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In vivo digestibility of kleingrass from fecal nitrogen excretion

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Abstract

It was proposed that the digestibility of organic matter (OMD) can be estimated from the relationship between total fecal nitrogen (TFN, as a % of organic matter intake (OMI)) and fecal nitrogen concentration (FNc) through the equation: OMD = 1 – TFN / FNc. Two assumptions are critical to this equation, total fecal nitrogen (as a % of OMI) is a constant and does not change within a range of diet crude protein and fecal nitrogen concentration is proportional to digestibility of organic matter. The objective of this study was to test if total fecal nitrogen (as a % of OMI) remains constant over 3 feeding levels, and if fecal nitrogen concentration decreases with decreasing organic matter digestibility of maturing forages. Total fecal nitrogen did not change (P = 0.94) with feeding level, but increased (P < 0.05) with evaluation period. The fecal nitrogen concentration correlated (r = 0.60; P < 0.001) to digestibility of organic matter. The results show that digestibility of organic matter cannot be estimated from total fecal nitrogen, unless time of the year is considered.

Key Words: fecal index, nutritive value, N fecal, rams.

Resumen

Se ha propuesto que la digestibilidad de la materia orgánica (DMO) puede ser estimada a partir de la relación entre el nitrógeno total fecal (NTF, como un % del consumo de materia orgánica (CMO)) y la concentración de nitrógeno fecal (cNF), en la ecuación: DMO = 1 - NTF / cNF. La aplicación de esta ecuación se sustenta en dos supuestos, el nitrógeno total fecal permanece constante dentro de un rango de proteína bruta de la dieta y la concentración de nitrógeno fecal es proporcional a la digestibilidad de la materia orgánica. El objetivo de este estudio fue determinar si el nitrógeno total fecal (como un % del CMO) permanece constante sobre tres niveles de alimentación y si la concentración de nitrógeno fecal disminuye con la disminución de la digestibilidad de la materia orgánica. El nitrógeno total fecal no se modificó (P = 0,94) con el nivel de alimentación, pero incrementó (P < 0,05) con el período de evaluación. La concentración de nitrógeno fecal se correlacionó (r = 0,60; P < 0,001) con la digestibilidad de la materia orgánica. Los resultados demuestran que no es posible estimar la digestibilidad de la materia orgánica, a partir de la utilización del nitrógeno total fecal, sin la consideración del período del año.

Simple estimates of nutritive value are often of little value if there is no information regarding the amounts that will be consumed (Ørskov and Ryle 1990). The estimation of forage intake in grazing systems is, perhaps, the most challenging question of animal production.

The determination of individual intake can be obtained from fecal production (F) and diet digestibility. The precision of organic matter digestibility (OMD) determination is affected by the accuracy with which the forage samples represent the actual diet of the animals (Burns et al. 1994, Coates and Penning 2000). The use of the in vitro technique to estimate forage digestibility is associated with several errors, including the effects of diet composition, between animal variations, intake level and physiological status of animals (Schneider and Flatt 1975). The fecal index technique, alternatively, does not require diet samples, but only routine chemical determinations of fecal material, and is currently being used to estimate intake of wild and domestic herbivores (Caughley and Sinclair 1996, Hodgman et al. 1996, Mésochina et al. 1998). Fecal N concentration (FNc) has been widely used as a fecal index (Le Du and Penning 1982), due to its easy determination and low variation within 24-hour periods (Bartiaux-Thill 1980).

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Lancaster (1949a,1949b) proposed that OMD can be estimated from the relationship between total fecal nitrogen (TFN, as a % of organic matter intake (OMI)) and FNc through the equation: OMD = 1 – TFN/FNc. Two assumptions are critical to this equation: (1) TFN (as a % of OMI) is a constant and does not change within a range to diet crude protein, and (2) FNc is proportional to OMD. The objective of this study was to test: a) if TFN (as a % of OMI) remains constant over 3 feeding levels, and b) if FNc decreases with decreasing organic matter digestibility of maturing forages.

Materials and Methods

The study was performed at the Facultad de Agronomía, Universidad Nacional de La Pampa, Santa Rosa, La Pampa, Argentina (36° 46' S, 64° 16' W, 210 m ASL), during 1995. The forage was obtained from a pasture sown in spring 1994, of pure Kleingrass (*Panicum coloratum* L.) cv. Verde. At the beginning of each growth season (early October), the pasture was cut at 5 cm above ground to eliminate all standing dead forage, and fertilized with 60 kg urea/hectare.

Measurements of intake and digestibility were carried out during 4 experimental periods (I to IV), each one lasting for 16 days, with 11 days of adaptation and 5 days of data collection. Beginning day of each period was: 21 March, 2 May, 13 June,

and 25 July, 1995. This was done to obtain forage of different nutritive value for each trial.

Seventeen Pampinta rams in each period (51.3 ± 5.1 kg, 56.7 ± 2.9 kg, 66.4 ± 11.7 kg and 74.0 ± 6.7 kg in period I to IV, respectively) were grouped by weight, and then randomly assigned within weight group, to 3 feeding levels (treatments): 0.5 maintenance level (feeding level = **L1**; 5 rams), 1.0 maintenance level (feeding level = **L2**; 5 rams), and ad libitum, 1.5 the actual intake of 2 days previous to feeding (feeding level = **L3**; 7 rams). Maintenance level was estimated according to the energy requirements (AFRC 1993) of rams, and the in vitro DM digestibility of the forage (Stritzler et al. 1996). All animals were dewormed 15 days before the beginning of the study and housed in individual pens under continuous light, with free access to water.

The forage, accumulated from the beginning of the growing season, was cut by sickle at 5 cm above ground, before each feeding time (0800 and 1730 hours). Samples of offered forage were obtained before feeding. Refused forage of each experimental animal was removed from the manger every day, and weighed immediately. The animals were fitted with feces bags, secured to harnesses. Total fecal production was measured for each animal by weighting feces twice daily. Samples of forage offered and refused, and feces produced were obtained twice a day, dried at 55° C for 72 hours and ground through a 1-mm screen in a Wiley mill.

Organic matter intake (OMI) was determined by difference between offered and refused OM of food (Burns et al. 1994). In vivo organic matter apparent digestibility (OMD) was estimated by the method of total fecal collection, using the following equation 1:

$$OMD = [1 - F / OMI] \times 100 \quad (1)$$

Where:

OMD = In vivo OM apparent digestibility,
F = daily OM feces output, and
OMI = OM intake.

Chemical analyses and in vitro OM digestibility

Dry matter was determined at 105° C for 48 hours, and ash content was measured gravimetrically by igniting samples in a muffle furnace at 550° C for 12 hours in forage and fecal samples. Aliquots of dried samples were analyzed for total N concentrations by the semi-micro Kjeldahl procedure (2040 Digestion Unit and 1026 Distilling Unit, Tecator, Högänas, Sweden). Forage samples were additionally analyzed for crude protein (CP, N x 6.25), neutral-detergent fiber (NDF), acid-detergent fiber (ADF), and acid-detergent lignin (ADL), as described by Van Soest and Robertson (1985). In vitro organic matter digestibility (IVOMD) was estimated as described by Tilley and Terry (1963) and modified by Alexander and McGowan (1966). Samples were incubated at 39° C for 48 hours in a rumen fluid-artificial saliva solution, followed by an additional 48-hour period in 20% hydrochloric acid-pepsin solution. Inoculum for the procedure was obtained from rumen cannulated steers fed alfalfa hay. The in vitro values were adjusted by in vivo standards in each batch. The IVOMD values of consumed forage were estimated for each experimental animal, from the amount of OM and IVOMD of offered and refused forage (Meijs et al. 1982).

The total fecal nitrogen (as a % of OMI) was estimated from the nitrogen concentration, feces production and OMI.

Statistical analysis

The trial was carried out within a randomized block design, with a factorial arrangement of treatments. To test for differences in total fecal nitrogen as influenced by feeding levels and periods, the following model was used: Y = mean + block + period + levels + period x levels + error, where Y = total fecal nitrogen (as a % of OMI); period = Periods I to IV, replications of feeding trial, in which 17 rams, different between periods, were fed at 3 different levels (L1, L2, and L3), as explained above, and error = residual error (Steel and Torrie 1980). Statistical significance was determined using the GLM procedure (SAS Institute Inc. 1999). Mean separations were made using LSD at P = 0.05. Simple correlation coefficients between fecal nitrogen concentration and organic matter intake were determined using PROC CORR procedure of SAS Institute Inc. (1999). Paired t-test compared in vitro organic matter digestibility and predicted in vivo organic matter digestibility from total fecal nitrogen and fecal nitrogen concentration.

Table 1. Chemical composition and in vivo organic matter digestibility of deferred Kleingrass at 4 periods of evaluation.

	Period[1]			
	I	II	III	IV
	(%)			
Ash	8.4	8.5	7.4	7.6
Crude protein	9.7	6.8	6.6	5.9
Neutral-detergent fiber	65.4	69.4	71.1	70.6
Acid-detergent fiber	37.7	39.1	40.0	40.5
Acid-detergent lignin	5.2	3.7	4.9	3.6
In vivo organic matter digestibility	56.0	54.3	48.6	51.0

[1]I = 21 March to 6 April; II = 2 to 18 May; III = 13 to 29 June; IV = 25 July to 10 August.

Results and discussion

The chemical composition and in vitro organic matter digestibility of the forage offered in each evaluation period are shown in Table 1. All analyses were performed on pooled samples of all days of data collection. The feed quality declined with evaluation period (from I to IV), but the highest differences were found between periods I and II (Table 1).

The interaction between feeding level and evaluation period, for total fecal nitro-

Table 2. Total fecal nitrogen (as a % of OMI) at 3 feeding levels and 4 evaluations periods.

Feeding	Period[1]				
Level[2]	I	II	III	IV	Mean
	(% of OMI)				
L1	0.569	0.627	0.746	0.705	0.662
L2	0.523	0.592	0.773	0.743	0.647
L3	0.523	0.605	0.757	0.707	0.653
Mean[3]	0.537[c]	0.608[b]	0.758[a]	0.715[a]	
SEM				0.016	

[1]I = 21 March to 6 April; II = 2 to 18 May; III = 13 to 29 June; IV = 25 July to 10 August.
[2]L1 = 0.5 maintenance, L2 = 1.0 = maintenance, L3 = ad libitum.
[3]Means followed by a common superscript are not significantly different at (P > 0.05).

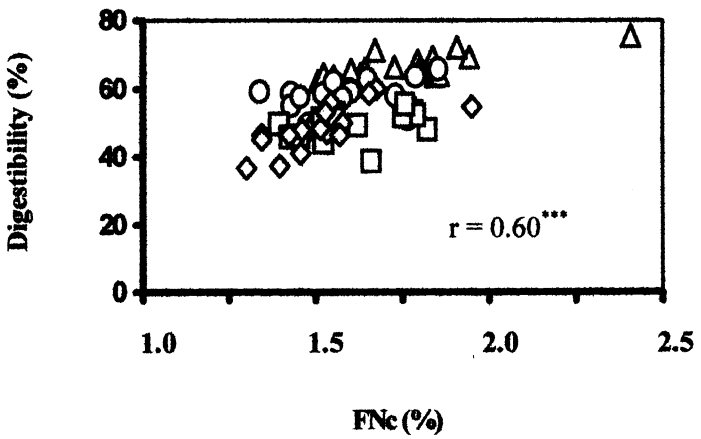

Fig. 1. Relationship between the fecal nitrogen concentration (FNc) and digestibility organic matter. Triangles correspond to 21 March to 6 April, circles 2 to 18 May, squares 13 to 29 June, and rhombus 25 July to 10 August period evaluation.

gen, was not significant (P = 0.70; Table 2). The total fecal nitrogen did not change (P = 0.94) with feeding level. This finding is in agreement with the results found by Lancaster (1949a) and Barrow and Lambourne (1962) and confirms Lancaster's first assumption that total fecal nitrogen (as a % of OMI) is constant and directly proportional to intake.

Insoluble N in feces comes largely from feed (Ørskov 1982), although a small contribution of N bound to indigestible cell wall of rumen bacteria should be also taken into account. The soluble N present in feces is mostly microbial, and includes considerable ammonia produced by cecum-colon bacteria (Van Soest 1994). Although much of the fecal N may originally have been endogenous, before excretion it has been converted to microbial N through fermentation in the hindgut (Mason 1969). The amount of excreted N depends, partially, on the microbial N yield (Van Soest 1994), on the digestion site (rumen or cecum-colon) (Ørskov et al. 1972) and the digestion extent (Arman et al. 1975). When forages of similar nutritive value are considered, total fecal nitrogen (as % of OMI) keeps constant with feeding level because microbial N yield is propotional to intake.

The total fecal nitrogen, however, increased (P < 0.05) from evaluation periods I to III. It seems likely that this increase is associated to changes in digestion site; according to Thomas (1988), with good-quality forages, 5–15% of cell wall carbohydrates are fermented in the cecum-colon. As forages mature, this proportion increases; Hogan et al. (1969) found that up to 25% of the total digestion of low quality grasses occurs in the hindgut. Rumen microbes, but not colon-cecum microbes, are exposed to the host animal's enzymes (Mason 1969); therefore the fermentation site might affect the amount of total fecal nitrogen (Ørskov et al. 1972). The increasing proportion of feed carbohydrates fermented in cecum-colon would affect mostly the soluble N in feces (Ørskov et al. 1972), whilst the insoluble N should not be changed.

The fecal nitrogen concentration was correlated with organic matter digestibility (r = 0.60; P < 0.001; Fig. 1) across all 4 periods. Correlations within each period were also obtained; they were all significant (P < 0.05) but not high (r = 0.77, 0.67, 0.50, and 0.66 for period I, II, III and IV, respectively), due to large between experimental animals variations of total fecal nitrogen excretion.

Although fecal nitrogen concentration increased with organic matter digestibility, the correlation across the 4 periods was not high. This would allow us to infer that total fecal nitrogen, as determined in this study, changes with period. In other words, for a given value of fecal nitrogen concentration obtained in different periods, the organic matter digestibility would be different. When analyses were run within each experimental period, the correlations were significant.

The estimations of digestibility by the in vitro technique and the fecal N index were not different (P > 0.05; Table 3) in 3 of the 4 periods. The means of both methods across periods were different (P < 0.05). However, this difference was only of 1.5%.

The comparison of indirect techniques to predict in vivo digestibility of consumed forage presents limitations. The main problem of the in vitro technique is the collection of samples representative of the diet consumed by the animal. The fecal N index technique requires a feeding trial to estimate total fecal nitrogen (as % of OMI), using the same forage to be grazed. The nitrogen fecal concentration is then assessed in fecal samples from the grazing animals, and diet digestibility can be predicted from the equation: OMD = 1 – TFN / NFc. The usefulness of this technique is restricted to situations where the forage to be grazed can also be cut to run a feeding trial simultaneously. The main advantages of the technique are that the analytical requirements are low and simple, and does not require diet sampling.

Table 3. Estimated organic mater digestibility (OMD) of consumed herbage using either in vitro organic matter digestibility or fecal N index in Periods I to IV (n = 17).

Period[1]	In vitro OMD	Fecal N index[2] OMD = (1 – TFN / FNc) * 100
	(%)	
I	59.5[b]	62.5[a]
II	54.6[a]	56.3[a]
III	49.3[a]	50.3[a]
IV	51.5[a]	51.9[a]
Media	53.8[b]	55.3[a]

Means in the same row followed by a common superscript are not significantly different at (P > 0.05).
[1]I = 21 March to 6 April; II = 2 to 18 May; III = 13 to 29 June; IV = 25 July to 10 August.
[2]TFN, total fecal nitrogen; FNc, fecal nitrogen concentration.

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