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Use of Neutral Detergent Insoluble Protein in Omasal Samples to Estimate Escape Protein

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Neutral detergent insoluble protein in omasal samples can be used to estimate the escape protein in forages. These escape protein values will help develop protein supplementation strategies for grazing cattle.

Summary

Three experiments were conducted with ruminally cannulated steers to evaluate neutral detergent insoluble protein (NDIP) content of omasal samples as an escape protein estimation technique. In experiment 1, brome hay escape protein (EP) values were determined with the NDIP technique and the omasal sampling purine technique (OSPT) with no difference in EP values. In experiment 2, smooth bromegrass EP values were estimated in June and August, 1996. In situ and OSPT estimated similar EP values, but EP estimates from NDIP were lower. Experiment 3 was conducted from May through July, 1997. OSPT estimated the highest EP values. In experiments 2 and 3, NDIP had better correlation with OSPT than the in situ technique.

Introduction

Estimation of forage protein degradation in the rumen is required to determine amount and type of protein needed to meet metabolizable protein requirements of grazing animals. Escape protein content of forages can be estimated with in vivo, in situ and in vitro techniques. Each technique has advantages and disadvantages.

Despite the difficulties of making in vivo measurements of protein degradation, such measurements are essential. They serve as a standard against which all chemical or in vitro methods of estimating protein degradation are compared. Usually, in vivo measurements are performed with surgically prepared animals equipped with ruminal and abomasal or small intestinal cannulae. Omasal sampling, however, can determine escape protein value of forages without abomasal or intestinal cannulae. Correct estimation of microbial and endogenous protein are the major problems of the in vivo technique. Neutral detergent (ND) solution has ability to remove microorganisms and endogenous N from omasal samples and can estimate undegradable protein content of forages. Additionally, the ND insoluble protein content of fecal samples can be used to determine digestibility of forage escape protein. The objective of these experiments was to determine whether ND insoluble protein content of omasal samples can be used to determine the escape protein content of brome hay and smooth bromegrass.

Procedure

In experiment 1, three ruminally cannulated steers (836 lb) were fed 1.5 of percent BW with coarsely chopped brome hay during two periods. In each period, dietary adaptation and marker steady-state conditions were established for seven days and omasal and fecal sample collection occurred on days seven and nine. In the first period, each animal received two different external markers (ytterbium and erbium) twice daily in two different forms (as a solution or as labeled hay). In the second period, the form of the two was reversed. Acid insoluble ash (AIA) was used an internal digesta particulate marker.

In experiment 2, four ruminally cannulated steers (774 lb) were used to collect diet, omasal and fecal samples when they were grazing smooth bromegrass in June and August 1996. In each month (period), the first 10 days were used for adaptation to grass while days 13, 16 and 19 were omasal and diet sampling days. Fecal grab samples were collected daily after day 10 and composited according to omasal sampling days.

Experiment 3 was conducted with three ruminally cannulated steers (722 lb) during May 9 through July 24, 1997. In addition to AIA, chromium releasing devices were used as external digesta flow markers and were replaced five times. In each period, days one through five were used to develop steady-state marker concentration while days five, eight, 11 and 14 were used for omasal and diet collection. After collection, samples were frozen and freeze dried. OSPT assumes there are three major fractions of N in the omasal sample: 1) plant N, 2) microbial N and 3) endogenous N. Microbial N was determined using purine analysis and purine:N ratio (.2), which assumes all plant purines are digested in the rumen and omasal purine contents originate from microorganisms. Amount of endogenous N used was 2.2 g for each kg DM intake. Plant N in the omasum was calculated by subtraction of microbial N and endogenous N from the total omasal N. NDIP technique used neutral detergent insoluble protein content of omasal samples as a measure of plant protein. Rumen undegraded portion of slowly degradable true protein (B3) and unavailable protein content (C fraction) of forage proteins were equal to EP in the NDIP. Rapid (B1) and intermediate (B2) degradable portions of the true protein are assumed to be degraded in the rumen. In both techniques, EP is expressed as a percentage of forage DM using internal and external particulate digesta marker ratios. In situ neutral detergent fiber nitrogen technique (1997 Nebraska Beef Cattle Report, pp. 13) was used to estimate EP values. Escape

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protein digestibility was determined using differences between neutral detergent insoluble protein content of omasal and fecal samples.

Results

In experiment 1, NDIP and OSPT estimates of brome hay EP were not significantly different (P>.05) when AIA was used as an internal marker (Table 2). Also, techniques estimated EP values similarly with external markers (P>.05), but external markers overestimated EP values. External markers did not accurately predict either particulate DM flow to the omasum or DM digestibility in the rumen. External and internal markers estimated rumen DM digestibilities of 18 and 49 percent , respectively. In experiment 2, EP values measured using the NDIP technique were different than OSPT and in situ EP values (P<.05; Table 2), but OSPT and in situ technique were not different (P>.05). In the last experiment, EP values measured by all three techniques were different when a chromium-releasing device was used as a particulate marker (P<.05). With AIA as the marker, NDIP and in situ techniques were not different (P>.05), but OSPT resulted in the highest EP values (Table 2). NDIP was better correlated with OSPT than the in situ technique in experiments 2 and 3 (Table 3). Coefficients of variation (CV) for the NDIP technique were always less than OSPT (Table 4). Escape protein digestibilities determined using NDIP content of fecal and omasal samples were 45, 32 and 57 percent of EP in experiments 1, 2 and 3, respectively.

Comparison of two omasal sampling techniques and an in situ technique is difficult because each technique has its own assumptions, making escape protein values relative, not absolute. Separation of microbial N and endogenous N from feed N has associated errors. No microbial marker can absolutely quantify microbial protein and microbial marker analysis requires multiple steps and complicated equipment. Purine was the marker chosen for this research. Purine: N ratio used was .2, but in the second period of experiment 2, a ratio of .14 (1997 Nebraska Beef Cattle Report, pp.

Table 1. Nutrient composition of brome hay and bromegrass on a dry matter basis.

Item[a]	Brome hay	SBG[b]	SBG[c]
Crude protein,%	8.16	15.03	17.51
ADF, %	43	—	37
NDF, %	72	64	64
IVDMD, %	55	62	62
AIA, %	2.93	3.27	3.83

[a] ADF-acid detergent fiber; NDF-neutral detergent fiber; IVDMD-in vivo dry matter disappearance; AIA-acid insoluble ash.
[b] Smooth bromegrass (SBG) diet samples collected from four ruminally cannulated steers in June and August, 1996.
[c] Smooth bromegrass (SBG) diet samples collected from three ruminally cannulated steers May 9 through July, 1997.

Table 2. Escape protein estimation with three different techniques on a dry matter basis.

	Technique			
	OSPT[a]	NDIP[b]	IN SITU	SE[c]
Experiment 1				
With AIA[d] ,%	1.04	1.29	—	.10
External markers, %	2.13	2.37	—	.18
Experiment 2				
With AIA,%	2.14	1.52	2.68	.19
Experiment 3				
With AIA, %	3.29	2.35	2.18	.16
With Cr[e], %	4.71	3.08	2.18	.19

[a]Omasal sampling purine technique.
[b]Omasal sampling neutral detergent insoluble protein technique.
[c]Standard error of techniques.
[d]Acid insoluble ash.
[e]Chromium.

Table 3. Correlation coefficients of techniques for estimating escape protein in forages.

	NDIP- OSPT	NDIP- In situ	OSPT-In situ
Experiment 2[a]	.66	.08	.15
Experiment 3[a]	.71	.13	.16
Experiment 3[b]	.62	-.12	-.05

[a]Acid insoluble ash used as a marker.
[b]Chromium releasing device used as a marker.

Table 4. Coefficients of variation of techniques for estimating escape protein in forages.

	NDIP[a]	OSPT[b]	In situ
Experiment 1			
AIA[c]	6.38	21.67	—
External markers[d]	22.66	50.14	—
Experiment 2			
AIA	25.31	32.83	21.71
Experiment 3			
AIA	19.06	41.75	21.44
Cr_2O_3	28.04	46.32	21.44

[a]Omasal sampling neutral detergent insolubleprotein technique.
[b]Omasal sampling purine technique.
[c]Acid insoluble ash as a digesta particle marker.
[d]Ytterbium, Erbium labeled hays and solutions used as external markers.

13) gave closer estimates to the other two techniques. Endogenous N values were estimated using reference values which vary. Endogenous N was assumed to be 2.2 g for each kg DM intake. The in situ technique is not an in vivo technique. Restriction of microbial access to the protein, bag size, bag porosity, sample quantity, sample particle size, incubation time, diet and animals are all concerns. Passage rate is required to calculate escape protein in the in situ technique. External markers are used to calculate passage rate, but they do not estimate lag prior to passage.

NDIP can estimate EP values simply using the ratio of internal particulate marker in the diet and the omasal contents with ND insoluble protein content of omasal contents. Therefore, NDIP can estimate EP contents of brome hay and smooth bromegrass more precisely, simply and faster than OSPT, and provide reasonable EP digestibility values. NDIP is superior to the in situ technique because it is an in vivo technique and better correlated with OSPT. Use of NDIP to measure the EP content of forages will enhance our understanding of the protein components of forages.

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