dried in a forced air oven at 65°C for 48 hours. Then, bags weighed and calculated degradability for DM, OM, NDF and ADF. Disappearance values of material at certain times and the parameters a, b, and c was estimated according to Ørskov and McDonald[27] $P = a + b(1 - e^{-ct})$, where P is the ruminal disappearance (g kg⁻¹) at time t, a is the soluble fraction (g kg⁻¹), b is the slowly degradable fraction (g kg⁻¹), c is the rate at which the b fraction is degraded (% h⁻¹) and t is time relative to incubation (h). Ruminal effective degradability (ED) was estimated as $ED = a + bc/(c + k)$, where k is the rumen flow rate (0.03 h⁻¹).

Calculations and statistical analysis

The collected data were analyzed using factorial methods, CRD design ANOVA method to compare for difference between treatments and Mixed model for to compare effects altitude and growth stages. For comparing average values of a, b and c parameters was used Duncan's Multiple Range test for least significant differences at the 5 percent probability level [30]. Statically model used for analysis of data as was:

$$Y_{ijk} = \mu + L_i + P_j + (L \times P)_{ij} + e_{ijk}$$

Where μ total mean, L_i altitude effect (i=1 &2), P_j growth stage effect (j= 1, 2&3), $(L \times P)_{ij}$ interaction of altitude in growth stage and e_{ijk} effect of test error.

RESULTS AND DISCUSSION

Table 1 shows the chemical composition of vetch plant. The amounts NDF, ASF, OM and DM increased and Ash decreased with progression of plant maturity. The effect of growth stage and altitude were significant on chemical composition (P<0.01). The interaction of altitude in growth stage were significant on fat, OM, ADF and DM contain of plant (P<0.05). The highest NDF was related to third stage of first site and the lowest NDF was belonged to first stage of second site. However, there were significant difference between growth stages and two sites undertake this study (P<0.01). In both altitudes were increased DM, NDF and ADF values with progression of plant growth and maturity that were similar with report of Hochen smith et al [14]. That would be due to increasing of roughage materials cell wall and ratio NDF to ADF and finally lignifications of plant with progression its growth and maturity. The concentration of cell wall material in roughages is variable and increases with stage of plant maturity. Lignin concentration also increases with maturation of plants and is associated with reduced cell wall digestibility and degradability [19, 21, 33]. Table 2 shows DM degradability in various times of ruminal incubation. The results obtained were observed the highest DM degradability for first stage of second site and the lowest it for third stage of first site (P<0.01). The effect of altitude, growth stage and interaction of altitude in growth stage were significant on times of ruminal incubation (P<0.01).

In both sites were significant decreased rate and amount DM degradability with progression of plant growth and maturity that due to either high soluble carbohydrates or organic matter [1]. Also, in the second site was determined higher degradability probability being due to lag maturity and freshness and blossoming plant [28]. Table 3 shows the degradability parameters of the vetch plant DM obtained from the fitted values. There was significant difference between growth stages both sites ($P < 0.01$).

Table 1. Mean chemical composition (%) of vetch plant (*Vicia canescens*.L) at two site and three phenological stages of growth.

Items		DM	OM	Ash	EE	ADF	NDF
The first site	First stage	95.37 ^d	90.96 ^c	9.04 ^a	6.36 ^{ab}	36.06 ^d	47.98 ^d
	Second stage	96.69 ^b	91.93 ^b	8.07 ^b	4.01 ^c	41.55 ^c	49.54 ^c
	Third stage	97.14 ^a	92.13 ^b	7.87 ^b	6.25 ^b	48.09 ^a	53.59 ^a
The second site	First stage	94.03 ^e	91.00 ^c	9.00 ^a	6.45 ^a	33.39 ^e	46.01 ^e
	Second stage	96.00 ^c	92.09 ^b	7.91 ^b	2.71 ^d	37.06 ^d	46.79 ^e
	Third stage	96.53 ^b	93.80 ^a	6.20 ^c	3.89 ^c	43.72 ^b	50.54 ^b
SE		0.09	0.15	0.15	0.36	0.34	0.32
Significant	Altitude	**	**	**	**	**	**
	Growth stage	**	**	**	**	**	**
	Altitude × Growth stage	*	**	**	**	*	n.s

SE= Standard error, Means in the same column with the different superscript are significantly different {($P < 0.05$)}. n.s= Non significant, ** = as 'statistically significant ($p = 0.01$ level) and * = as 'statistically significant ($p = 0.05$ level)

Table 2. Mean value of ruminal DM degradability (%) of vetch plant (*Vicia canescens*.L) at two site and three phenological stages of growth and different hours of incubation.

Items		0	2	4	8	16	24	48	72
The first site	First stage	35.05 ^b	45.21 ^b	57.15 ^c	60.22 ^b	65.53 ^b	74.12 ^b	78.08 ^c	82.09 ^b
	Second stage	32.58 ^d	42.09 ^e	55.04 ^d	58.08 ^d	62.12 ^e	70.08 ^e	75.18 ^d	79.00 ^f
	Third stage	30.29 ^f	41.07 ^f	52.14 ^f	56.15 ^e	60.10 ^f	67.42 ^f	72.09 ^f	76.51 ^e
The second site	First stage	36.22 ^a	47.18 ^a	59.14 ^a	63.10 ^a	67.16 ^a	77.10 ^a	80.12 ^a	85.15 ^a
	Second stage	33.32 ^c	44.08 ^c	57.58 ^b	60.12 ^b	65.06 ^c	72.31 ^c	78.27 ^b	81.07 ^c
	Third stage	31.10 ^e	42.82 ^d	54.77 ^e	58.52 ^c	62.80 ^d	70.34 ^d	74.33 ^e	79.40 ^d
SE		0.08	0.04	0.05	0.04	0.04	0.05	0.06	0.05
Significant	Altitude	**	**	**	**	**	**	**	**
	Growth stage	**	**	**	**	**	**	**	**
	Altitude × Growth stage	*	*	**	**	**	**	**	**

SE= Standard error, Means in the same column with the different superscript are significantly different {($P < 0.05$)}. n.s= Non significant, ** = as 'statistically significant ($p = 0.01$ level) and * = as 'statistically significant ($p = 0.05$ level)

The highest soluble fraction (a) related to first stage of second site (46.55 %) and lowest it was belong to third stage of first site (40.95%). Due to higher soluble carbohydrates during the earlier growth stages of plants as a result of the lagging of plants maturity in high altitudes than low altitudes DM and OM degradability of vetch plant observed more for second site. The greatest b fraction of vetch plant related to the first growth stage of second site and so there was significant difference between growth stages ($P < 0.01$). Generally, degradability is affected by cell wall as higher cell wall lower degradability. This finding is in agreement with that of the previous studies as reported by [5]. Ruminal effective degradability (ED) had significant difference between three growth stages and two sites in passage rate of 2, 5 and 8%/h ($P < 0.01$). The highest values of ED were in first stage of second site and the lowest value of ED observed in third stage of first site. Hoffman et al [15] found that DM effective degradability reduced with progression of plant growth and maturity that was in agreement with the results of present study for DM, NDF, ADF and OM effective degradability. The reason for this decline could be increasing NDF, ADF and ADIN as have been reported previously [6, 10].

Cell wall degradation parameters of vetch plant for NDF are presented in table 4. Results in the current study, further revealed that, most of the DM, and cell wall (NDF, ADF) components were lost within the first 8 and 48 h after incubation (table 2) suggesting high ruminal degradability of this material. The effect of growth stages and both sites was significant on rate of degradability ($P < 0.05$) and the other obtained degradability parameters ($P < 0.01$). The interaction of altitude in growth stage was significant on b fraction, ED at the rumen flow rate (0.03 h^{-1} , 0.05 h^{-1} and 0.08 h^{-1}) of vetch plant ($P < 0.05$) but it was not significant for soluble fraction (a) and rate of degradability (c).

Table 3. Dry matter degradation kinetics (%) of vetch plant (*Vicia anescens*.L) at two site and three phenological stages of growth.

Items		a (%)	b (%)	c (/h)	PD (%)	ED (0.02/h)	ED (0.05/h)	ED (0.08/h)
The first site	First stage	44.49 ^b	36.67 ^b	0.062	81.17 ^b	72.43 ^b	65.08 ^b	60.80 ^b
	Second stage	42.46 ^c	35.71 ^d	0.06	78.17 ^d	69.33 ^d	62.07 ^c	57.92 ^c
	Third stage	40.95 ^f	34.51 ^f	0.06	75.47 ^f	66.85 ^f	59.78 ^f	55.78 ^f
The second site	First stage	46.55 ^a	37.30 ^a	0.06	83.85 ^a	74.92 ^a	67.40 ^a	63.05 ^a
	Second stage	44.32 ^c	36.20 ^c	0.06	80.52 ^c	71.80 ^c	64.53 ^c	60.27 ^c
	Third stage	42.69 ^d	35.21 ^e	0.06	77.92 ^e	69.42 ^e	62.33 ^d	58.20 ^d
SE		0.06	0.05	0.001	0.06	0.02	0.02	0.03
Significant	Altitude	**	**	**	n.s	**	**	**
	Growth stage	**	**	**	n.s	**	**	**
	Altitude × Growth stage	n.s	*	n.s	n.s	*	n.s	**

“a” Fraction that is soluble or immediately degraded. “b” Potentially degradable but insoluble fraction. “PD”:

Maximum potential degradability. “c”: Rate of degradation of the sample “b” fraction (per h). ED: Effective degradability values at 0.02, 0.05 and 0.08 per h outflow rate. SE= Standard error, Means in the same column with the different superscript are significantly different ($P < 0.05$). n.s= Non significant, ** = as 'statistically significant ($p = 0.01$ level) and * = as 'statistically significant ($p = 0.05$ level)

Table 4. NDF degradation kinetics (%) of vetch plant (*Vicia anescens*.L) at two site and three phenological stages of growth.

Items		a (%)	b (%)	c (/h)	PD (%)	ED (0.02/h)	ED (0.05/h)	ED (0.08/h)
The first site	First stage	30.35 ^b	33.99 ^b	0.05 ^a	64.30 ^c	54.00 ^b	46.60 ^b	42.75 ^b
	Second stage	28.07 ^c	31.59 ^c	0.04 ^a	59.70 ^d	48.85 ^c	41.85 ^d	38.35 ^d
	Third stage	21.98 ^e	33.67 ^b	0.04 ^a	55.65 ^f	44.35 ^d	36.85 ^f	33.10 ^f
The second site	First stage	34.00 ^a	36.29 ^a	0.04 ^a	70.30 ^a	57.90 ^a	49.80 ^a	45.80 ^a
	Second stage	30.37 ^b	35.71 ^a	0.03 ^b	66.05 ^b	52.85 ^b	44.80 ^c	41.00 ^c
	Third stage	23.73 ^d	33.83 ^b	0.04 ^b	75.55 ^e	46.55 ^d	39.05 ^e	35.30 ^e
SE		0.23	0.38	0.002	0.41	0.13	0.11	0.13
Significant	Altitude	**	**	*	**	**	**	**
	Growth stage	**	*	*	**	**	**	**
	Altitude × Growth stage	n.s	**	n.s	*	**	**	*

“a” Fraction that is soluble or immediately degraded. “b” Potentially degradable but insoluble fraction. “PD”:

Maximum potential degradability. “c”: Rate of degradation of the sample “b” fraction (per h). ED: Effective degradability values at 0.02, 0.05 and 0.08 per h outflow rate. SE= Standard error, Means in the same column with the different superscript are significantly different ($P < 0.05$). n.s= Non significant, ** = as 'statistically significant (at the $p = 0.01$ level) and * = as 'statistically significant (at the $p = 0.05$ level)

The greatest b fraction of cell wall vetch plant related to the first growth stage of second site and the lowest it for second growth stage of first site. In both sites were significant in the first and second plant growth stage from b fraction point of view ($P < 0.01$) but it not observed in the third plant growth stage. Potential degradability (PD) was significant between plant growth stages.

The highest values of PD were in first stage of second site and its lowest value obtained in third stage of first site. In both altitudes were significantly decreased the values of NDF and ADF potential degradability with advancing plant maturity of vetch plant ($P < 0.01$). Resistance to degradation of the cell wall depended on the chemical composition, plant anatomy, morphology, the thickness of the cell wall, low mesophyll, high xylem groups and high lateral areas in cell wall tissue [24]. The relatively low content of fibre (NDF: 46-47% and ADF: 33-36%) and the high degradability observe in the current study (Table 1&4) are positive indicators of the good feeding value of vetch plant (*Vicia anescens*). Thus, the low level of fibre can facilitate the colonization of the vetch feed by the rumen microbial population, which in turn might induce even higher fermentation rate, therefore improving its digestibility [32]. The rumen degradation parameters of ADF vetch plant are presented in Table 5. The effect of growth stages and altitude sites was significant on all of ADF degradability parameters ($P < 0.01$). The interaction of altitude in growth stage was significant on all the parameters excluding rate of degradability (c) of vetch plant ($P < 0.01$). The greatest a fraction of vetch plant related to the first growth stage of second site but it was at least for the third growth stage of first site. In both sites were significant decreased a fraction with progression of plant growth and maturity ($P < 0.01$). Messman et al [25] reported that ADF degradability is altered by with progression of plant growth and maturity and they found that rate of degradability (c) of vetch plant significant decreased with progression of plant growth. Also, effective degradability (ED) at the different rumen flow rate significant decreased with advancing maturity for both sites ($P < 0.01$) and the highest amount was related to the first growth stage of second site.

Table 5. ADF degradation kinetics (%) of vetch plant (*Vicia canescens*.L) at two site and three phenological stages of growth.

Items		a (%)	b (%)	c (/h)	PD (%)	ED (0.02/h)	ED (0.05/h)	ED (0.08/h)
The first site	First stage	27.22 ^c	33.95 ^c	0.06 ^a	61.15 ^c	52.55 ^b	45.55 ^b	41.55 ^b
	Second stage	24.12 ^d	30.64 ^d	0.06 ^{ab}	54.75 ^d	46.60 ^e	40.20 ^e	36.65 ^e
	Third stage	20.96 ^e	30.93 ^d	0.05 ^b	51.90 ^f	43.00 ^f	36.45 ^f	33.00 ^f
The second site	First stage	30.22 ^a	35.57 ^a	0.05 ^b	65.80 ^a	55.20 ^a	47.50 ^a	43.45 ^a
	Second stage	27.94 ^b	34.54 ^{bc}	0.04 ^c	62.50 ^b	51.50 ^c	43.95 ^c	40.05 ^c
	Third stage	26.64 ^c	35.25 ^{ab}	0.03 ^d	61.85 ^{bc}	48.80 ^d	40.90 ^d	37.10 ^d
SE		0.21	0.23	0.002	0.22	0.08	0.05	0.04
Significance	Altitude	**	**	*	**	**	**	**
	Growth stage	**	*	*	**	**	**	**
	Altitude × Growth stage	**	**	n.s	**	**	**	**

“a” Fraction that is soluble or immediately degraded. “b” Potentially degradable but insoluble fraction. “PD”:

Maximum potential degradability. “c”: Rate of degradation of the sample “b” fraction (per h). ED: Effective degradability values at 0.02, 0.05 and 0.08 per h outflow rate. SE= Standard error, Means in the same column with the different superscript are significantly different ($P < 0.05$). n.s= Non significant, ** = as 'statistically significant (at the p = 0.01 level) and * = as 'statistically significant (at the p = 0.05 level)

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