

# Evaluation of Different Exogenous Enzymes to Improve Fiber Digestibility of Soybean Hulls

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## Introduction

Reducing feed cost is the primary driver for utilizing exogenous fibrolytic enzymes in livestock production. Feed accounts for 60 to 70% of total costs in most livestock enterprises, with energy as the major constituent of feed. Differences in digestibility explain most of the variation in energy content of a feed. In addition to starch, digestible fiber can be a major contributor to the energy content of the diet. For the past 30 years, feed enzymes have played an important role in helping to improve the efficiency of production by increasing digestibility of major nutrients in feeds. Creep feeding is a common management practice of providing supplemental feed to calves before weaning. Weaning weight is a big profit-driver of any cow/calf enterprise. Creep feeding can be a profitable practice if the expected increase in weaning weight out-weighs the cost of feeding creep. One of the factors to consider in creep feeding is feed prices and the use of co-products will help lower the cost of feed. One such co-product that has been used over the years is soybeans hulls. Due to limitations of calves to effectively utilize the fiber portion of their diets, supplementing with specific exogenous enzymes improves the nutritional value of such diets by increasing fiber digestion. The objective of this study was to identify enzyme additives that consistently increase fiber digestibility of soybean hulls used in creep rations by using the in vitro gas and in situ evaluation techniques.

## Materials and methods

### *Study 1. Initial evaluation study using the in vitro batch culture*

The batch culture technique was used to evaluate and identify enzyme additives that increase fiber digestibility of soybean hulls. The enzymes were obtained from industry collaborators and key enzyme activities of cellulase, xylanase and mannanase were considered to choose enzyme products with potentials for high activity in the rumen. Ten enzyme treatments (NSPase, ABM, DYX, AMA, CUL, Mix1, Mix2, Mix3, Mix4 and Mix5) were evaluated using the in vitro gas incubation technique. Approximately 0.5 g of creep feed (Table 1) was weighed into filter bags (Ankom Technology, Macedon, NY, USA) and sealed. Sealed bags were placed in serum bottles. Ruminal fluid was collected 2 h after feeding from three ruminally-fistulated steers (1521 lbs body weight) fed a high forage diet. All animal procedures were approved by the NDSU Animal Care and Use Committee. Whole ruminal contents were obtained from the rumen, composited, and immediately transferred to the laboratory and held at 39°C in a water bath. Each serum bottle received 45 mL of McDougall's buffer (artificial saliva) and 15 mL of strained ruminal fluid. Bottles were flushed with CO<sub>2</sub>, capped, and crimp sealed. Sealed bottles were incubated in an oscillating shaker at 39°C for 6, 12, and 24 h. After incubation, bags were removed from bottles, washed and dried in an oven for 48 h. Dry matter disappearance was determined by subtracting the loss of DM from the bags from the initial DM incubated.

**Table 1. Ingredients and chemical composition of diet.**

Ingredients	Composition (% DM)
Rolled corn	20.5
Modified DGS	20.0
Soybean hulls	55.0
Mineral supplement	1.85
Vitamin supplement	2.60

Chemical composition <sup>1</sup>	Composition (% DM)
Dry matter, %	89.3
Crude protein	18.1
Ether extract	2.14
eNDF	1.10
NEg, Mcal/lb	65.1
Calcium	0.79
Phosphorous	1.08
Potassium	1.07

<sup>1</sup>Values for the experimental diets were calculated from NRC (1996) feed library table based on the ingredient composition.

### *Study 2. In situ ruminal digestibility of DM and fiber.*

The best two enzymes (NSPase and ABM) were further evaluated using the in situ technique. The in situ study was conducted with the same steers from the in vitro study above and fed the same high forage diet. Ruminal DM and NDF degradability was determined using polyester bags. Quadruplicate samples of each enzyme treatment were incubated in the rumen of three steers. About 5 g of the same diet used in the in vitro study was incubated for 6, 12, and 24 h. Immediately after removal from the rumen, bags were immersed in ice-water to stop or minimize microbial activity and then washed with cold water in a washing machine for 35 min. Ruminal DM and NDF disappearance was calculated by subtracting the loss of DM and NDF from the bags from the initial DM and NDF incubated.

## **Results**

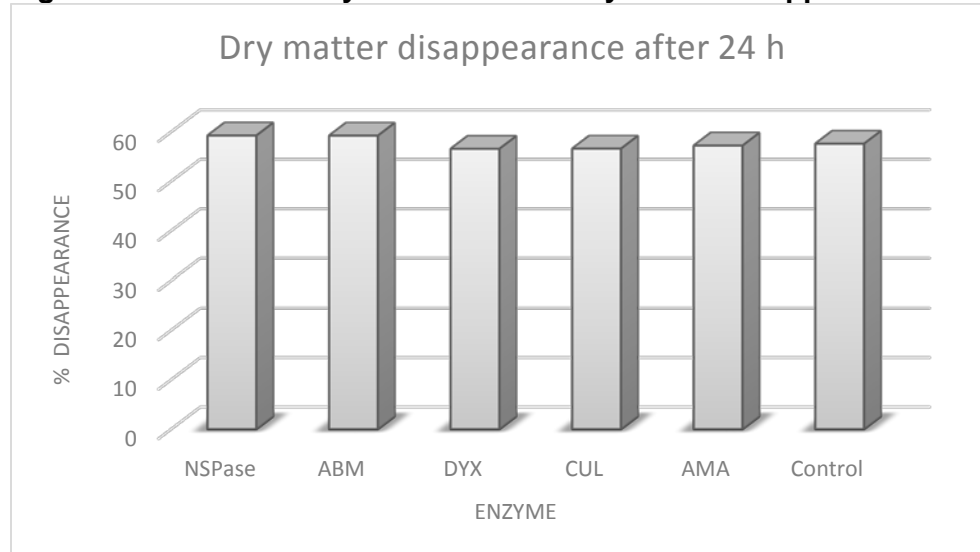
The in vitro batch culture technique is used to determine the nutritive value of different diets. Gas is produced when carbohydrates in the diets are fermented to short-chain fatty acids (SCFA) like acetate and butyrate. One major advantage of the gas measurement technique is that it focuses on the appearance of fermentation products as soluble but non-fermentable substrates do not contribute to gas production. Gas production reflects the generation of SCFA and microbial mass. Higher ( $P<0.05$ ) asymptotic gas volume was noted for NSPase, ABM and DYX treatments compared with the other enzymes (Table 2). Enzyme treatments did not result in higher DM disappearance but NSPase and ABM treatments had numerically higher (3% more) DM disappearance values compared with the control (Figure 1). The implication is that these enzymes will result in improved DM and fiber digestibility.

**Table 2. Effects of exogenous enzymes on in vitro gas production kinetics of creep diet.**

Enzymes	M	c (h <sup>-1</sup> )	Lag time (h)
NSPase	200	7.85	2.69
ABM	199	7.08	1.50
DYX	194	6.95	1.33
AMA	151	6.85	0.10
CUL	162	7.57	1.36
Mix1	158	7.96	2.22
Mix2	146	8.21	2.52
Mix3	133	9.58	2.97
Mix4	148	10.2	3.40
Mix5	140	10.7	3.36
Control	139	8.49	2.67
SEM	22.3	0.769	0.537
LSD at <i>P</i> < 0.05	21.2	3.9	3.3

M = asymptotic gas volume (ml/g DM); c = rate of fermentation.

**Figure 1: Effects of enzymes on in vitro dry matter disappearance after 24 h of fermentation.**



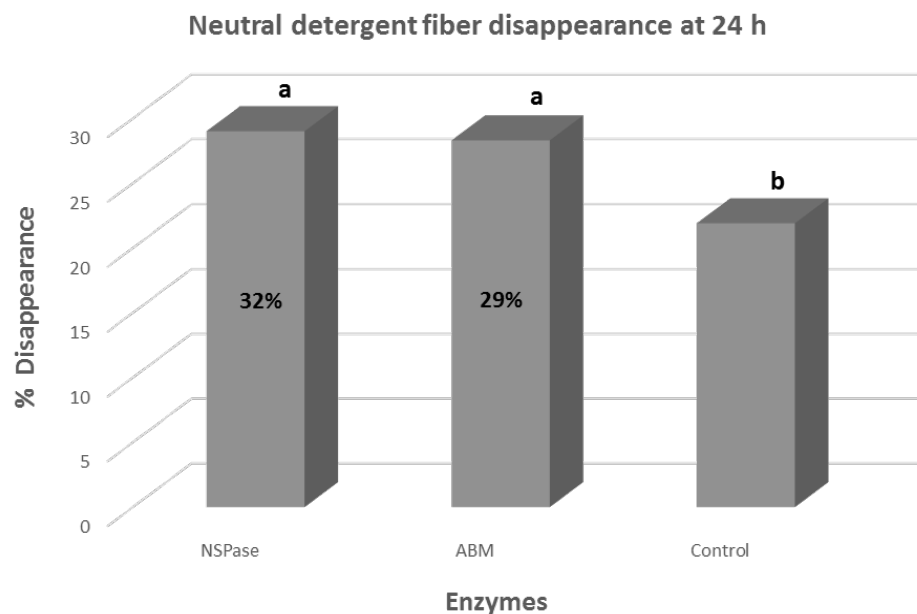
Compared with the control (no enzyme), both NSPase and ABM resulted in higher effective DM digestibility at low, medium and high feeding levels (Table 3). Differences in DM digestibility as a result of enzyme inclusion ranged from 4 to 9%. Additionally, enzyme treatments resulted in 32 and 29% increase in NDF digestibility for NSPase and ABM, respectively (Figure 2). Overall, these two (in vitro and in situ) techniques were utilized to identify an exogenous enzyme that can potentially increase DM and fiber digestibility of soybean hulls used in a creep feed.

**Table 3. Non-linear estimates and effective degradability coefficient of the creep diet.**

	ABM	NSPase	Control	SEM	<i>P</i> value
A	0.241 <sup>b</sup>	0.244 <sup>b</sup>	0.243 <sup>ab</sup>	0.0005	0.036
B	0.406 <sup>b</sup>	0.433 <sup>a</sup>	0.399 <sup>b</sup>	0.0051	0.003
k	0.132 <sup>a</sup>	0.116 <sup>b</sup>	0.097 <sup>c</sup>	0.0036	<0.001
Lag	0	0	0.157	0.0564	0.130
ED (2%)	0.594 <sup>b</sup>	0.614 <sup>a</sup>	0.573 <sup>c</sup>	0.0031	<0.001
ED (5%)	0.536 <sup>b</sup>	0.547 <sup>a</sup>	0.505 <sup>c</sup>	0.0018	<0.001
ED (8%)	0.494 <sup>b</sup>	0.500 <sup>a</sup>	0.459 <sup>c</sup>	0.0015	<0.001
Undegraded	0.352 <sup>a</sup>	0.323 <sup>b</sup>	0.358 <sup>a</sup>	0.0054	0.003

<sup>abc</sup> Means in the same column with different superscripts differ ( $P < 0.05$ ).

A = The portion of DM solubilized at initiation of incubation; B = the fraction of DM insoluble but degradable in the rumen; k = the constant rate (percentage per hour) of disappearance of fraction B; Lag = lag phase, (hours) prior to the commencement of degradation of fraction B; ED = Effective degradability at three rumen solid outflow rates of 2, 5 and 8% per h, which is representative for low, medium and high feeding levels.

**Figure 2: Effects of enzymes on in situ ruminal neutral detergent fiber digestibility.**

Bars with different letters differ ( $P < 0.05$ ).