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Rumen degradation and transit kinetics of particle size fractions from three different roughages

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Abstract. Although both degradation and transit kinetics parameters are needed to estimate the effective degradability, no attempts appear to have been made in order to determine them on the same particle size populations. This is important as degradation kinetics of the whole diet should be a composite of individual rates of the different particles sizes making up that diet multiplied by their relative contribution. However, some interactions between microbial populations attaching to different particles may occur making this not being so. A similar concern arises when studying transit kinetics. It has been clearly demonstrated that the kinetic behaviour of the whole digesta cannot be determined by its labelling with external markers, which should only be applied to particles in a relatively narrowly defined range of sizes when estimates of particle mean retention time are to be made. The objectives of the present study were: (1) to determine the effect of forage quality (untreated barley straw, ammonia-treated barley straw and lucerne hay) and particle size on their *in situ* degradation kinetics and rates of outflow, and (2) to determine if the behaviour of the whole forage may be estimated from their particle sizes. In a changeover design, chopped untreated straw, ammonia-treated straw and lucerne hay were offered *ad libitum* to three sheep fitted with rumen and duodenal cannulae. Milled (3-mm screen) subsamples of these forages and five particle size families (> 1.2, 1.2 to 0.6, 0.6 to 0.3, 0.3 to 0.15 and 0.15 to 0.045 mm) obtained by wet sieving of the ground material were incubated in the rumen for up to 96 h to study degradation kinetics. Intraruminal doses of Yb-labelled whole ground (3-mm screen) forages and particle size fractions were also given to estimate outflow rates. Extent and fractional rate of degradation of both dry matter and neutral detergent fibre varied between whole roughages and between particle size populations within each forage, with no specific pattern of variation between these latter. Rumen fractional outflow rate was also affected by roughage and particle size, the smaller the size the faster the rate. Although there were large differences between degradation kinetics, fractional outflow rate and effective degradability of whole ground samples and the weighed means estimated from the different particle size populations, these did not reach statistical significance. It was concluded that the nylon bag technique does not completely mimic the behaviour of the composite of the different particle sizes constituting the material incubated, which may lead to severe errors when estimating degradation and transit.

Keywords. Degradation – Forage – Particle size – Transit.

Dégradation et cinétique de transit dans le rumen de différentes tailles de particule de trois fourrages

Résumé. *Bien que la dégradation et la cinétique de transit sont deux paramètres nécessaires pour estimer la dégradabilité effective, aucun essai ne semble avoir été fait afin de les déterminer sur les populations de même taille de particules. Ceci est important car la cinétique de dégradation de l'ensemble du la ration doit être un composite des taux individuels de différentes tailles de particules qui forment ce régime multipliés par leur contribution relative. Cependant, certaines interactions entre les populations microbiennes attachées aux différentes particules peuvent se produire compromettant le résultat. Une préoccupation similaire se pose lors d'étude de la cinétique de transit. Il a été clairement démontré que le comportement de la cinétique de l'ensemble du digesta ne peut être déterminé par son marquage avec des marqueurs externes, qui devrait seulement être appliquée aux particules dans une gamme relativement étroite de tailles définie quand estimations de temps de rétention sont à réaliser. Les objectifs de la présente étude ont été : (1) déterminer l'effet de la qualité du fourrage (paille d'orge non traitée, paille d'orge traitée à l'ammoniac et foin de luzerne) et la taille de particules sur leur cinétique de dégradation* in situ *et le taux de passage, et (2) déterminer si le comportement de l'ensemble du fourrage peut être estimée à partir de leurs tailles de particules. Dans une analyse* change-over, *la paille non traitée haché, la*

paille traitée à l'ammoniac et de foin de luzerne ont été offert in vivo *à trois brebis fistulés au niveau de rumen et de duodénale. Sous-échantillons broyés (tamis de 3 mm) de ces fourrages et cinq familles de la taille des particules (> 1,2; 1,2 à 0,6; 0,6 à 0,3; 0,3 à 0,15 et 0,15 à 0,045 mm) obtenues par tamisage humide de la matière broyée ont été incubés dans le rumen pendant 96 h pour étudier la cinétique de dégradation. Doses intraruminales des fourrages broyés (tamis de 3 mm) et des fractions granulométriques marqués avec l'Yb ont également été donnés pour estimer les taux de passage. L'étendue et le taux fractionnaire de la dégradation de la matière sèche et de la fibre détergent neutre variaient entre fourrages grossiers entiers et entre les populations granulométriques au sein de chaque fourrage, sans une règle spécifique de variation entre ces derniers. Le taux de passage fractionnaire du rumen a été également affectée par le type de fourrage et par la taille des particules, plus la taille est petite, plus le taux est rapide. Bien qu'il y avait de grandes différences entre la cinétique de dégradation, le taux de passage fractionnaire et la dégradabilité effective des échantillons broyés et les moyennes proportionnelles estimées à partir des différentes populations granulométriques, ceux-ci ne sont pas atteint une signification statistique. Il a été conclu que la technique des sachets de nylon ne imité pas complètement le comportement du composite de différentes tailles de particules constituant le matériau incubé, ce qui peut conduire à des erreurs graves lors de l'estimation de la cinétique de dégradation ou de transit.*

Mots-clés. *Dégradation – Fourrage – Taille des particules – Passage.*

I – Introduction

The nutritive value of forages for ruminants is mainly determined by their intake and digestibility, which in turn is affected to a great extent by the degradation and retention time in the rumen (Poppi *et al*., 1981). Degradation has long ago been determined by *in situ* studies which are widely used for forage evaluation due to their simplicity and low cost. Among other factors affecting *in situ* degradability values, particle size has been suggested as one of most relevant (Huntington and Givens, 1995). Degradation kinetics of different particle sizes have been widely investigated, although most of the papers dealt with feedstuffs ground through various screen sizes and measured rates of degradation of the whole material rather than with well-defined particle populations. Only the work by Ehle *et al*. (1982) and Emanuele and Staples (1988) considered the effect of narrowly defined particle sizes on their degradation kinetics. Their conclusion was that the mean particle size can differ among forages after grinding through the same screen size, and that different particle size fractions potentially can have different rates and extents of degradation. Degradation kinetics of the whole diet should then be a composite of the individual rates of the different particles sizes making up that diet multiplied by their relative contribution. However, some interactions between microbial populations attaching to different particles may occur making this not being so. A similar concern arises when studying transit kinetics. Faichney *et al*. (1989) clearly demonstrated that the kinetic behaviour of the whole digesta cannot be determined by its labelling with external markers, which should only be applied to particles in a relatively narrowly defined range of sizes when estimation of particle mean retention time is to be made. Although both degradation and transit kinetics parameters are needed to estimate the effective degradability, no attempts appear to have been made in order to determine them on the same particle size populations.

The objectives of the present study were: (1) to determine the effect of forage quality (untreated barley straw, ammonia-treated barley straw and lucerne hay) and particle size on their *in situ* degradation kinetics and rates of outflow; and (2) to determine if the behaviour of the whole forage may be estimated from their particle sizes.

II – Materials and methods

In a changeover design, chopped (5 cm) untreated straw (US), ammonia-treated straw (TS) and lucerne hay (LH) were offered *ad libitum* to three individually-housed sheep fitted with rumen and

duodenal cannulae, and fed at 2-h intervals. Milled (3-mm screen; 3Ø) subsamples of these forages and five particle size families (> 1.2, 1.2 to 0.6, 0.6 to 0.3, 0.3 to 0.15 and 0.15 to 0.045 mm) obtained by wet sieving of the ground material were incubated in the rumen for up to 96 h to study degradation kinetics. Particle size populations from each forage were incubated only in animals consuming that forage. Intraruminal doses of Yb-labelled whole ground (3-mm screen) forages and particle size fractions were also given to estimate outflow rates.

Outflow rates from rumen of Cr-EDTA were subjected to a one-way analysis of variance, whereas extent and fractional rate of degradation of DM in nylon bags, and fractional outflow rates of Yb-labelled particles were analysed as a split-plot design, with diet as main plot and particle size as subplot. Differences between treatment means were identified by the least significant difference (LSD). Statistical analysis was performed using the SAS statistical package (version 8.01). Extent and fractional rate of degradation of DM, DM fractional outflow rate and DM effective degradability of ground diets were compared to the composite made up from data for each particle size and their proportion in the ground material. Differences were tested for significance using paired t-tests.

III – Results and discussion

Degradation kinetics of DM (Table 1) changed with particle size. A significant interaction ($P<0.05$) diet x particle size was found for DM extent of degradation and as such there were no differences between diets for particles >1.2, 0.6 to 0.3 and 0.3 to 0.15 mm, whereas particles 1.2 to 0.6 and 0.15 to 0.045 mm showed lower values with diets LH and US, respectively. Extent of degradation of ground samples was lower for TS and higher for LH (differences significant at $P<0.05$), whereas US showed intermediate values ($P>0.05$). Differences between particle sizes varied with diet and hence 3Ø was not different from particles 1.2 to 0.3 and 0.15 to 0.045 mm for diet US, from particles >0.6 mm for diet TS and from particles 0.6 to 0.045 mm for diet LH. With respect to DM fractional rate of degradation, diet LH showed the highest value ($P<0.05$), whereas 3Ø was not different ($P>0.05$) from particles 1.2 to 0.3 and 0.15 to 0.045 mm. No interaction between diet and particle size was found for this variable.

Table 1. Extent (a+b; %) and fractional rate of degradation (c; h^{-1}) in nylon bags of dry matter from untreated barley straw (US), ammonia-treated barley straw (TS) and lucerne hay (LH) ground through a 3-mm screen (3Ø), and from their particle size fractions

Particle size (mm)		3Ø	> 1.2	1.2-0.6	0.6-0.3	0.3-0.15	0.15-0.045	mean	s.e.m.[1]
a+b	US	64	52	58	68	75	70	64	
	TS	59	56	65	73	80	81	69	3.8
	LH	71	48	49	67	76	80	65	
mean	65	52	57	69	77	77			
s.e.m.[2]				1.8					
c	US	0.017	0.022	0.023	0.022	0.022	0.036	0.024	
	TS	0.054	0.043	0.040	0.035	0.030	0.035	0.040	0.0084
	LH	0.102	0.058	0.072	0.092	0.070	0.102	0.083	
mean		0.058	0.041	0.045	0.050	0.040	0.058		
s.e.m.[2]				0.0047					

s.e.m.[1]: standard error of the mean of the analysis of variance for comparisons between diets.
s.e.m.[2]: standard error of the mean of the analysis of variance for comparisons between particle sizes.

The fractional outflow rates of Cr-EDTA and Yb-labelled particles are shown in Table 2. There were no significant differences between diets in the fractional outflow rates of Cr-EDTA (P>0.05). For the Yb-labelled particles there were differences between diets and size of particles (P<0.01), diet LH showing the higher fractional outflow rates with no statistical differences between straws. The smaller the size of the particle, the faster the rate of outflow, with particles >0.3 mm leaving the rumen at a significantly slower rate than particles of 0.3 to 0.045 mm. Ground diets also passed through the rumen significantly slower than these small particles.

Table 2. Rumen fractional outflow rates (h^{-1}) of Cr-EDTA and Yb-labelled particle size fractions (Yb-lab.) as affected by diet (US, untreated barley straw; TS, ammonia-treated barley straw; LH, lucerne hay) and particle size

Particle size (mm)		3Ø	> 1.2	1.2-0.6	0.6-0.3	0.3-0.15	0.15-0.045	mean	r.s.d.[1]
Cr-EDTA	US							0.069	
	TS							0.072	0.0327
	LH							0.100	
Yb-lab.	US	0.039	0.017	0.024	0.030	0.038	0.050	0.033	
	TS	0.022	0.060	0.053	0.054	0.052	0.076	0.053	0.0203
	LH	0.066	0.047	0.053	0.073	0.104	0.123	0.078	
mean		0.042	0.042	0.043	0.052	0.065	0.083		
r.s.d.[2]					0.0202				

3Ø: forages ground through a 3-mm screen. r.s.d. [1]: residual standard deviation of the analysis of variance for comparisons between diets. r.s.d. [2]: residual standard deviation of the analysis of variance for comparisons between particle sizes. There were four missing values.

Table 3. Extent (a+b; % ± S. E.) and fractional rate of degradation (c; h^{-1} ± S. E.) of dry matter, fractional outflow rate of Yb-labelled particles (k; h^{-1} ± S. E.) and dry matter effective degradability (DMED; % ± S. E.) of untreated barley straw (US), ammonia-treated barley straw (TS) and lucerne hay (LH,) ground through a 3 mm screen (3Ø), and of the composite (C) made up from data of the different particle sizes and their proportions in the ground samples, together with the mean differences (± S. E.) between 3Ø and C

	US	TS	LH
a+b			
3Ø	63.9 ± 5.15	59.3 ± 5.29	70.9 ± 2.29
C	68.4 ± 5.73	73.2 ± 2.70	69.3 ± 1.10
3Ø-C	-4.5 ± 5.06	-13.9 ± 2.99	1.5 ± 1.20
C			
3Ø	0.017 ± 0.0038	0.054 ± 0.0094	0.102 ± 0.0021
C	0.023 ± 0.0019	0.039 ± 0.0027	0.077 ± 0.0168
3Ø-C	-0.006 ± 0.0033	0.015 ± 0.0107	0.025 ± 0.0157
k			
3Ø	0.039 ± 0.0153	0.022 ± 0.0050	0.066 ± 0.0260
C	0.025 ± 0.0030	0.062 ± 0.0084	0.064 ± 0.0046
3Ø-C	0.013 ± 0.0234	-0.042 ± 0.0166	0.006 ± 0.0400
DMED			
3Ø	27.8 ± 6.77	46.0 ± 5.78	53.7 ± 1.66
C	43.3 ± 4.73	38.3 ± 0.49	48.2 ± 1.63
3Ø-C	-14.0 ± 6.73	6.1 ± 10.15	6.5 ± 3.97

Table 3 shows the extent and rate of degradation of DM, the fractional outflow rate of the particulate phase and the DM effective degradability of both the ground diets and the composite (C) made up from data of particle size fractions weighted by their proportion in the ground material. In general, estimated values (C) differed greatly from 3 Ø (up to 191% for TS fractional outflow rate). Correlation coefficients between 3 Ø and C were 0.336 ($P>0.1$), 0.829 ($P<0.01$), 0.213 ($P>0.1$) and 0.473 ($P>0.1$) for extent and fractional rate of degradation, fractional outflow rate and effective degradability, respectively. However, the comparison of 3 Ø and C within each dietary source of variation by means of paired t-tests did not result in statistically significant differences (except for TS extent of degradation, $P<0.05$).

IV – Conclusions

Although the high variability avoided statistical differences between 3 Ø and C, the large discrepancy between them led to the conclusion that the nylon bag technique has to be used carefully as it does not completely mimic the behaviour of the composite of the different particle sizes constituting the material incubated, and this may lead to severe errors when estimating degradation kinetics or flow of undegraded digesta out of the rumen. In addition, estimations of fractional outflow rate should be made from a narrow range of Yb-labelled particle sizes which depends on the diet considered.

References

Ehle F.R., Murphy M.R. and Clark J.H., 1982. *In situ* particle size reduction and the effect of particle size on degradation of crude protein and dry matter in the rumen of dairy steers. *J. Dairy Sci.,* 65, p. 963-971.

Emanuele S.M. and Staples C.R., 1988. Effect of forage particle size on *in situ* digestion kinetics. *J. Dairy Sci.,* 71, p. 1947-1954.

Faichney G.J., Poncet C. and Boston R.C., 1989. Passage of internal and external markers of particulate matter through the rumen of sheep. *Reprod. Nutr. Dévelop.,* 29, p. 325-337.

Huntington J.A. and Givens D.I., 1995. The *in situ* technique for studying the rumen degradation of feeds: a review of the procedure. *Nutr. Abstr. Rev. (Series B),* 65, p. 63-93.

Poppi D.P., Minson D.J. and Ternouth J.H., 1981. Studies of cattle and sheep eating leaf and stem fractions of grasses. III. The retention time in the rumen of large feed particles. *Aust. J. Agric. Res.,* 32, p. 123-137.