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Nutritive value of on-farm common vetch-oat hays. II. Ruminal degradability of dry matter and crude protein

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Abstract — Rumen degradation characteristics and effective degradability (ED) of dry matter (DM) and crude protein (CP) of 11 samples of common vetch-oat hay were determined using the nylon bag technique in three rumen fistulated wethers. Animals were fed at an intake level of 40 g DM·kg^{-1}·BW$^{0.75}$ with a mixed diet of vetch-oat hay and concentrate (2:1 on DM). Mean values of ED of DM and CP, calculated for rumen outflow rates determined in each sheep, presented a moderate variation (mean = 65.8%; CV = 7.9% for DM and mean = 79.3%; CV = 4.7% for CP). Vetch proportion in hay was positively correlated with ED of DM (r = 0.412; $P < 0.05$) and ED of CP (r = 0.583; $P < 0.001$). Predictive equations of ED from chemical composition were obtained using the step wise procedure. The contents of neutral detergent fibre and acid detergent lignin explained 79.4% of the total variation of ED of DM. The best prediction of ED of CP was obtained with the concentration of cellulose as an independent variable ($r^2 = 0.839$). Degradation studies in these hays also indicated that the content of bypass protein, potentially digestible in the small intestine, was low. Thus, most N available to animals was derived from rumen microbial synthesis.

forage quality / vetch-oat hay / rumen degradability / protein / sheep

Résumé — **Valeur nutritive des foins de vesce-avoine pour les ovins. II. Dégradabilité dans le rumen.** Les caractéristiques de dégradation et la dégradabilité théorique (DT) dans le rumen de la matière sèche (MS) et des matières azotées totales (MAT) de 11 foins de vesce-avoine ont été mesurées avec des sachets de nylon sur trois moutons munis d'une canule du rumen. Les animaux ont été nourris avec une ration composée de foin de vesce-avoine et d'aliment concentré dans des

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proportions 2:1 (sur MS) à un niveau de 40 g MS·kg^{-1}·P^{0,75}. Les valeurs de DT, calculées avec des taux de sortie des particules du rumen mesurés pour chaque animal, ont été peu variables tant pour la MS (moyenne = 65,8 % ; CV = 7,9 %) que pour les MAT (moyenne = 79,3 % ; CV = 4,7 %). La proportion de vesce du foin a été positivement reliée avec la DT de la MS (r = 0,412 ; $P < 0,05$) et des MAT (r = 0,583 ; $P < 0,001$). Des équations de prédiction de la DT à partir de la composition chimique ont été obtenues. Dans le cas de la MS, les teneurs en parois cellulaires et en lignocellulose expliquent 79,4 % de la variation totale observée pour la DT. En revanche, la meilleure prédiction de la DT des MAT a été obtenue avec la teneur en cellulose comme variable indépendante ($r^2 = 0,839$). Les études de dégradation ont aussi montré que ces foins ont une faible teneur en protéine non dégradée dans le rumen et potentiellement digestible dans l'intestin. De cette façon, la plus grande partie de l'azote disponible pour l'animal est fournie par la synthèse microbienne dans le rumen.

qualité des fourrages / foin de vesce-avoine / dégradabilité dans le rumen / matières azotées / ovins

1. INTRODUCTION

A non-irrigated intercrop of annual forage legumes and winter cereals for hay production is a traditional and extended cropping practice in the Mediterranean basin. In these areas, the production of these hays represents an important resource allowing to meet the forage deficit derived from the seasonality and low-quality of the available natural resources (natural pasture, cereal stubble and straw, shrub-steppe formations). Moreover, these crops have positive ecological functions when rotated with winter cereal grains, stabilising grain yields by breaking the pest and disease cycles and reducing soil nitrogen depletion [4].

Among these hays, the species most usually employed are common vetch (*Vicia sativa* L.) and oat (*Avena sativa* L.) [3]. The intercropping abilities of common vetch and oat have been reported [5] but studies on the nutritive value of vetch-oat hays are scarce. In particular, there is an important lack of information related to their rumen protein degradability, which is a parameter necessary for the application of new schemes of protein rationing for ruminants. Between the possible factors that could influence protein degradability of forages, the stage of maturity at harvest and forage species [1] seem to have a superior importance in a grass-legume forage such as vetch-oat hays.

The objectives of this research were: (i) to obtain information on the rumen degradability of common vetch-oat hays and (ii) to study the possible relationship of the botanical and chemical composition of these hays on their degradation.

2. MATERIALS AND METHODS

2.1. Study sample

A total of 11 common vetch-oat hays from different farms within the Castile-La Mancha region (South-central Spain) were obtained. Sampling procedures, stage of maturity and chemical and botanical composition of these hays have been reported in the companion paper [9] using the same identification, that was assigned by increasing the vetch proportion in hays, which ranged from 125 to 917 g·kg^{-1} DM. Nevertheless, one of the 12 samples included in the previous work was lost contingently before degradation assays, so, the H3 sample was not included in the present study.

2.2. Animals and feeding

Three caged male castrated sheep (2–3 years old) of the Manchega breed provided with a rumen cannula (inside diameter 80 mm) were used in the experiment.

Animals were fed a mixed (2:1) hay to concentrate diet containing (on DM) 14% CP and 38% NDF. A common vetch-oat hay was used as dietary hay (55.2 vetch, 42.6 oat and 2.2% weed on DM) and the concentrate pellet contained (on DM basis) 19.2% CP and 29.7% NDF. The diet was distributed at a level of 40 g DM·kg^{-1}·BW$^{0.75}$ in two equal meals at 09.00 and 17.00 h.

2.3. Ruminal degradation of DM and CP

The hays were incubated in nylon blutex bags (pore size 46 µm, (reference 120T, Tissages Tissues Techniques, Villeneuve la Garenne, France) of 11 cm × 7 cm (inner dimensions), heat-sealed (Preci-Pack P30N, Dover Pack S.A., Barcelona, Spain) and filled with approximately 3 g (air dry basis) of feed samples ground through a 2 mm screen. Two series of hay samples were incubated in the rumen of each animal. Within each series, rings of 12 bags were sequentially incubated. Each ring encompassed a randomly associated pair of hay samples that was introduced in the rumen just after the 09.00 h meal and removed after 3, 6, 12, 24, 48 and 72 h. After incubation, the bags were removed, washed with tap water, and stored, frozen. After being defrosted, the bags were washed 3 times for 5 min in a turbine washing machine, dried in an air-forcing oven for 48 h at 80 °C and weighed for DM determination. Residues were homogenised and analysed for N [2]. Three additional bags of each hay were reserved for the zero incubation which involved a washing procedure without prior rumen incubation.

The pattern of DM or CP disappearance (d) with the incubation time (t) was described for each hay and animal using the model proposed by Ørskov and McDonald [11]: $d = a + b\,(1–e^{-kd*t})$. In this model, the constant a (%) represents the soluble or rapidly degradable fraction and b (%) represents the non-soluble degradable component, which disappears at a constant fractional rate k_d (h^{-1}). The undegradable fraction (u, %) was estimated as $100 – (a + b)$ and ruminal effective degradability (ED, %) was determined as: $ED = a + (bk_d / (k_d + k_p))$.

For this last purpose, between both incubation series of the hay study sample, the rate constant describing the passage of dietary hay particles (k_p) from the rumen was determined by supplying, immediately before the first daily meal, a pulse dose (50 g) of labelled hay in each animal's stall. Previously, the hay was washed with sodium lauryl sulphate in an automatic washing machine as proposed by Uden et al. [16] to eliminate the soluble components and labelled with ytterbium (Yb). Labelling was done by immersion for 24 h as previously described [7], using a dose of 10 mg Yb per g of dry feed residue. A total of 19 samples of faeces was obtained from the rectum of each animal, the first before supplying the marked hay and the rest between 20 and 144 h afterwards. These samples were dried, milled and incinerated at 550 °C and then digested by boiling with a solution of 1.5 M HNO_3 and KCl (3.81 g·L^{-1}). The resultant solutions were analysed for Yb by atomic absorption spectrometry (Smith-Hieftje 22, Thermo Jarrel Ash, MA, USA) using predosed samples of faeces to prepare common-matrix standards. The pattern of Yb concentrations in faeces with time was described by fitting the model proposed by Dhanoa et al. [6] and rate constants derived from the decreasing phase of concentrations were used as k_p values for all the tested hays.

2.4. Statistical analysis

A least-squares non-linear iterative process was used to fit the modelling curves of DM and CP disappearance and faecal marker excretion by the NLIN procedure of the Statistical Analysis System for Windows software, version 6.08 [15]. This same program was employed to perform uni- and multivariate regression analyses between ruminal degradation characteristics and botanical and chemical composition of the hays.

3. RESULTS

Ruminal degradation characteristics of DM and CP of the hays are presented in Tables I and II, respectively. Estimates of ED were obtained with a k_p value of 2.91 ± 0.32%·h^{-1}, derived from the three employed sheep. Both in DM as in CP results, the undegradable fraction (*u*) and the fractional rate of degradation (k_d) showed higher variations than both the soluble (*a*) or potentially degradable (*b*) fractions. The lowest relative dispersion was observed for ruminal ED of both DM and CP, which presented mean values of 65.8% and 79.3%, respectively.

Degradation kinetic characteristics of DM and CP were regressed on botanical and chemical composition (Tabs. III and IV, respectively).

Correlations of most degradation characteristics of DM, except for the *u* fraction, and vetch proportion in hays were significant. All these coefficients of correlation were positive except for the *b* fraction (r = –0.629, $P < 0.001$). Conversely, as the vetch proportion increases the rate of degradation of this fraction also increases. This parameter has the closest correlation to the vetch proportion (r = 0.821, $P < 0.001$). Among the chemical parameters, NDF content showed the closest correlation with ED of DM and its degradation characteristics. Conversely, the *u* fraction was mainly linked to the ADIN proportion (r = 0.657, $P < 0.001$) and ADL content (r = 0.607, $P < 0.001$).

For CP degradation (Tab. IV), the vetch proportion in hay did not alter the *a* and *b* fractions nor the degradation rate. Vetch proportion in hay, however, was negatively correlated with the undegradable fraction (r = –0.508, $P < 0.01$) and positively correlated with the ruminal ED of CP (r = 0.583, $P < 0.001$). Significant correlations with chemical parameters were mainly obtained for k_d, *u* and ED. Degradation rate was negatively related to the NDIN proportion (r = –0.698; $P < 0.001$). The *u* fraction was negatively correlated with the hay CP

Table I. Degradation kinetics and effective degradability (ED) of dry matter of common vetch-oat hays.

Hay	*a*	*b*	*u*	k_d (%·h^{-1})	ED (%)
		(%)			
H1	29.8	50.7	19.5	5.05	62.0
H2	22.3	59.0	18.7	4.92	59.6
H4	35.4	50.5	14.1	5.83	68.8
H5	37.3	45.3	17.4	6.64	68.8
H6	31.1	46.3	22.6	5.41	61.3
H7	34.0	47.1	18.9	7.96	68.6
H8	29.3	43.5	27.2	7.71	60.9
H9	41.0	41.4	17.6	9.03	72.3
H10	28.2	45.0	26.8	8.09	61.4
H11	30.2	48.6	21.2	9.20	67.2
H12	39.0	43.1	17.9	11.8	73.4
Mean	32.5	47.3	20.2	7.42	65.8
CV (%)	16.7	10.3	20.1	28.4	7.9

a, *b* and *u* represent soluble, non-soluble degradable, and undegradable fractions, respectively; k_d, fractional degradation rate of fraction *b*; values for each hay are means of three sheep.

Table II. Degradation kinetics and effective degradability (ED) of crude protein of common vetch-oat hays.

Hay	*a*	*b*	*u*	k_d (%·h^{-1})	ED (%)
		(%)			
H1	40.3	42.8	16.9	23.1	78.3
H2	36.4	52.1	11.5	6.42	72.3
H4	36.4	49.6	14.0	11.9	76.1
H5	41.0	49.0	10.0	16.6	82.5
H6	47.3	38.6	14.1	11.2	78.0
H7	38.2	51.6	10.2	12.3	79.9
H8	39.9	43.9	16.2	17.2	77.4
H9	41.0	50.7	8.3	15.0	83.2
H10	37.6	47.7	14.7	13.7	77.0
H11	41.8	48.9	9.3	17.7	83.9
H12	41.8	49.5	8.7	17.7	84.1
Mean	40.2	47.6	12.2	14.8	79.3
CV (%)	7.7	8.8	25.6	29.7	4.7

a, *b* and *u* represent soluble, non-soluble degradable, and undegradable fractions, respectively; k_d, fractional degradation rate of fraction *b*; values for each hay are means of three sheep.

Table III. Correlation coefficients between chemical and botanical composition and in situ degradation characteristics of dry matter of common vetch-oat hays.

Fraction[b]	In situ degradation characteristics[a]				
	a	*b*	*u*	k_d	ED
CP	0.514**	–0.352*	–0.246	0.655***	0.614***
CF	–0.661***	0.325	0.473**	–0.418*	–0.695***
NDF	–0.866***	0.652***	0.341	–0.744***	–0.833***
HCEL	–0.715***	0.609***	0.195	–0.651***	–0.643***
CEL	–0.796***	0.647***	0.256	–0.740***	–0.791***
ADL	0.028	–0.523**	0.607***	0.393*	–0.102
NDIN	–0.649***	0.641***	0.068	–0.626***	–0.598***
ADIN	–0.778***	0.301	0.657***	–0.251	–0.743***
Vetch proportion[c]	0.362*	–0.629***	0.296	0.821***	0.412*

[a] ED: effective degradability, other ruminal degradation characteristics as in Table I.
[b] CP: crude protein; CF: crude fibre; NDF: neutral detergent fibre; ADF: acid detergent fibre; ADL: acid detergent lignin; HCEL: hemicellulose (determined as NDF-ADF); CEL: cellulose (determined as ADF-ADL); NDIN: insoluble nitrogen in NDF reactive solution; ADIN: insoluble nitrogen in ADF reactive solution.
[c] On DM basis.
*, **, *** $P < 0.05$, 0.01 and 0.001, respectively.

content (r = –0.873; $P < 0.001$) and positively correlated, even if at a minor level, with fibre fractions and ADIN. The variation of ED of CP was negatively associated to variations of fibre and structural carbohydrate fractions and fibre bound N proportions, but specially with cellulose (r = –0.916; $P < 0.001$) and NDIN (r = –0.831; $P < 0.001$).

Table IV. Correlation coefficients between chemical and botanical composition and in situ degradation characteristics of crude protein of common vetch-oat hays.

Fraction[b]	In situ degradation characteristics[a]				
	a	*b*	*u*	k_d	ED
CP	0.342	0.402*	–0.873***	–0.069	0.655***
CF	–0.416*	–0.173	0.635***	–0.212	–0.735***
NDF	–0.209	–0.343	0.665***	–0.255	–0.768***
HCEL	–0.216	–0.247	0.541**	–0.035	–0.527**
CEL	–0.304	–0.241	0.618***	–0.493**	–0.916***
ADL	0.604***	–0.426*	–0.009	0.081	0.252
NDIN	–0.259	–0.064	0.337	–0.698***	–0.831***
ADIN	–0.201	–0.248	0.530**	–0.329	–0.681***
Vetch proportion[c]	0.232	0.210	–0.508**	0.0079	0.583***

[a] ED: effective degradability, other ruminal degradation characteristics as in Table II.
[b] For abbreviations see Table III.
[c] On DM basis.
*, **, *** $P < 0.05$, 0.01 and 0.001, respectively.

Conversely, ED of CP was positively correlated, although at a lower level, with the CP content.

Using the step-wise procedure and chemical fractions as independent variables, we obtained regression equations to estimate ED:

Dry matter

1) ED = 94.6 – 0.06 NDF
RSD = 0.921; $R^2 = 0.694$; $P < 0.001$

2) ED = 104.8 – 0.06 NDF – 0.16 ADL
RSD = 0.772; $R^2 = 0.794$; $P < 0.001$

Crude protein

1) ED = 102.3 – 0.10 CEL
RSD = 0.498; $R^2 = 0.839$; $P < 0.001$

For DM degradability, NDF and ADL contents explained 79.4% of the total variation, while for CP degradability, the cellulose content explained 83.9%.

4. DISCUSSION

Disappearance data of DM and CP fitted well with the model used, and did not show evidence of lag time for any hay. Mean ED values of DM (65.8%) and CP (79.3%) agreed with those observed by Rodríguez [12] for a common vetch-oat hay (64.7% and 78.9%, respectively). For DM, mean ED was also intermediate to the values of common vetch (68.7%) and oat (63.8%) hays obtained by Rubio [14]. On the contrary, mean ED of CP was higher than the values obtained by this author for both vetch (78.4%) and oat (68.2%) hays.

With the exception of the degradation rate (k_d), most ruminal DM degradation characteristics were better correlated with chemical components than with the vetch proportion in hays. Comparative studies on ruminal degradation [14] reported a higher degradation rate of DM for a common vetch hay ($11.8\%\cdot h^{-1}$) than for an oat hay ($4.9\%\cdot h^{-1}$). Grenet and Demarquilly [8] also reported a higher k_d for legumes than for grass species.

Among the chemical constituents in our study sample, NDF showed the highest influence on ruminal degradation characteristics and ED of DM. Cell-wall concentration was negatively correlated with the *a* fraction and the fractional degradation

rate (k_d), and positively correlated with the *b* and *u* fractions. These results indirectly showed that the soluble fraction (*a*) was linked to the cell content (complementary to the NDF fraction) while the *b* and *u* fractions were mainly linked to fibre components and lignin, respectively. Consequently, the variability of the *a* and *b* fractions was linked to the variability of the fibre fractions while a higher variability of the undegradable *u* fraction was related to the higher variability of lignin and ADIN.

The increasing vetch proportion in hay was associated with an increase of cell contents and a decrease of cell walls [9] and, therefore, the vetch proportion had a positive effect on the *a* fraction and k_d value and a negative effect on the *b* fraction. As a result, the net effect of the vetch proportion on the estimated ED of DM was positive, but this correlation only reached a moderate level ($r = 0.4118$, $P = 0.0173$), as a consequence of the contrasting correlations with vetch proportions of the independent chemical fractions obtained for the prediction of ED of DM. The increase of both NDF or ADL contents reduces ED, but a higher vetch proportion increases ADL and tends to reduce NDF [9].

On average, some 80% of total CP in the vetch-oat hays was apparently degraded in the rumen. Nevertheless, when this value was corrected for microbial contamination, using the equation proposed by Rodriguez et al. [13] which was obtained with the same methodology and with similar diet and ingestion conditions, the results showed that 85% of the protein of these roughages is diverted to microbial ruminal fermentation. The ED underestimation, produced by the microbial contamination, represented from 4.7 to 8.6% (mean value 6.6%) and was negatively correlated to the vetch proportion in hay ($r = –0.68$; $P < 0.05$). This relation was in agreement with the higher error due to microbial contamination observed in grasses than in legumes [1, 10, 12]. In the above reported equation [13], this error was positively related to cellulose content, and negatively to the concentration of CP and its apparent ED. The cellulose content was higher and the CP and ED of CP were lower in vetch hay than in oat hay [9] and, therefore, microbial contamination was lower in vetch hay. In this manner, the variation of corrected values of CP degradability was lower than that of apparent values (CV: 3.4 vs. 4.7%, respectively).

In the study of Rubio [14], the soluble fraction (*a*) and the potentially degradable fraction (*b*) of CP were 1% and 9% lower in oat hay than in common vetch hay. The undegradable CP fraction (*u*), however, was 46% higher in oat than in common vetch. These findings may explain why, in our study sample, the only significant correlation between vetch proportion and the kinetic parameters for degradation was found for the undegradable fraction ($r = –0.508$, $P < 0.01$). The higher variability observed for the *u* fraction (Tab. II) than for the *a* or *b* fractions was also explained considering that vetch and oat contribution only had a differential effect for the undegradable fraction.

The two components (microbial and dietary) of this fraction should be considered jointly to explain their correlations with chemical composition. Rodriguez [12], with a total of 14 samples of different feeds, observed that the microbial component of the undegradable CP fraction is positively related to the content of structural carbohydrates (mainly cellulose) and negatively related to the N concentration of feed. On the contrary, the positive correlation observed between this CP fraction and ADIN could be related to the N of lignin and Maillard complexes included in the feed component of the undegradable CP fraction. Both this latter fact and the high CP degradability values indicate that rumen undegraded CP, that could also be digested in the small intestine, should be very limited. Therefore, most N available to animals from these hays is derived from ruminal microbial protein.

The ED of CP in our study sample increased as vetch proportion increased ($r = 0.583$, $P < 0.001$) as a result of the higher CP degradability of vetch than oat [14]. An increasing vetch proportion in hay would mean a higher CP content [9] and a lower proportion of the undegradable CP fraction and, therefore, a higher nitrogen value of hay for ruminants.

When ruminal degradation is extensive, as for CP in these hays, ED tends to be linked to the undegraded fraction. In this sense, the cellulose content was the chemical fraction more closely related to the *u* fraction and to the rumen escape CP portion of the *b* fraction, through its negative relation with k_d (Tab. IV). The cellulose content of the hays was also negatively correlated with the vetch proportion [9], supporting the observed effect of vetch contribution on ED of CP.

5. CONCLUSION

Common vetch-oat hays are characterised by an extensive CP degradation, which is also responsible for a low content of digestible bypass protein in the small intestine. Most N available to animals from these hays is derived from microbial protein synthesised in the rumen. The increase of vetch proportion in hays has a positive effect on its nutritive value.

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