

# Development of highly digestible animal feed from lignocellulosic biomass Part 2: Oxidative lime pretreatment (OLP) and shock treatment of corn stover<sup>1</sup>

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**ABSTRACT:** Oxidative lime pretreatment (OLP) increases lignocellulose digestibility by removing lignin and hemicellulose acetyl content. Digestibility is improved further by adding mechanical shock treatment, which subjects aqueous slurry of biomass to an explosive pressure pulse. Shock treatment mechanically disrupts the microscopic structure while maintaining the macroscopic integrity of the biomass particle. This study determined the effectiveness of these pretreatments to enhance the ruminant digestibility of corn stover. In terms of compositional changes, OLP and shock treatment should negatively affect the feed value of corn stover; however, digest-

ibility analysis provides a significantly different conclusion. With corn stover, shock + OLP improved the 48-h neutral detergent fiber digestibility (NDFD) to 79.0 g neutral detergent fiber (NDF) digested/100 g NDF fed, compared to 49.3 for raw corn stover. The 48-h in vitro total digestible nutrients (TDNom, g nutrients digested/100 g OM) was 51.9 (raw), 59.7 (OLP), and 72.6 (shock + OLP). Adding extracted corn stover solubles to shock + OLP increased TDNom to 74.9. When enough solubilized chicken feathers were added to match the protein content of corn grain, TDNom increases to 75.5, which is only 12.6 less than corn grain.

**Key words:** lignocellulose, lime pretreatment, ruminant animal feed, shock pretreatment

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## INTRODUCTION

Corn grain has been widely used as livestock feed. For ruminants, it is possible to displace corn with lignocellulose, the most abundant organic material on earth (Rajaratnam et al., 1989).

Falls et al. (2017) describes a combination of oxidative lime pretreatment (OLP) and ball milling to generate highly digestible forage sorghum to supplement or replace corn grain as ruminant animal feed. Even though rumen digestibilities of this pretreated forage sorghum are high based on laboratory assays, ball-milled material has no practical use as feed because it is extremely expensive. Furthermore, there is the pos-

sibility that fine particles produced by ball milling can readily escape from the rumen before they are digested.

Even though the study reported by Falls et al. (2017) did not result in a practical treatment, the following valuable lessons were obtained: (1) use feedstock with low protein content, which prevents its loss in OLP; (2) extensively wash the biomass to remove ash from OLP; and (3) select a mechanical pretreatment that maintains fiber integrity so it is retained in the rumen until digested.

The purpose of this work is to generate highly digestible corn stover to supplement or replace corn grain as ruminant animal feed. Compared to forage sorghum, corn stover has lower protein content (Lesson 1). Furthermore, prior to OLP, the corn stover is extracted with water to remove soluble protein and other solubles (e.g., free sugars, hemicellulose). After OLP, it is extensively washed to reduce the ash content of the feed (Lesson 2). In contrast to ball milling, which finely divides the biomass, shock treatment mechanically disrupts the microscopic structure while maintaining the macroscopic integrity of the biomass particle (Lesson 3). To render the biomass

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more digestible, OLP was combined with mechanical shock treatment. To determine the nutritive value of the generated feed, composition and in vitro digestibility were determined by university and commercial laboratories.

## MATERIALS AND METHODS

### Biomass Feedstocks

Corn stover was provided by the National Renewable Energy Laboratory and was dried to uniform moisture content (< 10%) and milled to pass 0.6-cm round screen. To wash extractives out of the corn stover, deionized (DI) H<sub>2</sub>O was used at a ratio of 10 mL DI H<sub>2</sub>O per mL corn stover. Corn stover and DI H<sub>2</sub>O were mixed on a roller bottle apparatus for 1 h before centrifugation; the solids were subsequently dried. The supernatant was concentrated using rotary evaporation, and then freeze-dried to a powder using a Labconco Lyph-Lock 6-L freeze dryer system (Model 77530, Labconco Corporation, Kansas City, MO). Cumberland Valley Analytical Services, Inc. (Hagerstown, MD) performed the compositional analysis for corn stover samples (Table 1); the analytical methods are reported by Falls et al. (2017). Two materials were used as con-

trols: cracked corn grain and alfalfa. Cumberland Valley Analytical Services, Inc. performed compositional analysis on these control materials as well (Table 1).

### Short-term Oxidative Lime Pretreatment

Corn stover was pretreated using short-term OLP as described by Falls et al. (2011). The pretreatment vessel was a 20-L stainless steel batch reactor [Fig. 1(a)]. Corn stover (500 g), excess calcium hydroxide (250 g), and distilled water (7.5 L) were loaded into the reactor. The reactor was sealed, heated to 110°C, and the stirring mechanism was activated. The reactor was then charged with  $6.89 \times 10^5$  Pa pure oxygen, and the reaction proceeded for 3 h. When complete, the heat and stirring were shut off, and the reactor was allowed to cool. Once the reactor was cool enough to handle, it was slowly vented to relieve pressure, and then opened. The pretreated slurry was removed and neutralized to pH 4.0 using 5-N HCl. The slurry was then vacuum filtered to isolate the pretreated solids. To wash out any residual lime, the pretreated solids were washed with distilled water a minimum of 3 times, until the pH of the collected wash was equal to that of fresh distilled water. The pretreated corn stover was air dried in metal pans. To prevent microbial

**Table 1.** Compositional analysis, neutral detergent fiber digestibility, and total digestible nutrients of corn grain, alfalfa, corn stover samples, solubilized protein, and balanced feeds

Sample	Corn stover						Solubilized protein	Combined feed <sup>3</sup>	Protein-balanced feed <sup>4</sup>	Corn grain	Alfalfa
	Raw	Shock treated	OLP	OLP <sup>2</sup> + shock	Shock + OLP	Solubles					
Moisture, %	9.5	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	14.2	7.4
Ash, %DM	7.9	6.6	8.7	10.3	8.3	31.4	7.0	12.4	12.2	1.3	9.4
CP, %OM <sup>1</sup>	7.1	6.6	3.2	4.1	3.1	28.0	95.9	6.5	9.7	8.6	16.6
ADF, %OM	48.3	59.9	72.6	77.2	75.4	1.0	0.4	65.0	62.7	4.8	39.3
NDF, %OM	77.3	88.1	81.9	87.6	87.1	1.7	1.0	75.2	72.6	11.4	49.2
NFC <sup>5</sup> , %OM	16.8	6.9	16.0	10.4	10.4	69.7	2.9	18.6	18.1	76.6	34.1
Lignin, %OM	10.4	13.5	13.4	9.1	7.2	0.3	0.2	6.2	6.0	2.4	9.6
Fat, %OM	1.0	0.4	0.7	0.6	0.8	1.2	0.6	0.8	0.8	4.0	2.1
Ca, %DM	0.4	0.4	2.9	1.4	1.3	1.1	3.3	1.2	1.3	0.0	1.6
P, %DM	0.1	0.1	0.0	0.0	0.0	0.6	0.0	0.1	0.1	0.3	0.2
Mg, %DM	0.2	0.1	0.1	0.0	0.0	1.0	0.0	0.2	0.2	0.1	0.7
K, %DM	1.8	0.6	0.0	0.1	0.1	11.7	0.3	2.1	2.1	0.4	2.3
Na, %DM	0.0	0.2	0.0	0.2	0.0	0.1	1.0	0.0	0.1	0.0	0.1
NDFD <sup>6</sup> , %NDF	49.3	43.9	79.0	79.3	76.0	n/a	n/a	n/a	n/a	63.2	47.9
TDN <sub>W</sub> , %DM	48.1	40.9	42.7	45.2	49.7	61.0	86.1	51.7	52.8	86.0	55.7
TDN <sub>OM</sub> , %OM	52.2	43.8	46.7	50.3	54.2	88.9	92.6	59.0	60.2	87.1	61.5
TDN <sub>N</sub> , %DM	47.8	37.5	54.5	62.5	66.6	n/a	87.5	65.6	66.3	87.0	53.9
TDN <sub>OM</sub> , %OM	51.9	40.2	59.7	69.7	72.6	n/a	94.1	74.9	75.5	88.1	59.4

<sup>1</sup>OM = NDF + NFC + Fat + CP.

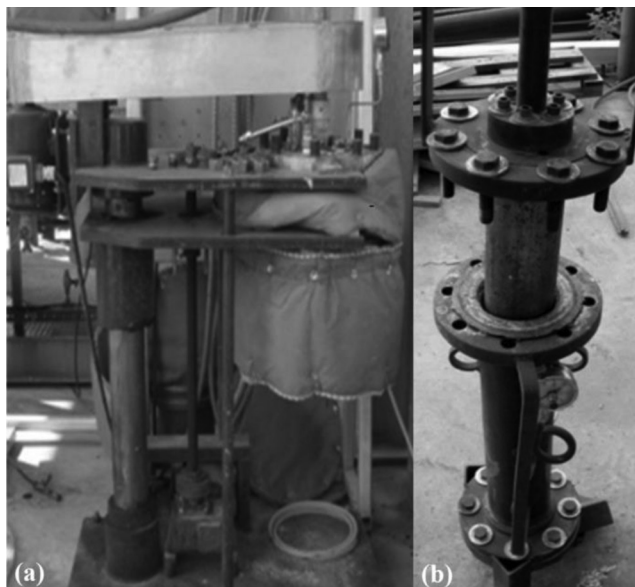
<sup>2</sup>OLP = oxidative lime pretreated.

<sup>3</sup>Combined feed = 17.8% corn stover solubles and 82.2% shock + OLP corn stover.

<sup>4</sup>Protein-balanced feed = 3.3% solubilized protein, 17.2% corn stover solubles, and 79.5% shock + OLP corn stover.

<sup>5</sup>NFC = non-fiber carbohydrates.

<sup>6</sup>NDFD = neutral detergent fiber digestibility (48 h). NDFD was measured by Texas A&M University Animal Science Department.



**Figure 1.** (a) Oxidative lime pretreatment 20-L reactor. (b) Shock tube reactor.

growth while drying, the biomass was stirred at least once every 24 h. A portion of the lime-pretreated corn stover was subjected to shock treatment.

### **Shock Treatment**

Shock treatment was performed in the shock tube pretreatment apparatus [Fig. 1(b)]. The shock tube is comprised of a carbon steel tube and carbon steel barrel connected by a 300-lb-class flange. The bottom tube is a 50.8-cm section of 10.2-cm Schedule (Sch.) 80 pipe, and the top barrel is a 70.0-cm section of 2.5-cm Sch. 40 pipe. The 2.5-cm barrel joined the 10.2-cm pipe through a 27.9-cm-long conical section. The conical section has an inner diameter of 2.2 cm at the barrel end, which increases to 9.0 cm at the tube end. The shock tube was placed in a temperature-controlled water bath (25°C), and loaded with 100 g dry corn stover and 2 L water. The barrel section was lowered onto the bottom tube, and the shock tube was sealed. A 12-gauge shotgun shell (Winchester Expert High Velocity 8.9 cm, 0.04-kg steel BB shot) was placed inside the top opening of the barrel and fired by releasing a steel plate firing pin onto the central metal surface of the shell. The flange was unbolted, and the barrel section of the shock tube was lifted away. The shock tube contents were placed in a product container and then filtered to remove lead shot and other shell remnants. The shocked corn stover was then air dried in metal pans to uniform moisture content (< 10%).

### **Solubilized Protein from Chicken Feathers**

Chicken feathers (provided by Texas A&M Poultry Science Department, College Station) were

washed, air-dried, and then completely dried at 105°C. The dried feathers were ground using a Thomas-Wiley laboratory mill (Arthur H. Thomas Company, Philadelphia, PA) and sieved through a 2-mm screen. Lime treatment was performed in a 1-L autoclave reactor with a temperature controller and mixer (1,000 rpm). Recommended treatment conditions were used: 100°C, 300 min, and 0.1 g Ca(OH)<sub>2</sub>/g dry feather. The treated slurry was centrifuged, and the supernatant was collected as the final product. The solubilized feather protein solution (3.3%, DM basis) was added to shock + OLP corn stover (79.5%, DM basis) and corn stover solubles (17.2%, DM basis) to determine the potential for development of a combined feed ingredient suitable for replacing corn grain in cattle diets.

### **Total Digestible Nutrients**

Total digestible nutrients were used to estimate the value of each prepared sample as an animal feed. Total digestible nutrient was calculated by Dairy One, Inc. and Cumberland Valley Analytical Services, Inc. using the Weiss model (TDN<sub>W</sub>, Weiss et al., 1992), which is based on true digestibility coefficients for available soluble carbohydrates, proteins, fatty acids, and fiber. The Weiss model is suitable for raw lignocellulose that has not been pretreated to enhance digestibility.

Texas A&M University Animal Science Department also calculated an adjusted TDN (TDN<sub>N</sub>) using the Tedeschi model (Falls et al. (2017) and Tedeschi et al., 2009), which is based on measured 48-h neutral detergent fiber digestibilities (NDFD<sub>48</sub>). The Tedeschi model includes an assessment of digestibility and hence is suitable for both raw and pretreated lignocellulose.

### **In Vitro Anaerobic Fermentation and Gas Production**

Texas A&M University Animal Science Department analyzed the in vitro anaerobic fermentation of corn stover using the gas production method described by Tedeschi et al. (2009). The in vitro fermentation chamber included an incubator with a multi-plate stirrer, pressure sensors attached to incubation flasks (125-mL Wheaton bottles), an analog-to-digital converter device, and a PC-compatible computer provided with appropriate software (Pico Technology, Eaton Socon, Cambridgeshire, UK). The pressure inside each flask was automatically recorded every 5 min for 48 h (2,880 data points). Each incubation flask was loaded with feed sample (200 mg), boiled distilled water that had been cooled to room temperature (2 mL), cysteine hydrochloride (14 mL), and filtered mixed ruminal bacteria inoculum (4 mL). Recording of the pressure was initiated once the fermentation chamber reached the fermentation temperature (39°C). Fermentation pH

was maintained between 6.8 and 6.9. Once fermentation was complete, 40 mL of neutral detergent solution was added to each bottle, the bottles were crimp sealed, and placed in an autoclave for 60 min at 105°C. The undegraded fiber was filtered using Whatman 54 filter paper, and neutral detergent fiber (NDF) was determined gravimetrically. For alfalfa and shock + OLP pretreated corn stover, experiments were repeated 4 times; for corn grain, shock pretreated corn stover, and OLP pretreated corn stover, experiments were repeated 5 times; for untreated corn stover and OLP + shock pretreated corn stover, experiments were repeated 6 times. The mean and standard deviation of obtained gas production and total nutrient digestion rate for each material were calculated.

### **Experimental Design**

Five corn stover samples and two control samples (corn grain and alfalfa) were analyzed: (1) raw, (2) OLP, (3) shock, (4) OLP + shock, and (5) shock + OLP. Cumberland Valley Analytical Services, Inc. analyzed compositional differences, estimated TDN<sub>W</sub>, and measured 30-h in vitro NDF digestibility. Texas A&M University Animal Science Department measured 48-h in vitro NDF digestibility and the gas production resulting from the anaerobic fermentations.

## **RESULTS AND DISCUSSION**

### **Compositional Analysis**

Compositional analysis was performed to determine changes in composition from pretreatment (Table 1). Corn grain had significantly higher non-fiber carbohydrate (NFC) content (76.6%) than both alfalfa (34.1%) and raw corn stover (16.8%), which is why corn grain is widely used in ruminant diets. Oxidative lime pretreatment of corn stover had a negligible effect on NFC; OLP corn stover had an NFC content of 16.0%. However, shock treatment alone significantly reduced NFC (6.9%). When combined, the order of pretreatments had little effect on NFC content (10.4%). The effect of shock treatment on NFC is not well understood, and needs to be further explored.

Raw corn stover had significantly higher NDF (77.3%) than alfalfa (49.2%) and corn grain (11.4%). The primary hurdle of implementing lignocellulose in high-quality ruminant feeds is overcoming the high NDF content, which is normally highly indigestible. Both OLP and shock pretreatment processes significantly increased NDF content. Oxidative lime pretreatment alone increased NDF to 81.9%, and shock treatment alone increased NDF to 88.1%. Similar to NFC, when combined, the order of pretreatments had

little effect on NDF changes. OLP + shock had similar NDF (87.6%) to shock + OLP (87.1%).

The CP content of raw corn stover (7.1%) is only slightly lower than corn grain (8.6%), but is considerably lower than alfalfa (16.6%). As discussed by Falls et al. (2017), a significant drawback to using OLP to generate animal feed is the unavoidable degradation of protein. To some extent, protein can be protected by prewashing the corn stover to recover protein prior to OLP. Oxidative lime pretreatment reduced corn stover CP to 3.2%, whereas shock treatment had negligible effect (6.6%). When combined, OLP + shock and shock + OLP had CP contents of 4.1 and 3.1%, respectively. If OLP is used to produce animal feed, it will be necessary to supplement it with a high-protein source, such as alfalfa, soybean meal, distillers' grains, or solubilized proteins (Coward-Kelly et al., 2006).

Neither OLP nor shock treatment significantly affected the mineral composition of corn stover. Slight increases of calcium were observed, particularly with OLP alone (2.9% DM), indicating that extensive washing was unable to fully remove all calcium. Oxidative lime pretreatment also removed the majority of potassium, only leaving trace amounts. The corn stover solubles had significant calcium (1.1% DM) and potassium (11.7% DM).

Overall, with OLP + shock pretreated corn stover, NDF increased whereas NFC and CP both decreased. Based on composition alone, OLP and shock treatment negatively affect the feed value of corn stover; however, digestibility analysis provides a significantly different conclusion.

### **48-h Neutral Detergent Fiber Digestibility**

The 48-h neutral detergent fiber digestibility (NDFD) of corn stover samples, corn grain standard, and alfalfa standard was measured using in vitro anaerobic fermentation (Table 1). Previous literature has reported that improving forage NDFD increases DM intake and milk yield in dairy cows (Oba and Allen, 1999). The corn grain and alfalfa standards had NDFD values (g NDF digested/100 g NDF fed) of 63.2 and 47.9, respectively. The NDFD of raw corn stover (49.3) was similar to alfalfa. Oxidative lime pretreatment alone improved NDFD to 79.0, whereas shock treatment alone reduced NDFD to 43.9. Shock + OLP corn stover (76.0) was slightly less digestible than OLP alone; however, OLP + shock corn stover was the most digestible (79.3).

### **Total Digestible Nutrients**

The TDN of the prepared corn stover samples, corn grain standard, and alfalfa standard were estimated using two methods: (1) Weiss formula using chemical

analysis results only ( $TDN_W$ ) and (2) Tedeschi formula which incorporates experimentally measured 48-h NDFD ( $TDN_N$ ). Table 1 shows the TDN results derived from both methods on both a DM and OM basis. In this section, all TDN results discussed are presented as g nutrients digested/100 g OM fed ( $TDN_{OM}$ ).

Because of its high NFC content, corn grain had the highest  $TDN_{OM_W}$  (87.1) and  $TDN_{OM_N}$  (88.1). Both methods estimated comparable values for alfalfa (61.5 and 59.4) and corn stover (52.2 and 51.9) for  $TDN_{OM_W}$  and  $TDN_{OM_N}$ , respectively. Because of the low NDFD for shocked corn stover, the models resulted in similar values: 43.8 ( $TDN_{OM_W}$ ) and 40.2 ( $TDN_{OM_N}$ ).

As discussed previously, OLP, OLP + shock, and shock + OLP all increased NDFD, resulting in differences between the 2 TDN estimation methods. In all 3 cases,  $TDN_{OM_N}$  was much greater than  $TDN_{OM_W}$  because it accounts for the improved NDFD resulting from the biomass pretreatment methods. For OLP and OLP + shock corn stover,  $TDN_{OM_N}$  were 59.7 and 69.7, respectively. Of the corn stover samples, shock + OLP demonstrated the highest  $TDN_{OM_N}$  (72.6), a difference of 18.4 from the calculated  $TDN_{OM_W}$  value. These modified TDN values show the effectiveness of the pretreatment processes, and demonstrate that traditional forage empirical models cannot predict the feed value of high-digestibility lignocellulose.

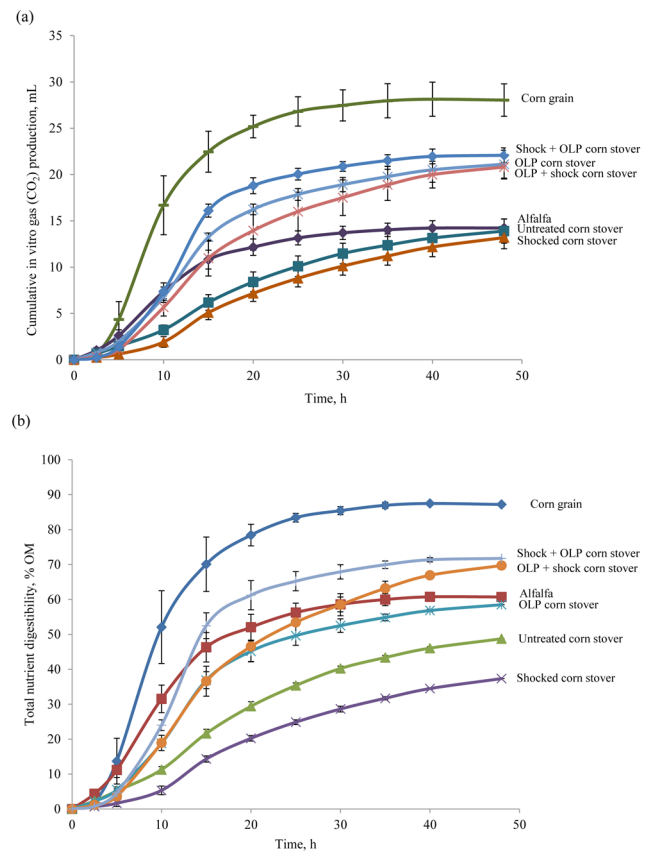
### In Vitro Gas Production

During the 48-h in vitro anaerobic fermentation used to measure NDFD, a pressure sensor was attached to the incubation flask. This pressure sensor measured gas production during fermentation. The sensor recorded the pressure every 5 min for the duration of the fermentation (48 h), resulting in 2,880 data points. The resulting gas production plot [Fig. 2(a)] can be correlated to fermentation rate. Combining  $TDN_{OM_N}$  and gas production, the rate of nutrient digestion can be plotted [Fig. 2(b)].

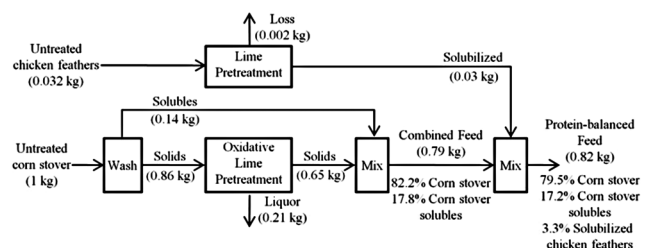
### Addition of the Soluble Extractives

The raw corn stover was thoroughly washed with hot water to extract soluble components (approximately 14% by dry weight). Table 1 shows the composition of the extractives. The extractives had a  $TDN_W$  (g nutrients digested/100 g nutrients fed) of 61.0 on a DM basis, or 88.9 on an OM basis. [Note: NDFD (48 h) was not determined for the extractives, so  $TDN_N$  could not be calculated; however, the NDF content was so low the 2 TDN methods should produce comparable values.]

Figure 3 shows a mass balance for each process step on a DM basis. Of untreated corn stover, 14% was soluble and OLP solids yield was 75%. Combining the corn stover



**Figure 2.** (a) Gas production (mL) of corn grain, alfalfa, and corn stover samples during in vitro anaerobic fermentation. (b) Total nutrient digestion rate of corn grain, alfalfa, and corn stover samples calculated using in vitro gas production and  $TDN_{OM_N}$  on an OM basis. (Note: Each curve is the average of multiple runs. Error bars represent  $\pm 1$  standard deviation.)



**Figure 3.** Mass balance for combining OLP corn stover with pre-washed corn stover soluble extractives and solubilized chicken feathers.

ver sample with the highest  $TDN_N$  (shock + OLP) with extractives is 17.8% extractives and 82.2% shock + OLP.

On a DM basis, shock + OLP corn stover had a  $TDN_N$  of 66.6, and the extractives had a  $TDN_W$  of 61.0. Their combined TDN is calculated as follows:

$$TDN \text{ of combined feed} = (0.822)(66.6) + (0.178)(61.0) = 65.6$$

This combined TDN (65.6 g nutrients digested/100 g nutrients fed) shows a slightly negative effect from adding the extractives to the treated corn stover, and is con-

siderably lower than corn grain (−20.5). This is because of the high ash content in the extractives material.

On an ash-free basis, the combined TDN can be calculated as follows:

$$\text{TDN of combined feed, ash-free basis} = \frac{(0.822)(66.6) + (0.178)(61.0)}{(0.822)(1 - 0.083) + (0.178)(1 - 0.314)} = 74.9$$

This compares more favorably to ash-free corn grain (−12.3).

### **Solubilized Protein**

Protein degradation is an unavoidable consequence of OLP, necessitating the development of protein supplementation strategies. Coward-Kelly et al. (2006) used lime treatment to solubilize chicken feathers, resulting in a liquid rich in amino acids and poly-peptides. One potential concern with using highly soluble protein is ammonia production in the rumen; however, solubilized protein from chicken feathers produces similar levels of ammonia as soybean meal or cottonseed meal, and substantially less than urea (Coward-Kelly et al., 2006).

This study determined the macronutrient and micronutrient (Table 1) composition of solubilized protein from chicken feathers. On an OM basis, the solubilized protein was comprised almost solely of CP (95.9%), with the second largest constituent being NFC (2.9%). The solubilized protein contained some ash (7% DM), which was primarily calcium (3.3% DM). Because of its low NDF content (1.0% OM), TDNom<sub>W</sub> and TDNom<sub>N</sub> were very similar (92.6% OM and 94.1% OM, respectively).

Adding solubilized chicken feathers to the combined feed (0.037 kg solubilized chicken feathers/kg combined feed) produces a protein-balanced feed with the same CP content of corn grain (Fig. 3). This feed is comprised of 79.5% shock + OLP corn stover, 17.2% corn stover solubles, and 3.3% solubilized chicken feathers; on an OM basis, the resulting TDNom<sub>N</sub> is 75.5 g nutrients digested/100 g OM. Table 1 shows the macronutrient composition of the combined and protein-balanced feeds, and provides the mineral content of each. Adding solubilized chicken feathers increases

TDN<sub>N</sub> of the combined feed (+0.7). The balanced feed is slightly less digestible than corn grain (−12.6). Of the 12.6 difference, lignin alone accounts for 6.0, making it difficult to narrow the gap further.

### **Conclusions**

With corn stover, shock + OLP improved the 48-h NDFD to 79.0 g NDF digested/100 g NDF fed, compared to 49.3 for raw corn stover. Shock treatment did not further improve NDFD, but did increase TDN. On an OM basis, shock + OLP corn stover had a TDNom<sub>N</sub> of 72.6, which approached that of corn grain (88.1). When extractives are added, TDNom<sub>N</sub> increases to 74.9, which is only 13.2 less than corn grain. When enough solubilized chicken feathers are added to match the protein content of corn grain, TDNom<sub>N</sub> increases to 75.5, which is only 12.6 less than corn grain.

### **LITERATURE CITED**

- Coward-Kelly, G., V. S. Chang, F. K. Agbogbo, and M. T. Holtzaple. 2006. Lime treatment of keratinous materials for the generation of highly digestible animal feed: 1. Chicken feathers. *Bioresour. Technol.* 97:1337–1343. doi:10.1016/j.biortech.2005.05.021
- Falls, M., D. Meysing, S. Lonkar, C. Liang, M. N. Karim, G. Carstens, L. Tedeschi, and M. T. Holtzaple. 2017. Development of highly digestible animal feed from lignocellulosic biomass Part 1: Oxidative lime pretreatment (OLP) and ball milling of forage sorghum. *Transl. Anim. Sci.* 1. doi:10.2527/tas2017.002
- Falls, M., R. Sierra-Ramirez, and M. Holtzaple. 2011. Oxidative lime pretreatment of dactoth switchgrass. *Appl. Biochem. Biotechnol.* 165:243–259. doi:10.1007/s12010-011-9247-6
- Oba, M., and M. S. Allen. 1999. Evaluation of the Importance of the Digestibility of Neutral Detergent Fiber from Forage: Effects on Dry Matter Intake and Milk Yield of Dairy Cows. *J. Dairy Sci.* 82:589–596. doi:10.3168/jds.S0022-0302(99)75271-9
- Rajaratnam, S., Z. Bano, and K. H. Steinkraus. 1989. *Pleurotus* mushrooms. Part III. Biotransformations of natural lignocellulosic wastes: Commercial applications and implications. *Crit. Rev. Food Sci. Nutr.* 28:31–113. doi:10.1080/10408398909527491
- Tedeschi, L. O., P. J. Kononoff, K. Karges, and M. L. Gibson. 2009. Effects of chemical composition variation on the dynamics of ruminal fermentation and biological value of corn milling (co) products. *J. Dairy Sci.* 92:401–413. doi:10.3168/jds.2008-1141
- Weiss, W. P., H. R. Conrad, and N. R. St. Pierre. 1992. A theoretically-based model for predicting total digestible nutrient values of forages and concentrates. *Anim. Feed Sci. Technol.* 39:95–110. doi:10.1016/0377-8401(92)90034-4