

# Evaluation of Pepsin-Insoluble Nitrogen as a Marker for Determination of Digestibility of Forages

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## Story in Brief

Digestibility of the different nutrient components of hay from four selections of Old World bluestems was estimated by a conventional method and use of pepsin insoluble nitrogen (PIN) as an internal marker. Digestibility values obtained by the conventional method were higher than those obtained by using PIN. Average recovery of PIN in the feces of lambs averaged 78.6 and varied from 72.1 to 86 percent for different varieties of the hay. This component of the forage would not be a satisfactory internal indicator for estimation of digestibility of various nutrient components of hay.

## Introduction

Various materials have been used as markers to estimate the digestibility of different nutrient components of feeds. Less labor is required than for determination of digestibility by the conventional total collection method, and estimates often can be made under conditions more similar to those common for producing animals. These advantages are partially offset, however, by certain factors which tend to limit the usefulness of markers. Some of the more important limiting factors include difficulty with analytical procedures, uneven distribution of some markers in the digesta, consumption of extraneous sources of a marker, and lack of complete recovery in the feces.

Acid insoluble ash is a natural marker useful for determination of digestibility under conditions where feed refusals are minimal and the animals do not have access to extraneous sources of the ash. Several researchers have evaluated lignin as a marker in digestibility studies of forages; however, variation in recovery of the material has been observed, possibly because of problems in the analytical procedures used. Other internal markers tested include plant chromogens and fecal nitrogen. The former are essentially limited to fresh green forages, and problems relating to specificity of the compounds actually measured have been noted. Efforts to develop equations to relate fecal nitrogen to digestibility of forages have not been entirely successful.

The purpose of this study was to evaluate pepsin insoluble nitrogen (PIN) as an internal marker for determining the digestibility of various nutrient components of hay.

## Materials and Methods

The samples for this study were collected during a conventional digestion trial in which digestibility of hays from four selections of Old World bluestems

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was determined (Londoño et al., 1982). The varieties compared using 16 young lambs in sequences of a replicated 4 × 4 Latin Square design were:

Variety code	Scientific name
WW-506	<i>Bothriochloa ischaemum</i> var. <i>sangarica</i>
WW-573 (WW Spar)	<i>Bothriochloa ischaemum</i> var. <i>ischaemum</i>
WW-477	<i>Bothriochloa ischaemum</i> var. <i>sangarica</i>
WW-517	<i>Bothriochloa intermedia</i> var. <i>indica</i>

Each hay was chopped and fed to the animals twice daily in sufficient quantity to allow some feed refusal in most instances. Soybean meal (75 g. per head per day) was added to assure adequate protein intake for maintenance. A mineral supplement containing 13 to 15 percent calcium, 7 percent phosphorus, and 30 to 36 percent salt was fed at the rate of 10 g. head per day. In addition, a trace mineral supplement was available for ad libitum consumption. Experimental periods were 2 weeks, with feed refusals and feces collections during the last six days of each period. Feces samples were dried in the oven (95°C) and next were ground through a 1 mm screen in a Wiley mill and kept for future composites. Pepsin-insoluble nitrogen was determined by method of Goering and Van Soest (1970).

## Results and Discussion

The chemical composition of the four varieties of hay fed in the digestibility trial was similar (Table 1). Crude protein content was relatively low, whereas acid detergent fiber (ADF) and neutral detergent fiber (NDF) were high, reflecting an advanced stage of maturity of the material at harvest.

Recovery of the pepsin insoluble nitrogen (PIN) consumed by the lambs averaged 78.6 percent. Presumably, a portion of the PIN in the hays was digested during passage through the digestive tract of the lambs. As noted above, the fecal samples were dried at 95°C which is higher than the recommended temperature for materials to be analyzed for PIN; however, drying at a lower temperature could only have resulted in even lower PIN values for the feces and a lower percentage recovery.

The percentage of the PIN intake recovered in the feces was lower than the value, i.e., 90 percent or higher, generally considered acceptable for a substance to be used as a marker in digestibility studies. Moreover, there was

**Table 1. Chemical composition of Old World bluestem hays<sup>a</sup>**

Variety	Crude protein	Acid detergent fibr	Neutral detergent fiber	Lignin	Cellulose	Pepsin insoluble nitrogen
			%			
WW-506 <sub>b</sub>	6.7	45.6	74.0	6.1	35.8	.54
WW-573	6.6	44.0	72.8	5.8	34.3	.48
WW-477	7.0	43.6	70.8	5.7	34.0	.54
WW-517	6.1	44.3	73.4	5.6	35.1	.45

<sup>a</sup>Dry matter basis.

<sup>b</sup>Released for production as WW-Spar.



considerable variation among varieties of the hay in the extent of PIN recovery in the feces of the lambs in this trial (Table 2). The low recoveries of PIN were reflected in considerably lower estimates of digestibility of different components of the forages (Table 3). After calculation of digestibility coefficients for different nutrient components using a correction factor for PIN in the feces based on the average recovery, significant differences ( $P < .05$ ) remained between methods of determination. Also, estimates of digestibility by the PIN method did not rank the varieties of hay in the same order as did estimates by the total collection method.

Based on the results of this trial, it was concluded that unless some change in the analytical procedure or other factor were to eliminate the variation among varieties of forage in PIN recovery in the feces of test animals, this component of forage would not be a satisfactory internal indicator of nutrient digestibility. Further research would be required to determine whether this component of feed could be used as a marker for determination of digestibility of other types of feeds.

### Literature Cited

Londoño, I., et al. 1982. Okla. Exp. Station. MP-112:123-126.

**Table 2. Total intake and excretion of pepsin insoluble nitrogen (PIN) by lambs**

Variety	PIN intake	PIN in feces	Recovery <sup>a</sup>
	(g)	(g)	(%)
WW-506	20.1	14.5	72.1
WW-573	18.4	15.0	81.5
WW-477	20.0	15.0	75.0
WW-517	17.1	14.7	86.0

<sup>a</sup>Pooled standard error for treatment mean was .94.

**Table 3. Digestibility of dry matter and crude protein using total collection method (TCM) and pepsin insoluble nitrogen (PIN) as a marker.**

Variety	Dry matter			Crude protein		
	TCM	PIN	Adj. PIN <sup>a</sup>	TCM	PIN	Adj. PIN <sup>a</sup>
	%					
WW-506 <sup>b</sup>	61.1	46.2	57.7	70.2	59.1	67.9
WW-573	60.7	51.7	62.0	69.6	62.6	70.6
WW-477 <sup>b</sup>	61.4	48.6	59.6	71.4	61.9	70.0
WW-517 <sup>b</sup>	59.0	52.1	62.3	68.7	63.5	71.3

<sup>a</sup>Values adjusted on basis of average recovery of pepsin insoluble nitrogen.

<sup>b</sup>Within nutrient components, differences between means for TCM and Adj. PIN statistically significant ( $P < .05$ ).