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A comparison between protein disappearance from the mobile bag and acid detergent solubility of nitrogen as estimates of protein digestibility in ruminants

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ABSTRACT

Protein disappearance from the mobile nylon bag and acid detergent solubility of protein (ADSN) were estimated in different concentrate feedstuffs (guar meal, rapeseed meal, cottonseed meal, sunflower meal, dried sugar beet pulp, corn distillers dried grain, maize feed meal, maize gluten feed, pig hair meal, feather meal, hair-feather meal). Estimates with the mobile bag method included a 16 h preincubation in the rumen. Protein degradability varied both within and between feeds and degradabilities of feeds of plant origin were higher than for feeds of animal origin. The N content was generally higher in feeds of animal origin compared to feeds of plant origin, which makes animal origin feeds a good source of undegraded dietary protein. For corn distillers dried grain, disappearance obtained with the mobile bag was higher than acid detergent solubilities, whereas the opposite was the case for all other feeds tested, and most pronounced for feeds of animal origin and for dried sugar beet pulp. Generally, there was a low correlation between results obtained with the mobile bag and with acid detergent. If it is accepted that protein disappearance from the mobile bag is a good estimate for *in vivo* true digestibility, our results indicate that ADSN is not a satisfactory method for estimation of protein digestibility.

KEY WORDS: degradability, digestibility, mobile bag, ADF, ADIN, nitrogen, protein, ruminants

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INTRODUCTION

Intestinal digestibility of undegraded feed protein is an important parameter in modern protein evaluation systems for ruminants. Several methods have been used in order to estimate true intestinal digestibility of feedstuffs (Hvelplund, 1985; Antoniewicz et al., 1992; Susmel et al., 1994). The mobile nylon bag method has been accepted as the most promising method for measuring intestinal digestibility of various feedstuffs such as forages and concentrates and for detecting the impact of, for example, treatment and extent of rumen degradation (Varvikko and Vanhatalo, 1990; Frydrych, 1992; Hvelplund et al., 1992).

The digestibility values used in the new protein evaluation systems are the true digestibilities of rumen undegraded feed protein. These digestibilities vary according to rumen degradability (Hvelplund et al., 1992). However, it was shown by Hvelplund et al. (1992) that the true digestibility of rumen undegraded protein can be estimated at an arbitrary rumen degradability if total true digestibility (or the true indigestible fraction) of the protein is known. Further, the estimation of total true digestibility using the mobile bag often requires rumen preincubation, which has been shown to be necessary for starch--containing feeds (Volden and Harstad, 1995), tropical forages (Mgheni et al., 1994) and temperate forages (Vanhatalo et al., 1996). An estimate of true digestibility of rumen undegraded protein can thus be obtained from a method which can estimate the true indigestible fraction of the protein. Although the mobile bag method is much less laborious than the alternative *in vivo* infusion method for estimation of true digestibility (Hvelplund, 1985), it is still too work-consuming for routine feed evaluation, moreover, it requires access to intestinally fistulated animals. Therefore, it is important to develop laboratory methods for estimation of digestibility. Different approaches have been used, including enzymatic methods (Antoniewicz et al., 1992; Calsamiglia and Stern, 1995) and acid detergent soluble nitrogen (Goering et al., 1972; Krishnamoorthy et al., 1982).

The aim of this study was to compare estimates of protein digestibility obtained with the mobile bag method and by acid detergent solubility of protein for various concentrate feedstuffs.

MATERIAL AND METHODS

Feed samples

Forty-five samples of different concentrate feedstuffs (guar meal, rapeseed meal, cottonseed meal, sunflower meal, dried sugar beet pulp, corn distillers

dried grain, maize feed meal, maize gluten feed, pig hair meal, feather meal, hair-feather meal) were examined. Table 1 shows the number of samples examined within feed type and the range of their nitrogen (N) content. For *in sacco* analysis, samples were ground through a 1.5 mm screen, except pig hair meal, feather meal, and hair-feather meal which already were finely ground during the processing. For chemical analysis the samples were ground through a 1 mm screen.

In sacco procedures

Rumen protein degradability was determined according to Madsen et al. (1995) using nylon bags (36x36 mm pore size) incubated in three cows fed a standard diet. Disappearance of N was measured with the mobile nylon bag technique according to Hvelplund et al. (1992) modified with rumen pre-incubation. Approx. 1 g of the sample was weighed out in each bag (9x9 mm pore size, external dimensions 6x6 cm), then the bags were heat sealed and preincubated in the rumen of cows for 16 h. After rumen preincubation the bags were incubated first in an HCl solution pH 2.4 for one hour to adjust pH and then in a pepsin-HCl solution (100 g pepsin (1:10000 Orthana 23208) per l of 0.004 M HCl solution, pH 2.4) for 2 h at 39°C. After pepsin-HCl incubation the bags were introduced into the small intestine through the duodenal cannula, 2 bags in each of three cows per sample. After recovery from the faeces the bags were washed thoroughly in running tap water before determination of the nitrogen content in the residues.

Chemical analysis

The feedstuffs and the residues in the bags were analyzed for dry matter (DM), nitrogen (N), acid detergent fibre (ADF) and acid detergent insoluble N (ADIN) according to conventional methods (AOAC, 1990). To avoid bumping during acid detergent boiling, the amount of sample was reduced to 0.5 g for pig hair meal, feather meal and hair-feather meal.

Calculations

Effective protein degradability was calculated on the basis of a 5% particle passage rate and corrected for particle loss from the nylon bags (Weisbjerg et al., 1990). Mobile bag disappearance of nitrogen was estimated as the loss of nitrogen from the bags. ADIN was calculated following the AOAC procedure (AOAC, 1990). Proc GLM (SAS, 1985) was used for regression analysis.

TABLE 1

Number of samples and range of N content, effective protein degradability, disappearance from the mobile bag, acid detergent fibre (ADF) and acid detergent soluble nitrogen (ADSN) of feedstuff groups

Feedstuffs	n	N content % in DM	Effective protein degradability, %[1]	Disappearance from the mobile bag, %[2]	ADF % in DM	ADSN % in N
Guar meal	2	7.0 – 7.1	64.0 – 66.1	91.9 – 94.3	14.7 – 15.8	99.0 – 99.2
Rapeseed meal	6	6.0 – 7.6	53.1 – 72.2	85.5 – 90.5	19.7 – 26.5	86.9 – 91.2
Cottonseed meal	10	5.4 – 8.7	36.9 – 61.2	88.9 – 96.4	8.3 – 30.2	89.8 – 97.8
Sunflower meal	4	5.9 – 7.2	69.5 – 75.0	95.0 – 96.1	22.1 – 33.8	93.4 – 95.4
Dried sugar beet pulp	6	1.4 – 1.9	41.7 – 56.8	73.8 – 85.5	26.0 – 28.9	86.9 – 92.4
Maize distillers dried grain	3	4.4 – 4.9	26.2 – 40.4	88.1 – 91.8	13.0 – 13.6	82.1 – 88.6
Maize feed meal	2	1.9 – 2.6	52.4 – 63.8	89.0 – 91.2	6.2 – 11.2	92.9 – 97.5
Maize gluten feed	2	3.5 – 3.7	72.4 – 72.8	92.2 – 92.5	8.6 – 11.7	95.5 – 97.5
Pig hair meal	4	14.8 – 15.0	9.3 – 15.5	57.8 – 60.9	10.8 – 29.9	86.0 – 90.0
Feather meal	3	14.1 – 14.4	9.7 – 12.6	68.0 – 69.3	4.1 – 9.8	86.1 – 91.1
Hair–feather meal	3	14.8 – 14.9	10.4 – 12.7	60.6 – 62.5	4.7 – 11.3	83.1 – 88.3

[1] on the basis of a 5% particle passage rate and correction for particle loss

[2] measured using the mobile nylon bag method including a 16 h preincubation in the rumen

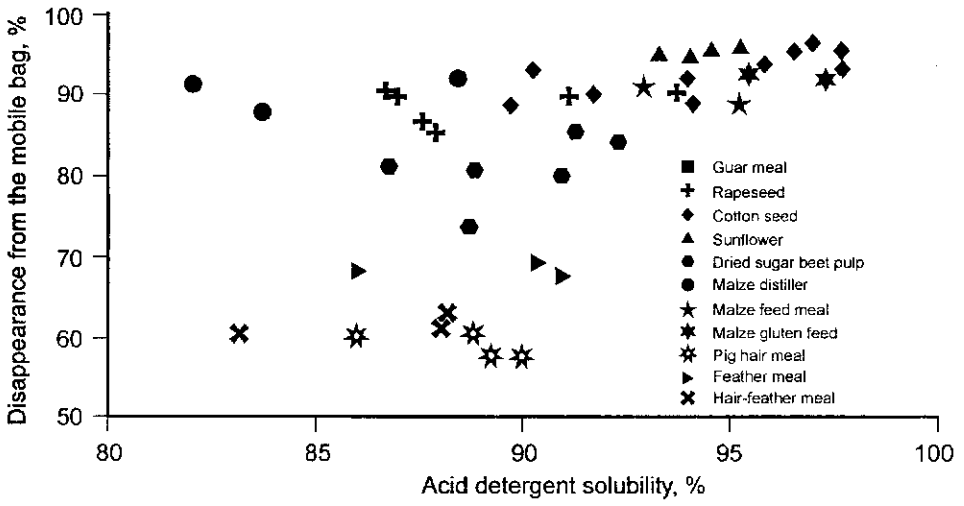

Figure 1. The relationship between nitrogen disappearance from the mobile bag and acid detergent soluble nitrogen

RESULTS

As shown in Table 1, the examined feed samples covered a wide spectrum of N content, ranging from 1.4% (dried sugar beet pulp) to 15.0% (pig hair meal) in DM. Rumen degradability, disappearance of N obtained with the mobile bag method and ADSN varied substantially among and within different protein sources. Effective rumen degradability varied between 9.3 (pig hair meal) and 75.0% (sunflower meal), while disappearance from the mobile bag varied between 57.8 (pig hair meal) and 96.4% (cottonseed meal). ADSN varied from 82.1 (corn distillers dried grain) to 99.2% (guar meal). The large variability between the samples makes this sample set very suitable for testing the correlation between the two methods.

Disappearance from the mobile bag is plotted against ADSN in Figure 1. It is obvious that the correlation between the two methods is very low. Generally, samples of animal origin had a low disappearance from the mobile bag compared to ADSN. In contrast, corn distillers dried grain had a low ADSN compared to the disappearance from the mobile bag. Regression analysis on all samples and the different feed types are shown in Table 2. The regression analysis shows, as did Figure 1, an unsatisfactory correlation between disappearance from the mobile bag and ADSN. Overall, the multiple correlation coefficient is as low as 0.30, and it does not improve by dividing feeds according to animal or plant

TABLE 2
Regression coefficients for the model: disappearance from the mobile bag, % = $\beta_0 + \beta_1$ ADSN, %

Feedstuff	N	β_0	β_1	P	R^2
Total	45	−61.6	1.59	<0.0001	0.30
Feeds of plant origin	35	30.8	0.64	0.0008	0.29
Feeds of animal origin	10	30.0	0.37	0.6	0.04
Rapeseed meal	6	62.8	0.29	0.4	0.15
Cottonseed meal	10	29.8	0.66	0.01	0.54
Dried sugar beet pulp	6	−10.9	1.02	0.3	0.26
Maize distillers dried grain	3	67.1	0.27	0.7	0.22
Pig hair meal	4	115.8	−0.63	0.3	0.46
Feather meal	3	64.4	0.04	0.9	0.03
Hair-feather meal	3	38.1	0.27	0.3	0.77

origin. Analysis according to individual feed type groups (where the sample number >2) shows that even within a feed group, the correlation is low and significant only for cottonseed meal.

DISCUSSION

ADIN has been used to detect heat damage of protein feedstuffs since Goering et al. (1972) showed it to be highly correlated to *in vivo* apparent digestibility on a number of silage samples, of which some were expected to be heat damaged. Based on the work of Webster et al. (1986) the British Metabolizable Protein System (AFRC, 1992) predicts the digestibility of rumen undegraded protein using ADIN analysis, using the assumption that ADIN is undegradable in the rumen and indigestible in the intestine and that 90% of the rumen undegraded protein which is not ADIN is digestible. It has been shown, however, that, especially for feeds like distillers grains, where Maillard reactions have increased the natural content of ADIN, part of that ADIN is degradable in the rumen and, therefore, ADIN is not satisfactory for determination of the unavailable fraction in this type of feeds (Weiss et al., 1989; Waters et al., 1992). However, ADIN analysis on the rumen undegradable fraction can predict digestibility of undegraded protein in distillers grains too (Chaudhry and Webster, 1993).

In accordance with this, the present experiment showed that ADIN heavily overestimated the unavailable fraction of corn distillers dried grain, as ADIN was up to twice as high as the fraction which was found to be indigestible using the mobile bag method.

For the animal feeds (feather and hair meal) tested in this experiment, the opposite problem appeared. For these feeds, the protein fraction which was

unavailable using the mobile bag method was two to three times as high as the ADIN fraction, which probably mainly consists of keratins and Maillard products. This is probably due to more efficient hydrolysis of sulphuric bridges using acid detergent compared to the more physiological conditions applied with the mobile bag method. For dried sugar beet pulp the ADIN fraction also seemed to underestimate the indigestible fraction compared with the mobile bag method. However, this could partly be due to microbial contamination of the undegraded residues in the mobile bags.

For the rest of the vegetable feeds the discrepancy was less pronounced, but the correlation between the two methods for estimating digestibility was poor, and only cottonseed meal showed a significant correlation. Therefore, the ADIN procedure seems to be unsatisfactory for estimation of digestibility, if it is accepted that the disappearance from the mobile bag is a physiologically reasonable estimate for *in vivo* true digestibility, as shown by Hvelplund et al. (1994). However, ADIN analysis is probably a valuable method for detecting heat damage within feed type.

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STRESZCZENIE

Porównanie rozkładalności białka oznaczanego metodą woreczków mobilnych i rozpuszczalności azotu w kwaśnym detergencie jako wskaźników strawności białka u przeżuwaczy

W różnych paszach treściwych (mączka z guaru, śruty pockstrakcyjne z rzepaku, bawełny, słonecznika, suszone wysłodki buraczane, suszony wywar z kukurydzy, mączka pastewna z kukurydzy, pastewny gluten kukurydziany, mączka z włosów świńskich, mączka z pierza, mączka z włosów i pierza) oznaczano ubytek białka z nylonowych mobilnych woreczków oraz rozpuszczalność białka w kwaśnym detergencie (ADSN). W metodzie mobilnych woreczków zastosowano 16 godz. pre-inkubację w żwaczu. Stopień rozkładu białka różnił się tak w obrębie jak i pomiędzy paszami, a rozkładalność pasz pochodzenia roślinnego była większa niż pochodzenia zwierzęcego. Zawartość N była ogólnie większa w paszach pochodzenia zwierzęcego niż w paszach roślinnych, co wskazuje, że pasze pochodzenia zwierzęcego są dobrym źródłem nierozkładalnego białka dawki. W przypadku suszonego wywaru z kukurydzy ubytek białka był większy przy zastosowaniu metody mobilnych woreczków niż ADSN w przeciwieństwie do wszystkich badanych pasz, i wyraźniejszy w przypadku pasz pochodzenia zwierzęcego oraz suszonych wysłodków buraczanych.

Podsumowując, otrzymano niską korelację pomiędzy wynikami otrzymanymi przy zastosowaniu metody woreczków mobilnych i ADSN. Jeżeli przyjmie się, że rozkład białka oznaczony przy pomocy woreczków mobilnych jest dobrym wskaźnikiem *in vivo* rzeczywistej strawności, to nasze wyniki wskazują, że ADSN nie jest zadowalającą metodą oznaczania strawności białka.