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*Published in:*

77th Cornell Nutrition Conference for Feed Manufacturers

*Publication date:*

2015

*Citation for published version (APA):*

Zontini, A. M., Foskolos, A., Ross, D. A., J., M., P.H., D., & Van Amburgh, M. E. (2015). Formulating diets for lactating cattle using multiple pools of NDF digestibility. In 77th Cornell Nutrition Conference for Feed Manufacturers Syracuse, NY.

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# RESEARCH UPDATE: FORMULATING DIETS FOR LACTATING CATTLE USING MULTIPLE POOLS OF NDF DIGESTIBILITY

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## INTRODUCTION

Fiber, compared to the other nutritional components, is slowly digestible and bulky, thus, creating a ballast into the digestive tract of ruminants. For this reason it is thought to be a primary regulator of feed intake in ruminants (Mertens and Ely, 1979). Furthermore, the fiber component (aNDFom) of the diets fed to cattle is not uniformly digestible. For instance, aNDFom contains a fraction that is not available to microbial digestion even if fiber fermentation could be extended for infinite time (Allen and Mertens, 1988, Huhtanen et al., 2006). This undigestible fraction is analyzed in laboratories with long term in-vitro fermentation and defined uNDF (Cotanch et al., 2014)). Furthermore the component available to microbial digestion is defined digestible NDF (dNDF) and can be obtained by subtracting the uNDF from total aNDFom ( $dNDF = aNDFom - uNDF$ ). Previous work demonstrated that the dNDF of forages can be composed of two digestible fractions (Van Soest et al., 2005), both fractions following first order behavior but with different digestion rates and defined as fast and slow digesting pools (Raffrenato and Van Amburgh, 2010); whereas in plant by-products the dNDF is identifiable as one fraction and disappearing with a first order behavior (Cotanch et al., 2014). The size of the various fractions or pools of dNDF and the associated digestion rates, when combined to create differences with a total mixed ration, might affect feed intake and rumination behavior, milk production and feed efficiency. The objective of this study was to balance diets for high producing dairy cattle using these concepts, and evaluate the effect that diets with different proportions of the aNDFom pools might have on feeding behavior, rumination, milk production and feed efficiency. Based on the previous data (Cotanch et al., 2014) our hypothesis was that cattle fed diets varying in uNDF content and related NDF pools would demonstrate differences in feed intake, rumination and feed efficiency.

## MATERIALS AND METHODS

### Animals, Treatments and Experimental Design

One-hundred and eight multiparous cows ( $1,602 \pm 146$  lbs BW;  $127 \pm 54$  DIM) and thirty-six primiparous cows ( $1,356 \pm 117$  lbs BW;  $106 \pm 46$  DIM) were enrolled in the study and housed at the Cornell University Research Center (CURC). Animals were stratified by DIM, BW, and milk production and distributed into 9 pens of 16 (12 multiparous and 4 primiparous) cattle. Cows had free access to water and were fed total mixed ration once a day at 0930 h, allowing 5% refusals. Feed push up occurred 3 times a day (after

milking). Cows received rBST (Posilac, Elanco Animal Health, Indianapolis, IN) every 14 days according to the farm management protocol.

Treatment diets were developed considering the fractionation of aNDFom that was determined by in-vitro digestibility into fast and slow digestible pools, and uNDF. Diet ingredients were analyzed as described in Raffrenato (2011) and Cotanch et al. (2014) to measure the amount of aNDFom by pools from each ingredient (Table 1).

Table 1. Nutrient composition of ingredients used in diets with the aNDFom fractionation.

Feed	DM,%	CP, %DM	aNDFom, %DM	uNDF, %aNDFom	Fast, %aNDFom	Slow, %aNDFom
Alfalfa silo B	89	17.6	38.8	51.3	35.7	13
Alfalfa silo 4	35	22.4	38.1	36.2	55.2	8.7
Alfalfa silo 5	41.2	21	40.9	42.8	33.4	23.9
BMR corn silage	26.1	7.6	43.8	23.7	67.8	8.5
Conv. corn silage	25.2	7.3	41.3	30.3	8.8	60.9
Citrus pulp dry	89.8	6.3	23.1	12	92	na
Corn gluten feed	88.7	19.7	29.8	12	88	na
Corn grain	88	8	11.1	2	98	na
Delinted whole cottonseed	92	23.5	50.3	43	57	na
Soybean hulls	91	10.3	65.9	3	97	na
Soy Plus	90.8	47.6	18.9	2	98	na
Sunflower seed hulls	92	6.3	75.4	74	26	na
Wheat midds	91.6	16.4	37.4	22	78	na

Three diets were developed, two diets with approximately 86% aNDFom from forages and the remaining diet with 34% aNDFom from forages. The formulated high forage diets were 32% uNDF (High uNDF, HUF) and 26% uNDF (low uNDF, LUF) and the low forage diet was formulated to match the uNDF of the High uNDF high forage diet (32% uNDF, HUNF) (Figure 1). Pens were randomly assigned to the three treatments (Table 2) in a 3x3 Latin square design with 21-d adaptation periods and 5-d sampling periods.

Diets were formulated to allow 95 lb of ME and MP allowable milk production at approximately 58 lb DMI using the Cornell Net Carbohydrate and Protein System (CNCPS v6.5 via AMTS.Cattle.Pro v4.0) using library values adjusted with available chemical composition of the actual ingredients. The experiment took place between April 8 and July 21, 2015.

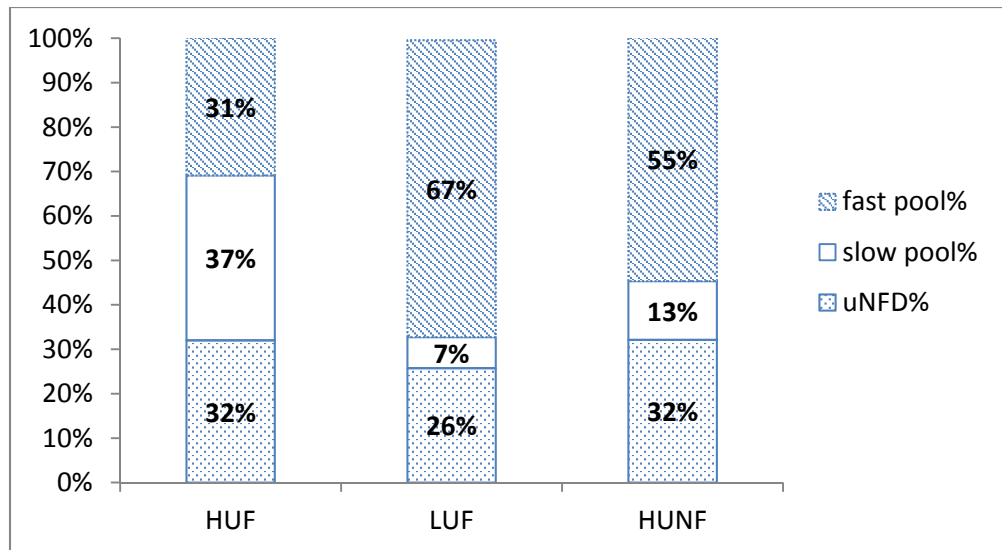


Figure 1. The formulated proportion of the fast and slow digestible pool, and uNDF in the three treatment diets. The diets were high forage, high uNDF (HUF) (~70% forage, 32% uNDF), high forage, low uNDF (LUF) (~70% forage, 26% uNDF) and low forage, high uNDF (HUNF) (30% forage, 32% uNDF).

### Sampling Procedure and Analysis

Representative samples of feed ingredients, TMRs and individual pen orts were taken and analyzed in triplicate for dry matter, chemical composition and NDF digestibility. Dry matter was determined by putting samples in a forced air oven at 55°C for 48 h. Feed samples were then ground using a Wiley mill (Arthur H. Thomas, Philadelphia, PA) with a 1-mm screen and collected in composites on a period basis for determination of nutrient content. Milk yield and sampling for composition, DMI and minutes of rumination using SCR monitors (SCR Global, Netanya, Israel) were measured during the first 3 days of the 5-d sampling periods were recorded. Furthermore, feces were collected on 72 randomly selected cows (8 per pen) at 0530, 1330, and 2130 on day 4, and at 0930, 1730, and 0130 on day 5 of the sampling period. At any fecal sampling 500ml of sample was obtained from each cow and frozen immediately at -20°C until further analysis. Fecal samples were then composited per cow, dried and ground as before, and analyzed for aNDFom digestibility at 30, 96, 120, and 240 h as time points. Samples were sent for analysis to Cumberland Valley Analytical Services Inc. (Maugansville, MD).

### Milk Production and Components

Cows were milked three times daily at 0830, 1630, and 2430 h and milk production from all milking's was recorded using Alpro herd management software (DeLaval International AB, SG). Milk samples were collected at each milking in the first 3 days of any 5-d sampling period, and preserved with 2-bromo-2-nitropane-1, 3-diol at 4°C until analyzed. Milk samples were analyzed by infrared methods (DairyOne Ithaca, NY) for fat, true protein, lactose, total solids, MUN (Foss Milkoscan FT+, Foss In., Eden Prairie, MN) and somatic cell count (SCC) (Fossomatic FC, Foss Inc., Eden Prairie, MN).

Table 2. The ingredient content and chemical composition of experimental diets

Ingredient, % DM	Diet		
	HUF	LUF	HUNF
Alfalfa hay	7.7	-	-
Alfalfa haylage (hi-uNDF)	9.9	-	-
Alfalfa haylage (lo-uNDF)	-	18.5	7.3
BMR corn silage	-	23.4	-
Conv. corn silage	24.3	-	9.5
Citrus pulp	2.4	1.5	5.1
Corn gluten feed	-	-	2.4
Corn grain ground	8.8	10.1	14.6
Cottonseed delinted	-	-	4.4
Soybean hulls	1.1	-	3.3
Soy Plus	4.0	5.1	3.5
Sunflower seed hulls	-	-	4.0
Wheat middlings	-	-	4.6
Minerals, vitamins	2.9	2.0	2.2
<i><u>Chemical Composition</u></i>			
DM	46	41	63
CP, % DM	16.1	16	16.1
aNDFom, %DM	32.9	33.1	33.1
Fast pool, %aNDFom	31	67	55
Slow pool, %aNDFom	37	8	13
uNDF, % aNDFom	32	25.7	31.2
Starch, %DM	24.4	24.7	25.1
Sugar, %DM	6.0	4.3	6.1
EE, % DM	4.0	4.2	4.4
Ca, %DM	0.86	0.96	0.63
P, %DM	0.36	0.41	0.38
ME, mcal/lb DM	1.13	1.13	1.11
ME allowable milk (lb/d)	96.56	96.78	98.77
MP allowable milk (lb/d)	103.40	105.38	106.26
Lys:Met	2.94	2.94	2.83

### Statistical Analysis

Data were analyzed using the following mixed effects model (JMPv.11 SAS Institute, Inc., Cary, NC):

$$Y_{ijkl} = T_i + PR_j + D_k + PN_l + PN(PR)_{l(j)} + B_m + \epsilon_{ijklm}$$

where,

- $Y_{ijkl}$  is the dependent, continuous variable,
- $T_i$  is the fixed effect of the  $i$ th treatment ( $i=1, 2, 3$ );
- $PR_j$  is the fixed effect of the  $j$ th period ( $j=1, 2, 3$ );
- $D_k$  is the fixed effect of the  $k$ th day ( $j=1, 2, 3$ );
- $PN_l$  is the random effect of the  $l$ th pen ( $j=1, \dots, 9$ );

$PN(PR)_{l(j)}$  is the random effect of the  $j$ th ( $j=1, 2, 3$ ) period nested within  $l$ th ( $l=1, \dots, 9$ ) pen,  
 $B_m$  is the covariate measurement for the  $m$ th pen ( $l=1, \dots, 9$ );  
 $\epsilon_{ijklm}$  is the residual error.

Overall treatment effects were analyzed using Tukey's test. Significance was declared at  $P$ -values  $<0.05$ .

## RESULTS AND DISCUSSION

### Diet formulation

Overall, milk yield of the cows on all treatments was high and cow health, aside from some mastitis was excellent. Characteristics of the feed ingredients allowed for the development of three diets similar in chemical composition, in particular the same aNDFom content but quite different aNDFom fractionation (Table 2). The primary differences between the high forage diets (HUF and LUF) was the difference obtained by using conventional corn silage and a blend of two low digestibility alfalfa silages (B and 5) for treatment HUF; and a BMR corn silage and high digestible alfalfa (4) for LUF (Table 1) diets, respectively. This allowed for the formulation of two diets that varied in the amount of aNDFom in the fast pool by 2x, but modest differences in the amount of uNDF (Figure 1). For the low forage, high byproduct diet, (HUNF), the objective was to develop a diet that contained the same uNDF pool size as the HUF diet to determine if the uNDF pool would provide regulation of DMI independent of the source of aNDFom, uNDF and to some degree particle size. For the HUNF treatment, sunflower seed hulls played a key role for the high aNDFom and uNDF content along with wheat middlings, soybean hulls and delinted cottonseed were also chosen for the high fiber content. Cattle fed the low forage, high by-product diet consumed approximately 9 lb more dry matter and ruminated about 1 hour less per day than cattle fed the two high forage diets. Cattle fed the two high forage diets (HUF and LUF) did not demonstrate any differences in DMI or chewing and rumination (Table 3). The formulated uNDF content of the LUF diet was approximately 6% less than the HUF diet and at the time of writing, the feed chemistry was not complete to verify that the formulated difference was maintained throughout the experiment, thus drawing conclusions about the lack of difference in DMI is not yet possible. The lack of difference in rumination time among the high forage diets was interesting since it was assumed that with the significant difference in the size of the fast pool in the LUF diet, the rate of aNDFom disappearance would have been faster and would have negatively influenced chewing and rumination. This lack of difference might provide some insight into a different model of rumen function and how digestibility impacts buoyancy and specific gravity to maintain a rumen mat that encourages regurgitation and rumination in addition to the long-held observation of particle size (Mertens, 1997). Finally BW change tended to increase with increasing DMI. This suggests that if BW change was accounted for and included as part of the energy allowable production, the low forage (HUNF) would look favorable to overall energy balance despite the reduced feed efficiency calculated only when looking at energy corrected milk (Table 4).

Table 3. Dry matter intake, rumination and body weight of cattle fed diets that varied in uNDF and NDF pools as determined by in-vitro digestibility measured at 30, 120 and 240 hr as formulated.

Item <sup>1</sup>	HUF	LUF	HUNF	SEM	P-value
DMI (lb/cow/d)	61.1 <sup>a</sup>	61.3 <sup>a</sup>	77.8 <sup>b</sup>	1.01	0.06
aNDFom intake, lb/d	20.1 <sup>a</sup>	20.2 <sup>a</sup>	23.4 <sup>b</sup>	3.8	0.96
uNDF intake, lb/d	6.4	5.2	7.4	0.09	0.0001
dNDF intake, lb/d	13.7	15.0	16.0	0.19	0.0001
Fast intake, lb/d	6.3	13.4	12.8	0.15	0.0001
Slow intake, lb/d	7.4	1.6	3.2	0.07	0.0001
aNDFom intake, %BW	1.30 <sup>a</sup>	1.30 <sup>a</sup>	1.48 <sup>b</sup>	0.02	0.99
uNDF intake, %BW	0.42	0.34	0.48	<0.01	0.0001
dNDF intake, %BW	0.89	0.98	1.01	0.01	0.22
<u>Rumination</u>					
Rumination (min/cow/d)	593 <sup>a</sup>	609 <sup>a</sup>	534 <sup>b</sup>	7.25	0.28
Rumination (min/lb NDF intake)	29.5 <sup>a</sup>	30.1 <sup>a</sup>	22.9 <sup>b</sup>	0.41	0.36
Rumination (min/lb dNDF intake)	43.3	40.7	33.2	0.58	0.0001
<u>BW and BCS</u>					
BW initial, lb	1,547	1,554	1,574	6.59	0.78
BW change, lb per treatment period	7.1	20.5	39.2	9.17	0.08
BCS change	0.01	0.01	0.1	0.05	0.63

<sup>1</sup>Values in columns with different superscripts differ P < 0.05.

The formulation of these diets created a problem with the terms that we have been using to describe the digestibility of the forages and that is “fast” and “slow”. This characterization is comparative and not discreet and the differences became apparent because as reported in Cotanch et al (2014), most of the high NDF byproducts have only one digestible aNDFom fraction and generally the rate of digestion of that pool is intermediate between the fast and slow pool of forages. Thus, when characterizing how digestion takes place and how to assign the fractions to pools, the absolute rates and the pool sizes might become important as we learn to apply these concepts.

Animal performance

Milk yield and ECM were different among treatments, although milk yield among all treatments was high, especially for the HUF and LUF diets that were approximately 70% forage. This demonstrates that although we were altering digestibility among NDF pools, the overall digestibility of the forages was good. That in itself is problematic when trying to design studies of this nature because treatment differences can only be as significant as your differences in forage digestibility for the year and season the work is being conducted in.

When the cattle were fed the LUF diet, which had the largest fast pool of aNDFom compared to the HUF diet, the cattle produced approximately 3 lb more ECM (Table 4). The inverse linear relationships among milk fat and protein as pools shifted is interesting and suggests differences in substrate supplies with very similar DMI and forage contents.

Table 4. Effects of the treatments on milk yield, milk composition, and gross feed efficiency.

Item <sup>1</sup>	Diet			SEM	P-value
	HUF	LUF	HUNF		
DMI (lb/cow/d)	61.1 <sup>a</sup>	61.3 <sup>a</sup>	77.8 <sup>b</sup>	1.01	0.056
Milk production					
Milk, (lb/d)	91.7	96.3	105.5	1.06	<0.0001
Energy corrected milk, lb/d)	94.1	96.8	100.1	1.15	<0.0001
Fat yield, (lb/d)	3.48 <sup>a</sup>	3.46 <sup>a</sup>	3.31 <sup>b</sup>	0.05	0.76
True protein yield, (lb/d)	2.67	2.84	3.20	0.05	<0.0001
Lactose yield, (lb/d)	4.36	4.67	5.09	0.067	<0.0001
Gross feed efficiency (ECM/DMI)	1.55 <sup>a</sup>	1.58 <sup>a</sup>	1.41 <sup>b</sup>	0.02	0.52
Milk composition					
Fat, (%)	3.79	3.58	3.18	0.04	<0.0001
True protein, (%)	2.91	2.95	3.05	0.01	0.04
Lactose, (%)	4.77	4.84	4.89	0.01	<0.0001
MUN mg/dl	11.7 <sup>a</sup>	8.8 <sup>b</sup>	10.4 <sup>a</sup>	0.12	0.04
SCC (log1000/ml)	119.5	113.6	147.6	22.58	0.34

<sup>1</sup>Values in columns with different superscripts differ P < 0.05.

When cattle were fed the diet with the largest proportion of aNDFom in slow pool (HUF) they had the highest concentration of fat in the milk, again suggesting differences in substrate supply for milk component yield. Finally, as the size of the fast aNDFom pool increased (LUF), the MUN decreased suggesting more microbial yield and lower rumen ammonia balances compared to the other two treatments due to increased microbial activity, and this needs to be further evaluated.

#### ACKNOWLEDGEMENTS

Special thanks to ADM Alliance Nutrition (Quincy, IL), ADM Oilseed Processing (Enderlin, ND) for supplying the sunflower seed hulls and to Sam Fessenden, Katie Andrews and Rodrigo Molano for support and help in conduct of the experiment and to the staff at CURC, especially Dr. Bill Prokop and Lisa Furman. Thanks also to Rick Grant and Kurt Cotanch for helpful discussion.

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