

Article

# Feed Value of Barn-Dried Hays from Permanent Grassland: A Comparison with Fresh Forage

Donato Andueza <sup>1,\*</sup>, Fabienne Picard <sup>1</sup>, Philippe Pradel <sup>2</sup> and Katerina Theodoridou <sup>3</sup>

<sup>1</sup> Université Clermont Auvergne, INRA, VetAgro Sup, UMR Herbivores, F-63122 Saint-Genès-Champanelle, France; fabienne.picard@inra.fr

<sup>2</sup> INRA-Herbipôle, Unité 1414, F-63122 Saint-Genès Champanelle, France; philippe.pradel@inra.fr

<sup>3</sup> Institute for Global Food Security, Queen's University Belfast, 19 Chlorine Gardens, Belfast BT9 5DL, UK; k.theodoridou@qub.ac.uk

\* Correspondence: donato.andueza@inra.fr; Tel.: +33-4736-24071

Received: 29 March 2019; Accepted: 28 May 2019; Published: 30 May 2019



**Abstract:** In mountain areas, hays are the main forage in winter diets for livestock. Barn-dried hays can be an alternative to traditional hays, which are generally characterized by a low feed value. The aim of this study was to compare the feed value of barn-dried hays with that of the fresh forage from a permanent meadow. The study was carried out over three periods during the first growth cycle of the meadow's vegetation (from 30 May to 3 June, from 13 to 17 June, and from 27 June to 1 July). Fresh forage and barn-dried hays of the same fresh forages were tested for dry matter digestibility (DMD), organic matter digestibility (OMD), and voluntary intake (VI). Both types of forage obtained each period were tested with an interval of 15 days. Chemical composition and OMD of forages did not change ( $p > 0.05$ ) according to the feeding method. However, the DMD values for barn-dried hays were higher ( $p < 0.05$ ) than for fresh forages at the end of the cycle. VI and digestible organic matter intake of barn-dried hays were higher ( $p < 0.05$ ) than that of fresh forages. In conclusion, barn-dried hays obtained from permanent grasslands presented a higher feed value than fresh forages.

**Keywords:** permanent grassland; fresh forage; barn-dried hay; chemical composition; digestibility; voluntary intake

## 1. Introduction

In many regions of Europe and particularly in mountain areas, semi-natural grasslands are the predominant land types, and milk and meat production are based on forages they supply. The grassland systems of these areas seek to maximize the use of grass for grazing purposes, but preserved forages are also needed for feeding during the winter or to offset shortages during summer droughts. However, poor knowledge of their feed value and the reduced performance obtained when they were compared to seeded forages, such as that provided by *Lolium perenne* L. grasslands [1], make farmers reluctant to use forages obtained from semi-natural grasslands. The results obtained by Bruinenberg et al. (2002) [1], partially disagree to those obtained by Andueza et al. (2010) [2] and Andueza et al. (2013) [3] which reported a broad variability in digestibility and voluntary intake (VI) of different permanent grasslands at different maturity stages.

Haymaking is the most popular forage preservation method in mountain areas. However, the nutritive quality of hays is not optimal as it is often conditioned by weather, particularly in spring. According to Rotz and Muck (1994) [4] average dry matter (DM) losses in hay making are estimated between 24% and 28% of the original forage. To overcome this problem, farmers increasingly use barn-dried hays. However, their relative nutritive value is often not well known. French National Institute for Agricultural Research (INRA) nutritive value estimates for permanent grasslands are not

exhaustive, while nutritive value of barn-dried hays is unknown, and the few relevant references in the literature mostly concern seeded forages. In general, in this preservation system, changes in the quality of forages are mainly related to plant respiration of grass after cutting. However changes in the feed value of artificially dried forages are often conflicting in the literature: Pasha et al. (2004) [5], reported higher digestibility and intake values for forced-air-dried hays than for frozen grass, but Archimède et al. (1999) [6] found higher organic matter digestibility (OMD) and VI for fresh *Digitaria decumbens* Stent than the equivalent forage dried for 20 h at 60 °C. Similar results were found in sheep by Dulphy and Rouel (1987) [7] for VI. Finally, Demarquilly (1970) [8] and Delaby and Peccatte (2008) [9] reported similar results between fresh forage and barn dried hay or those dehydrated at low temperatures, but Andueza et al. (2009) [10] found higher OMD values for lucerne hays than the same dehydrated forages, although no differences between preservation methods were found for VI. From these results, we hypothesized that the feed value of barn-dried permanent grassland hays might be similar to that of fresh forage.

Our aim in this work was to assess the effects of the barn-dried preservation method as a management alternative option to fresh forage feeding, for a permanent grassland across its first growth cycle on the chemical composition and feed value of the forage.

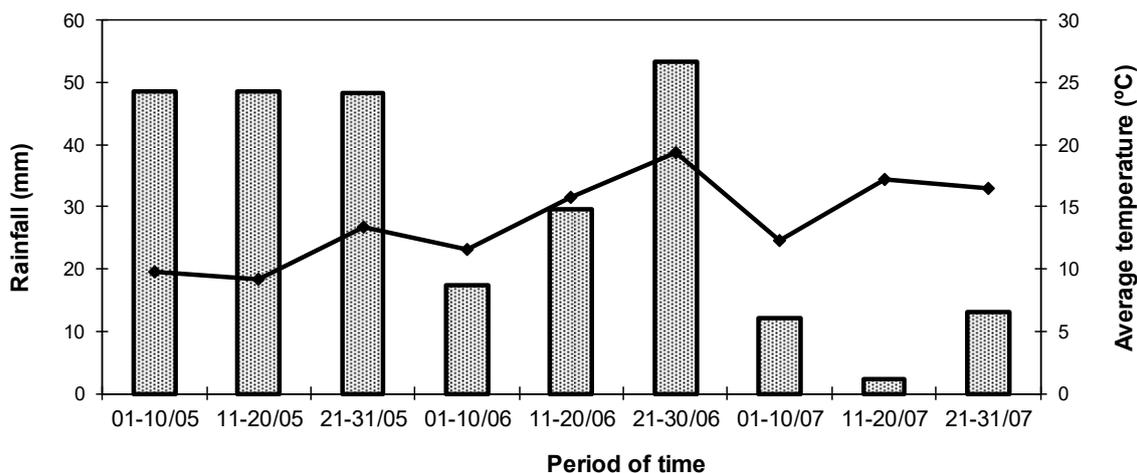
## 2. Materials and Methods

The study was conducted indoors at the French National Institute for Agricultural Research of the Marcenat experimental farm (INRA-Herbipôle) in France. The animals were handled by specialized personnel who applied animal care and welfare in accordance with European Union Directive no. 609/1986, under agreement no. A63 565. The experimental procedures complied with French regulations for the use of experimental animals (statutory order no. 87-848, guideline of 19 April 1988).

### 2.1. Forages and Climatic Conditions

One permanent grassland, located at Marcenat (Cantal, France 2°49' E, 45°18' N) at 1060 m above sea level, was used. It received 33 kg of N/ha in March 2005. The experimental period started on 20 May and lasted 8 weeks including an 8-day period of adaptation to the experiment (between 20 to 29 May). The sward was used three times during the first growth cycle of 2005 for fresh forage or barn-dried forage (from 30 May to 3 June, from 13 to 17 June and from 27 June to 1 July). Every Wednesday, (1, 15, and 29 June) an area of the 0.5 ha of this plot was cut, and forage was harvested for drying in an experimental barn. The rest of the Wednesdays between 18 May and 29 June, forages were barn-dried but they were not used in the current study. Briefly, air was heated to a maximum of 40 °C and then blown into the bottom of a box (2 × 2 × 2 m<sup>3</sup>) containing the forage harvested using a fan and an electric heating element. The 3 kW centrifuge fan produced an air flow of 4000 m<sup>3</sup>/h. The ambient air at the inlet of the ventilator was heated by 6 °C using a 9-kW electric heating element. Two boxes were used. The drying procedure lasted between 4 and 7 days according to the dry matter content of the fresh forage. Forage was manually turned once a day during each drying period to ensure consistent dehydration. Finally, 300 kg of dried forage was obtained each week.

Total rainfall during the growth cycle (1 April–30 July) was 300 mm. Total rainfall during the period of the study was 31 mm (Figure 1). Average temperatures ranged between 14 °C and 18 °C. Previous management of the grassland consisted in general on cutting the plot for hays at the end of June and then grazed in August and October. Plots received usually 30 kg of N/ha early in spring.



**Figure 1.** Gausson ombrothermic diagram of the study site (Marcenat, France) from 1 May until 31 July in the period of 10-days including the experimental period when forage tested was grown (from 30 May to 1 July). Bars indicate rainfall. Average temperatures are line joined.

## 2.2. *In Vivo* Digestibility Trials

Twelve 2-year old Texel wethers (mean live weight: 63 kg) were used. Each forage type (fresh or barn dried forage) was randomly assigned to six wethers. The *in vivo* digestibility trials were run in each of the three measurement periods over 15 days, in which the first 10 days were devoted to adaptation to diet and the last 5 days (dates showed previously) to data collection (daily offering, refusals and feces). The offered diet consisted of fresh unpreserved forage cut daily from the grassland, or barn-dried hay, chopped to a length of 5–7 cm and offered *ad libitum* twice a day, at 8:00 a.m. and 4:00 p.m. [11]. The amount of forage offered was adjusted daily on the basis of the previous day's intake. A refusal of 0.1 of the offered quantity was allowed. Throughout the experimental period, the animals had free access to water and vitamin–mineral blocks. Barn-dried hays were tested 15 days after testing the same forage cut daily and offered fresh.

## 2.3. Samples

During the data collection of each period in the digestibility trials, three samples of about 200 g were randomly collected per day and per forage before chopping of herbage (total = 15 samples). These samples were stored at  $-20^{\circ}\text{C}$  and were used for the determination of the botanical composition and the phenological stage. Botanical composition was determined by hand separation and weighing of samples of different plant species on five samples per forage. Senescent material (Sm) was considered globally for all plants. Phenological stage was determined on three samples per forage using 50 random grass tillers according to an adaptation of the method proposed by Moore et al. (1991) [12]. The mean plant stage by weight (MPW) for each sample was calculated by

$$\text{MPW} = \frac{\sum (C_i D_i)}{D} \quad (1)$$

where  $C_i$ , is the code of stage  $i$  as defined in Table 1,  $D_i$  the total dry weight for tillers in stage  $i$ , and  $D$  the total dry weight for all tillers.

Furthermore, during digestibility trials, forage offered refusals and the feces of each animal were sampled daily. At the end of each period, three samples of forage and one sample of refusals and feces were made up and they were used for chemical analyses.

**Table 1.** Definition of the plant stages (in grasses) used to characterize the medium plant stage of the grasslands (adapted from [12]).

Stage	Code	Description
Vegetative		
Vegetative leaf development	1.5	
Stem Elongation		
Elongation (beginning)	2	First node palpable/visible
Elongation	2.5	Nodes palpable/visible
Elongation (end)	3	Boot stage
Reproductive/Floral Development		
Inflorescence emergence	3.1	First spikelet visible
Inflorescence	3.3	Spikelets fully emerged/peduncle not emerged
Inflorescence emerged	3.5	Inflorescence emerged/peduncle fully elongated
Anther emergence/anthesis	3.8	
Seed Development and Ripening		
Caryopsis visible	4	
Milk	4.1	
Dough	4.4	
Endosperm hard/physiological maturity	4.7	
Endosperm dry/seed ripe	4.9	

#### 2.4. Analyses

All the samples (hand-separated plant species, forage offered, refusal, and feces) were oven-dried at 60 °C for 72 h. Forage offered, refusal and feces samples were ground through a 0.8 mm screen. Samples of forage offered and refusals were analyzed for crude ash (CA) according to AOAC, (1990) [13]. Forages offered were analyzed for nitrogen (N) [13], water soluble carbohydrates (WSC) [14] neutral detergent fiber (NDF) according to the method described by Van Soest et al. (1991) [15] and for acid detergent fiber (ADF) and acid detergent lignin (ADL) according to Van Soest and Robertson (1980) [16]. Neutral detergent fiber and ADF analyses were performed using the Ankom apparatus (Ankom® Tech. Co., Fairport, NY, USA). Analyses of CA were determined in feces samples.

#### 2.5. Calculations

Crude protein (CP) was obtained by multiplying the N concentrations by 6.25. The relative proportions of grasses, legumes, and forbs were calculated on a DM basis from botanical composition data. The in vivo results and DM and CA data were used to calculate dry-matter digestibility (DMD), OMD, and digestible organic matter intake (DOMI). Voluntary intake (calculated as the average of a 5-day period of daily forage offered minus refusal) and DOMI were expressed in g DM/kg of metabolic body weight ( $BW^{0.75}$ ).

#### 2.6. Statistical Analysis

Data on chemical composition of the samples for the two types of forages in the experimental periods underwent to repeated-measures ANOVA according to the model

$$Y_{ijk} = \mu + F_i + P_j + (F \times P)_{ij} + R(F \times P)_{ijk} + \varepsilon_{ijk}, \quad (2)$$

where Y is the dependent variable,  $\mu$  is the overall mean, F is type of forage (1 df), P is the period (2 df),  $F \times P$  is the interaction between type of forage and period (2 df), R is the replicate effect (2 df), and  $\varepsilon$  is the experimental error. Replicate was considered as a random effect, and period as a repeated measure.

For ANOVA of in vivo digestibility data (DMD, OMD, and DOMI), we used the model

$$Y_{ijk} = \mu + F_i + P_j + (F \times P)_{ij} + A(F)_{ik} + \varepsilon_{ijk}, \quad (3)$$

where Y is the result for a single animal,  $\mu$  is the overall mean, F is type of forage (1 df), P is the effect of period (2 df), A is the effect of animal (5 df), F  $\times$  P is the interaction between type of forage and period (2 df), and  $\epsilon$  is the experimental error. Each animal was considered as a block factor and as a random variable. Single degree-of-freedom orthogonal polynomial contrasts were used to detect linear and quadratic effects of time on chemical composition, digestibility coefficients, daily feed intake, and DOMI. Because forage samples were taken at 665, 822, and 1059 growing degree days (GDD), polynomial contrasts were adjusted for unequally spaced time effects (i.e., 665, 822, and 1059 GDD). The relationships between temperature accumulation from 1 February using 0 °C as minimum base temperature and 18 °C as maximum base temperature [17], phenological stage determined by MPW, chemical composition variables, in vivo digestibility, and intake data were evaluated by Pearson correlation coefficients. All analyses were performed using the mixed procedure of the SAS statistical package [18].

### 3. Results

#### 3.1. Botanical Composition and Phenological Stage

The average botanical composition of the permanent meadow during the three periods of the experiment is reported in Table 2. Permanent meadow was characterized by higher proportions of grasses than that of forbs. The ratio between the two groups increased with the advance of the phenological cycle. At young stages, the proportion of grasses was 0.76 and at more mature stages, the proportion of grasses increased to 0.83. Proportion of legumes was always less than 0.01. Initially, grassland species >0.10 were *Agrostis capillaris* L., *Dactylis glomerata* L., *L. perenne*, and *Taraxacum officinale* F.H. Wigg., whereas at more advanced phenological stages dominated species were *A. capillaris*, *L. perenne*, and *Festuca rubra* L. Mean plant weight at P1 (665 GDD) was 2.65 (boot stage, Table 1) and increased until P2 (822 GDD) before finally levelled off between P2 and P3 (1059 GDD) (Table 2).

**Table 2.** Botanical composition (percentage of dry matter) and mean plant weight (MPW) (dimensionless) phenological stage of the permanent grassland in the three periods of the study.

	P1 <sup>1</sup>	P2 <sup>2</sup>	P3 <sup>3</sup>
<b>Grasses</b>			
<i>Agrostis capillaris</i> L.	20.23	12.96	29.87
<i>Trisetum flavescens</i> L.	4.41	4.01	7.02
<i>Bromus mollis</i> L.	1.03	0.55	0.00
<i>Dactylis glomerata</i> L.	15.39	4.92	0.84
<i>Festuca rubra</i> L.	3.14	2.02	9.97
<i>Anthoxanthum odoratum</i> L.	0.19	0.00	1.61
<i>Poa pratensis</i> L.	6.06	7.37	8.25
<i>Holcus lanatus</i> L.	1.44	0.19	6.34
<i>Lolium perenne</i> L.	18.81	56.46	16.80
<i>Phleum pratense</i> L.	5.64	0.27	2.62
<b>Forbs</b>			
<i>Cerastium fontanum</i> Baumg.	9.57	1.54	5.41
<i>Taraxacum officinale</i> P.H. Wigg.	11.64	2.54	3.16
<i>Urtica dioica</i> L.	0.00	1.01	0.02
Sum of grasses+forbs	97.55	93.84	91.91
<b>Others</b>			
Senescent material	0.72	4.70	6.36
MPW	2.65	3.79	3.82

<sup>1</sup> P1 = first period (from 30 May to 3 June; 665 GDD); <sup>2</sup> P2 = second period (from 13 to 17 June; 822 GDD);

<sup>3</sup> P3 = third period (from 27 June to 1 July; 1059 GDD). Only those species that presented a percentage greater than 1 percent are shown.

### 3.2. Chemical Composition

There were no differences between types of forages ( $p > 0.05$ ) for any determinations of chemical composition (Table 3). Concerning the period or the cumulation of temperature, for the CA content, there were no differences ( $p > 0.05$ ) between values found among periods. The CP content decrease in a quadratic pattern along the first cycle of growth with a more pronounced decrease at the beginning (between the first and second period) and a weaker one at the end of the phenological cycle (between the second and third period). Neutral detergent fiber, ADF and ADL concentrations increased linearly over the beginning (first period) and the end of the experiment third period. Finally, for WSC concentration there were no significant ( $p > 0.05$ ) differences between forages obtained across the growth cycle (in the three periods of the study); this concentration was approximately 100 g/kg DM. For all determinations, the interaction between type of forage and period (F  $\times$  P) was non-significant ( $p > 0.05$ ).

**Table 3.** Crude ash (CA) (g/kg dry matter (DM)), crude protein (CP) (g/kg DM), neutral detergent fiber (NDF) (g/kg DM), acid detergent fiber (ADF) (g/kg DM), acid detergent lignin (ADL) (g/kg DM), and water-soluble carbohydrates (g/kg DM) of the permanent grasslands throughout the first growth cycle.

	Forage		Period			SEM <sup>6</sup>	Significance			
	FF <sup>1</sup>	BDF <sup>2</sup>	P1 <sup>3</sup>	P2 <sup>4</sup>	P3 <sup>5</sup>		Period	Contrast	Forage	Period $\times$ Forage
CA	80	79	82	71	84	10.2	ns <sup>10</sup>	ns	ns	ns
CP	11.9	12.1	15.6	10.6	9.9	0.60	***	L <sup>8,*</sup> Q <sup>9,*</sup>	ns	ns
NDF	598	592	545	590	652	16.8	***	L *	ns	ns
ADF	320	322	287	321	355	6.3	***	L *	ns	ns
ADL	45	40	33	42	53	5.9	*	L *	ns	ns
WSC	104	92	99	105	90	11.1	ns	ns	ns	ns

<sup>1</sup> FF = fresh forage; <sup>2</sup> BDF = barn-dried forage; <sup>3</sup> P1 = first period (from 30 May to 3 June; 665 GDD); <sup>4</sup> P2 = second period (from 13 to 17 June; 822 GDD); <sup>5</sup> P3 = third period (from 27 June to 1 July; 1059 GDD). <sup>6</sup> SEM = standard error of the mean; <sup>7</sup> P = Probability; <sup>8</sup> L = linear; <sup>9</sup> Q = quadratic; <sup>10</sup> ns:  $p > 0.05$ ; \*,  $p < 0.05$ ; \*\*\*,  $p < 0.001$ .

### 3.3. Digestibility and Intake

Dry matter digestibility and OMD declined with the increase in accumulated temperature ( $p < 0.001$ ) (Table 4). For the DMD, this decline did not varied with the type of forage ( $p < 0.10$ ) but a significant ( $p < 0.001$ ) interaction between type of forage and GDD at which forages were cut. For DMD of fresh forages, linear effects ( $p < 0.05$ ) were observed, whereas the DMD of preserved forages decreased, showing linear and quadratic effects ( $p < 0.05$ ). The main differences in DMD between barn-dried hays and fresh forages were obtained at 1059 GDD (P3) (0.09 points between the two types of forage ( $p < 0.001$ )). No significant differences between forages ( $p > 0.05$ ) were found for OMD, and this declined linearly ( $p < 0.05$ ) along the development cycle.

Barn-dried hays showed higher mean VI values ( $p < 0.05$ ) and higher mean DOMI values ( $p < 0.01$ ) than fresh forage (65.01 vs. 55.70 g/kg BW<sup>0.75</sup> respectively for VI and 36.11 vs. 28.46 g/kg BW<sup>0.75</sup> respectively for DOMI) (Table 4). Voluntary Intake and DOMI for both types of forage decreased quadratically ( $p < 0.001$ ) along the cycle of growth. For these determinations, the interaction type of forage by period was non-significant ( $p > 0.05$ ).

Temperature accumulation (GDD) was closely correlated with digestibility coefficients values and chemical composition, particularly with cell wall content and cell wall partitioning (Table 5). A close correlation was also observed with VI of barn-dried hays than with VI of fresh forages. Furthermore, phenological stage code was more closely correlated with CP than with cell wall contents. On the other hand, it was also less closely correlated compared to temperature accumulation with digestibility coefficients of both fresh forages and barn-dried hays.

**Table 4.** Average values of the grassland in the three cuts, and significance levels obtained in the analysis of variance developed for dry matter digestibility (DMD) (g/g), organic matter digestibility (OMD) (g/g), voluntary intake (VI) (g/kg BW<sup>0.75</sup>), and digestible organic matter intake (DOMI) (g/kg BW<sup>0.75</sup>).

	Fresh Forage					Barn-Dried Forage					Significance		
	P1 <sup>1</sup>	P2 <sup>2</sup>	P3 <sup>3</sup>	SEM <sup>4</sup>	Contrast	P1	P2	P3	SEM	Contrast	F <sup>5</sup>	P <sup>6</sup>	F × P
DMD	0.64	0.56	0.41	0.564	L <sup>7,*</sup>	0.64	0.55	0.50	0.013	L * Q <sup>8,*</sup>	†	***	***
OMD	0.66	0.60	0.50	0.012	L *	0.66	0.59	0.52	0.012	L *	ns <sup>9</sup>	***	ns
VI	67.07	50.27	49.75	4.408	L * Q *	80.87	60.96	53.20	4.408	L * Q *	*	***	ns
DOMI	35.94	26.95	22.48	2.665	L * Q *	49.54	33.60	25.18	2.665	L * Q *	**	***	ns

<sup>1</sup> P1 = first period (from 30 May to 3 June; 665 GDD); <sup>2</sup> P2 = second period (from 13 to 17 June; 822 GDD); <sup>3</sup> P3 = third period (from 27 June to 1; 1059 GDD); <sup>4</sup> SEM = standard error of the mean; <sup>5</sup> P = period; <sup>6</sup> F = forage; <sup>7</sup> L = linear; <sup>8</sup> Q = quadratic; <sup>9</sup> ns =  $p > 0.05$ ; †,  $p < 0.10$ ; \*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ; \*\*\*,  $p < 0.001$ .

**Table 5.** Correlations between sum of temperatures, phenological stage, chemical composition, botanical composition, digestibility coefficients, voluntary intake and digestible organic matter intake of fresh forage and barn dried forage.

	ST <sup>1</sup>	MPW	Ash	CP	NDF	ADF	ADL	WSC	DMDff	OMDff	Viff	DOMIff	DMDbdf	OMDbdf	Vibdf	DOMIbdf	Gram	Leg	for
MPW <sup>2</sup>	0.66																		
Ash	0.31	-0.51																	
CP <sup>3</sup>	-0.87	-0.94	0.19																
NDF <sup>4</sup>	0.99 *	0.67	0.29	-0.88															
ADF <sup>5</sup>	0.99 †	0.74	0.20	-0.92	0.99 †														
ADL <sup>6</sup>	0.99 *	0.70	0.26	-0.90	0.99 *	0.99 *													
WSC <sup>7</sup>	-0.39	0.44	-0.99 †	-0.11	-0.36	-0.28	-0.33												
DMDff <sup>8</sup>	-0.99 *	-0.60	-0.38	0.83	-0.99 †	-0.98	-0.99 †	0.45											
OMDff <sup>9</sup>	-0.99 *	-0.63	-0.34	0.86	-0.99 *	-0.99 †	-0.99 †	-0.41	0.99 *										
Viff <sup>10</sup>	-0.81	-0.97	0.30	0.99 †	-0.82	-0.87	-0.84	-0.23	0.76	0.79									
DOMIff <sup>11</sup>	-0.95	-0.86	-0.01	0.98	-0.96	-0.98	-0.97	0.08	0.92	0.94	0.95								
DMDbdf <sup>12</sup>	-0.96	-0.84	-0.04	0.97	-0.97	-0.98	-0.98	0.12	0.94	0.95	0.94	0.99 †							
OMDbdf <sup>13</sup>	-0.99 †	-0.74	-0.20	0.92	-0.99 †	-0.99 *	-0.99 *	-0.28	0.98	0.99 †	0.87	0.98	0.99 †						
Vibdf <sup>14</sup>	-0.93	-0.88	0.04	0.99 †	-0.94	-0.97	-0.95	-0.03	0.91	0.92	0.97	0.99 *	0.99 †	0.97					
DOMIbdf <sup>15</sup>	-0.96	-0.84	-0.03	0.98	-0.96	-0.98	-0.97	0.10	0.93	0.95	0.94	0.99 *	0.99 *	0.98	0.99 *				
Gram <sup>16</sup>	0.45	0.97	-0.71	-0.83	0.47	0.55	0.50	0.65	-0.38	-0.42	-0.89	-0.70	-0.68	-0.55	-0.74	-0.69			
leg <sup>17</sup>	-0.05	0.72	-0.96	-0.45	-0.02	0.07	0.01	0.94	0.12	0.08	-0.55	-0.27	-0.23	-0.07	-0.31	-0.24	0.87		
for <sup>18</sup>	-0.62	-0.99 *	0.54	0.93	-0.64	-0.71	-0.67	-0.48	0.56	0.60	0.96	0.84	0.81	0.71	0.86	0.82	-0.98	-0.75	
sm <sup>19</sup>	0.94	0.87	-0.03	-0.98	0.95	0.97	0.96	-0.05	-0.91	-0.93	-0.96	-0.99 *	-0.99 *	-0.97	-0.99 *	-0.99 *	0.73	0.30	-0.85

<sup>1</sup> ST = sum of temperatures; <sup>2</sup> MPW = phenological stage mean plant weight; <sup>3</sup> CP = Crude protein; <sup>4</sup> NDF = neutral detergent fibre; <sup>5</sup> ADF = acid detergent fibre; <sup>6</sup> ADL = acid detergent lignin; <sup>7</sup> WSC = water soluble carbohydrates; <sup>8</sup> DMDff = dry matter digestibility for fresh forages; <sup>9</sup> OMDff = organic matter digestibility for fresh forages; <sup>10</sup> Viff = voluntary intake for fresh forages; <sup>11</sup> DOMIff = digestible organic matter intake for fresh forages; <sup>12</sup> DMDbdf = dry matter digestibility for barn-dried hays; <sup>13</sup> OMDbdf = organic matter digestibility for barn-dried hays; <sup>14</sup> Vibdf = voluntary intake for barn-dried hays; <sup>15</sup> DOMIbdf = digestible organic matter intake for barn-dried hays; <sup>16</sup> gram = grasses; <sup>17</sup> leg = legumes; <sup>18</sup> for = forbs; <sup>19</sup> sm = senescent material. †,  $p < 0.1$ ; \*,  $p < 0.05$ .

#### 4. Discussion

The grassland studied was composed mainly of *L. perenne*, *A. capillaris*, and *T. officinale*, although its composition varied throughout the growth cycle as reported also by Andueza et al. (2016) [19]. At young stages, competitive species, as *L. perenne* and *D. glomerata* characterized according to Gross et al. (2007) by fast growth rate and high specific leaf area, formed the largest group, whereas at late stages, conservative species as *A. capillaris*, *Trisetum flavescens* L., and *F. rubra* characterized by slow growth rate and low specific leaf area Gross et al. 2007 were dominant. Other authors [20] recorded changes in the proportion of *A. capillaris* from 0.19 to 0.69 of the total biomass in different periods of the growing season in permanent grassland. The sward used in our study can be considered representative of many situations where forage from seeded temperate grasslands provides a major share of ruminant diets [1].

The present study covers the period between grazing and the time when most permanent grasslands were cut for hays. Despite the study duration (665–1059 GDD) the MPW ranged only between 2.65 (elongation) and 3.82 (anthesis). The presence of different species characterized by a different phenology and the heterogeneity of phenological stages within each one, a typical characteristic of permanent grasslands, [1] can explain this short evolution of maturity stages observed in the permanent grassland used in the present study.

The absence of any significant differences ( $p > 0.05$ ) in chemical composition between fresh forages and barn-dried hays is difficult to explain. In the literature, DM losses due to respiration are well documented, and have been estimated at 2–5% [21–23]. According to these authors, WSC and CP were the main constituents involved in respiratory losses. However, Rees (1982) [21], in his review, point out that some researchers have produced evidence that crops can sometimes increase their weight of DM after cutting. This may be due to continuing of photosynthesis and the weight of nutrients assimilated being greater than that used for respiration [21]. In the present study, the balance between the nutrient assimilated in the photosynthesis after cutting and that used for respiration might explain the lack of differences between fresh forages and the barn-dried forages despite the different conditions of plant species proportions, maturity stage, temperature, and moisture of forages processed.

An interesting result of the present study was the closer relationship between CP and MPW than between MPW and structural carbohydrates or lignin. The evolution of cell wall content and the partitioning components remain more closely correlated to temperature accumulation in agreement with other reported results [24]. In general, factors such as age and maturity stage are confounded, but Buxton (1996) [24] states that under most circumstances, maturity stage rather than age can be more closely related to quality. The results found in the present study for CP agree with this statement, but not those obtained for structural carbohydrates, which are more closely related to the digestibility values. The coexistence in the permanent grassland of different species and different phenological stages within each one at a given date could explain these results.

Differences between forages were not significant ( $p > 0.05$ ) for OMD, in agreement with chemical composition results. However, a surprising result of the present study is the differences between DMD and OMD values for the two types of forage and specifically differences for the last period. These results suggest that the digestibility of minerals could be greatly diminished in P3 for fresh forages. There is no obvious explanation for this lower DMD for fresh forages at P3. The possible presence of antinutritive compounds (e.g., condensed tannins) [25] in fresh forages which could be inactivated by the drying process might explain these results.

The presence of condensed tannins is influenced by the environmental conditions under which the plant is grown. It is likely that longer photoperiod and higher temperatures lead to an increased biosynthesis of tannins, as observed by Lees et al. (1994) [26] on lotus. In our study, the average temperatures were higher in the last period (18 °C) than in the first two (14 °C). In addition, it has been found that CT concentration tends to increase with phenological stage [27], so we would expect the concentration to be higher at P3 than in the other periods in our study. In line with this finding, Frutos et al. (2004) [28] state that condensed tannins are chelating agents that can reduce the availability

of certain metal ions. By contrast, Scharenberg et al. (2007) [29] report a lower digestibility of Ca, P, Mg, and Na of dried and ensiled sainfoin without polyethylene glycol than of dried and ensiled sainfoin with added polyethylene glycol. This suggests that complexes of condensed tannins formed with the minerals were the most likely reason for the low mineral digestibility of sainfoin without condensed tannins.

Another interesting result of this study is the higher VI of barn-dried hays than of fresh forage. Fresh forages are usually consumed in spring, summer, and autumn, and hays in winter when fresh forages do not grow. As seasons influence the VI of animals [30], comparison of fresh forages and hays is difficult to carry out. In some studies, fresh forage is stored frozen until the comparison trial is performed [5]. In the present study, digestibility and VI trials for fresh forages were run only 15 days before trials for the same forages preserved as barn-dried hays, thus minimizing the influence of season on the VI of animals. In the literature, the reported effects of artificial drying on forage intake are conflicting. Whereas Archimède et al. (1999) [6] reported higher intake of *Digitaria decumbens* when it was ingested as fresh forage than as barn-dried hay, Demarquilly (1970) [8] found no differences between the VI of fresh forages and those dehydrated at low temperature. However, Estrada et al. (2004) [31] reported higher VI for lactating cows when ryegrass was offered partially dried than when it was offered after cutting. The results of Archimède et al. (1999) [6] and those of Demarquilly (1970) [8] could be explained by possible losses associated with the barn drying process, mainly respiratory losses, [23]. By contrast, Estrada et al. (2004) [31] explain their results by the influence of water content of fresh grassland forage, which could limit their intake. However, in the present study, this effect still does not fully explain the differences between VI of barn-dried hays and fresh forages. In the first period (proportion of DM in the fresh forage of 0.18) the difference between VI of barn-dried hays and fresh forages was 13.8 g DM/kg BW<sup>0.75</sup>, whereas in P3 the difference between VI of barn-dried hays and fresh forages, (proportion of DM 0.29), was 3.45 g DM/kg BW<sup>0.75</sup>. Other factors that might explain these results are the possible presence of antinutritive compounds in fresh forage, which could depress the VI of animals, and which could be inactivated during the barn drying, or also possible differences in factors associated with ruminal fermentation, which could influence forage intake [5].

## 5. Conclusions

Barn-drying hay is a very good method of preserving forages from permanent grasslands. The results of the present study fail to support the hypothesis that the feed value of barn-dried hays is similar to that of fresh forages. We conclude that feed value of barn-dried hays of permanent grasslands is higher than that of fresh forage. This higher feed value of barn-dried hays of permanent grasslands is mainly a consequence of their higher VI in relation to that of fresh grass. More research is now needed to find a cogent explanation for this higher VI of hays in relation to fresh forages, and particularly to determine the influence of botanical composition of permanent grassland in this effect.

**Author Contributions:** D.A. carried out the experimental design, data interpretation, manuscript writing, and editing. He was also involved in the data recovery. F.P. was involved in the experimental design, data recovery, data analysis and manuscript revision. P.P.: contributed in the experimental design and data recovery. K.T. contributed in the interpretation of results, manuscript writing, and manuscript revision. All authors read and approved the final manuscript.

**Funding:** This work was partially performed thanks to the program “Investissement d’Avenir” (16-IDEX-0001 CAP 20-25) funded by the Agence Nationale de la Recherche of the French government.

**Acknowledgments:** The authors thank the Herbipôle team of the Marcenat experimental farm for their technical help.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

1. Bruinenberg, M.H.; Valk, H.; Korevaar, H.; Struik, P.C. Factors affecting digestibility of temperate forages from seminatural grasslands: A review. *Grass Forage Sci.* **2002**, *57*, 292–301. [[CrossRef](#)]
2. Andueza, D.; Cruz, P.; Farruggia, A.; Baumont, R.; Picard, F.; Michalet-Doreau, B. Nutritive value of two meadows and relationships with some vegetation traits. *Grass Forage Sci.* **2010**, *65*, 325–334. [[CrossRef](#)]
3. Andueza, D.; Picard, F.; Jestin, M.; Aufrere, J. The effect of feeding animals ad libitum vs. at maintenance level on the in vivo digestibility of mown herbage from two permanent grasslands of different botanical composition. *Grass Forage Sci.* **2013**, *68*, 418–426. [[CrossRef](#)]
4. Rotz, C.A.; Muck, R.E. Changes in forage quality during harvest and storage. In *Forage Quality, Evaluation, and Utilization*; Fahey, G.C., Collins, M., Mertens, D.R., Moser, L.E., Eds.; American Society of Agronomy, Inc.; Crop Science Society of America, Inc.; Soil Science Society of America, Inc.: Madison, WI, USA, 1994; pp. 828–868.
5. Pasha, T.N.; Prigge, E.C.; Russell, R.W.; Bryan, W.B. Influence of moisture-content of forage diets on intake and digestion by sheep. *J. Anim. Sci.* **1994**, *72*, 2455–2463. [[CrossRef](#)]
6. Archimede, H.; Poncet, C.; Boval, M.; Nipeau, F.; Philibert, L.; Xande, A.; Aumont, G. Comparison of fresh and dried *Digitaria decumbens* grass intake and digestion by Black-belly rams. *J. Agric. Sci.* **1999**, *133*, 235–240. [[CrossRef](#)]
7. Dulphy, J.P.; Rouel, J. Effect of wilting on changes in the voluntary feed-intake in cattle compared to sheep. *Ann. Zootech.* **1988**, *37*, 31–41. [[CrossRef](#)]
8. Demarquilly, C. Effect of low temperature-dehydration on forage feed-value. *Ann. Zootech.* **1970**, *19*, 45–51. [[CrossRef](#)]
9. Delaby, L.; Peccatte, J.R. Feeding value of ventilated hay from multi-specific pastures. *Fourrages* **2008**, *195*, 354–356.
10. Andueza, D.; Delgado, I.; Muñoz, F. Effect of lucerne preservation method on the feed value of forage. *J. Sci. Food Agric.* **2009**, *89*, 1991–1996. [[CrossRef](#)]
11. Demarquilly, C.; Chenost, M.; Giger, S. Pertes fécales et digestibilité des aliments et des rations. In *Nutrition des Ruminants Domestiques. Ingestion et Digestion*; Jarrige, R., Ruckebusch, Y., Demarquilly, C., Farce, M.H., Journet, M., Eds.; INRA Éditions: Paris, France, 1995; pp. 601–647.
12. Moore, K.J.; Moser, L.E.; Vogel, K.P.; Waller, S.S.; Johnson, B.E.; Pedersen, J.F. Describing and quantifying growth-stages of perennial forage grasses. *Agron. J.* **1991**, *83*, 1073–1077. [[CrossRef](#)]
13. AOAC. *Official Methods of Analysis*, 15th ed.; Association of Official Analytical Chemists: Arlington, VA, USA, 1990; p. 1298.
14. Somogyi, M. Notes on sugar determination. *J. Biol. Chem.* **1952**, *195*, 19–23.
15. Van Soest, P.J.; Robertson, J.B.; Lewis, B.A. Methods for dietary fiber neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* **1991**, *74*, 3583–3597. [[CrossRef](#)]
16. Van Soest, P.J.; Robertson, J.B. *Systems of Analysis for Evaluating Fibrous Feeds*; Pidgen, W.J., Balch, C.C., Graham, M., Eds.; IDRC No 134; International Development Research Centre: Ottawa, ON, USA, 1980; pp. 49–60.
17. Otto, S.; Masin, R.; Chiste, G.; Zanin, G. Modelling the correlation between plant phenology and weed emergence for improving weed control. *Weed Res.* **2007**, *47*, 488–498. [[CrossRef](#)]
18. SAS. *SAS/STAT UsersGuide, Version 6.12*; Statistical Analysis System Institute: Cary, NC, USA, 1998.
19. Andueza, D.; Rodrigues, A.M.; Picard, F.; Rossignol, N.; Baumont, R.; Cecato, U.; Farruggia, A. Relationships between botanical composition, yield and forage quality of permanent grasslands over the first growth cycle. *Grass Forage Sci.* **2016**, *71*, 366–378. [[CrossRef](#)]
20. Todorova, P.A.; Kirilov, A.P. Changes in the permanent grassland composition and feeding value during the growing season. *Grassl. Sci. Eur.* **2002**, *7*, 170–171.
21. Rees, D.V.H. A discussion of sources of dry-matter loss during the process of haymaking. *J. Agric. Eng. Res.* **1982**, *27*, 469–479. [[CrossRef](#)]
22. Dulphy, J.P. Fenaison: Pertes en cours de récolte et de conservation. In *Les Fourrages Secs: Récolte, Traitement, Utilisation*; Demarquilly, C., Ed.; INRA: Paris, France, 1987; pp. 103–124.
23. McGechan, M.B. A review of losses arising during conservation of grass forage. 1. Field losses. *J. Agric. Eng. Res.* **1989**, *44*, 1–21. [[CrossRef](#)]

24. Buxton, D.R. Quality-related characteristics of forages as influenced by plant environment and agronomic factors. *Anim. Feed Sci. Technol.* **1996**, *59*, 37–49. [[CrossRef](#)]
25. Montossi, F.; Liu, F.; Hodgson, J.; Morris, S.; Barry, T.; Risso, D. Influence of low-level condensed tannins concentrations in temperate forages on sheep performance. In Proceedings of the XVIIIth International Grassland Congress, Saskatoon, SK, Canada, 8–19 June 1997; pp. 8.1–8.2.
26. Lees, G.L.; Hinks, C.F.; Suttill, N.H. Effect of high-temperature on condensed tannin accumulation in leaf tissues of big trefoil (*Lotus Uliginosus* Schkuhr). *J. Sci. Food Agric.* **1994**, *65*, 415–421. [[CrossRef](#)]
27. Theodoridou, K.; Aufrere, J.; Andueza, D.; Le Morvan, A.; Picard, F.; Stringano, E.; Pourrat, J.; Mueller-Harvey, I.; Baumont, R. Effect of plant development during first and second growth cycle on chemical composition, condensed tannins and nutritive value of three sainfoin (*Onobrychis viciifolia*) varieties and lucerne. *Grass Forage Sci.* **2011**, *66*, 402–414. [[CrossRef](#)]
28. Frutos, P.; Hervas, G.; Giraldez, F.J.; Mantecon, A.R. Review. Tannins and ruminant nutrition. *Span. J. Agric. Res.* **2004**, *2*, 191–202. [[CrossRef](#)]
29. Scharenberg, A.; Arrigo, Y.; Gutzwiller, A.; Wyss, U.; Hess, H.D.; Kreuzer, M.; Dohme, F. Effect of feeding dehydrated and ensiled tanniferous sainfoin (*Onobrychis viciifolia*) on nitrogen and mineral digestion and metabolism of lambs. *Arch. Anim. Nutr.* **2007**, *61*, 390–405. [[CrossRef](#)]
30. Michalet Doreau, B.; Gatel, F. Annual variations in voluntary food-intake of wethers. *Ann. Zootech.* **1988**, *37*, 151–158. [[CrossRef](#)]
31. Estrada, J.I.C.; Delagarde, R.; Faverdin, P.; Peyraud, J.L. Dry matter intake and eating rate of grass by dairy cows is restricted by internal, but not external water. *Anim. Feed Sci. Technol.* **2004**, *114*, 59–74. [[CrossRef](#)]



© 2019 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<http://creativecommons.org/licenses/by/4.0/>).