

## FORAGES AND FEEDS: *Short Communication*

# Cottonseed characteristics related to beef cattle consumption: Protein degradability, digestibility, and gossypol content\*

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### ABSTRACT

**Objective:** Our objectives were to (1) evaluate effects of whole cottonseed (WCS) storage conditions on protein and digestibility characteristics and (2) characterize protein degradability, gossypol content, and seed characteristics of cotton (*Gossypium hirsutum* L.) in the southern United States.

**Materials and Methods:** Seed with 0% heat damage (NN), 50% heat damage (N/B), or 100% heat damage (BB) was analyzed for in situ digestibility and protein degradability in Exp. 1. In Exp. 2, seed from 88 cotton breeding lines was obtained from a public cotton breeding program and analyzed for CP, undegradable intake protein (UIP), degradable intake protein (DIP), gossypol concentrations, and seed size.

**Results and Discussion:** In situ DM disappearance was greatest ( $P < 0.001$ ) for NN after 48 h, and N/B was greater ( $P < 0.05$ ) than BB. Cottonseed that was not burned had a greater ( $P = 0.01$ ) amount of N degradable at a definable rate (B fraction) than BB, and the undegradable fraction (C fraction) tended to be less ( $P = 0.06$ ) for NN than BB and N/B. Cotton breeding lines had a mean CP value similar to those reported by the NASEM (2016), whereas DIP, UIP, and gossypol values had greater variability than CP in the present study. Correlations between seed size and protein were low, but there was a positive correlation with free gossypol.

**Implications and Applications:** Heat-damaged WCS has decreased DM disappearance and N disappearance. Characterization of DIP, UIP, and gossypol in cottonseed may provide baseline selection criteria for cotton breeding

programs, as well as aid in ration formulation strategies. Measuring WCS quality can help cotton breeders, animal scientists, and livestock producers develop seed value and feed recommendations for use.

**Key words:** whole cottonseed, degradable intake protein, undegradable intake protein, gossypol, overheating

### INTRODUCTION

Whole cottonseed (WCS) is a byproduct feedstuff used as both a protein and energy supplement for beef cattle operations, particularly during the winter months. The effects of seed handling or storage of cottonseed influence feed quality. Depending on cotton moisture concentration entering the storage period after ginning, cottonseed may naturally undergo heating. When feeds are heat damaged, nutrients such as proteins and sugars are bound via the Maillard or browning reaction and become less digestible (Goering et al., 1973). Beef cattle operators may choose to use overheated cottonseed as an economical supplement source, but the effects of heat damage on protein characteristics and subsequent seed feed value are not well defined.

Recommendations for seed consumption are primarily based on seed oil content being the limiting factor in ruminant diets (Myer et al., 2013). Cotton also produces a secondary metabolite, gossypol, which can cause sterility in livestock if a threshold is met (Chase et al., 1994; Chenoweth et al., 2000). These recommendations and thresholds were largely established in the late 1990s, and since that time, cottonseed composition has changed. Campbell et al. (2011) reported that cotton breeders have driven partitioning during the plant selection phase toward cotton fiber development rather than developing seed. As yield has increased in modern cultivars, seed size has decreased (Snider et al., 2016), which may alter nutrient composition in terms of ruminant livestock feed values.

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It is unknown what the diversity for these nutrients is among the existing cotton germplasm (Bertrand et al., 2005). There are few reports of published trials for protein degradability characteristics of WCS that exist, and the gossypol levels and oil content in newer genetic material has not been well described.

The objectives of this study were to investigate (1) the effects of storage conditions on WCS protein and digestibility characteristics and (2) protein degradability, gossypol content, and seed characteristics of cotton breeding lines adapted to the southern United States.

## MATERIALS AND METHODS

### Exp. 1

**Animal Care and Use.** All procedures were approved by the Institutional Animal Care and Use Committee of the University of Georgia (AUP # A2018 10-023-Y2-A0).

**Treatments.** Two samples of WCS exhibiting different degrees of heat damage were obtained from a cattle operation in Hazlehurst, Georgia, which was sourced from South Georgia Cotton Gin in Hazlehurst. The heat-damaged WCS was collected from a lot described by the gin as heat damaged due to fire; all seeds appeared dark in color. Additionally, samples were collected from a lot that did not appear damaged, and the seeds were white in color. Experimental treatments included 0% heat damaged (NN), 50% heat damaged (N/B), or 100% heat damaged (BB). The N/B treatment was constructed by mixing 50% of NN and BB by weight. Samples were submitted to the Agricultural and Environmental Services Laboratory (Athens, GA), and the nutrient analysis is presented in Table 1. Cotton samples were ground to pass through a 2-mm screen (Wiley Mill, Thomas Scientific) for the subsequent *in situ* analysis.

**In Situ Digestibility.** Five grams of substrate was weighed in sextuplet into preweighed Dacron bags (10 cm × 20 cm; ANKOM Technology) of 50- $\mu$ m porosity and were triple sealed using an Impulse sealer (American International Electric). Ten samples, 2 replicates for each of 5 time points, were placed sequentially into each of 3 ruminally cannulated beef steers in a completely randomized design. Steers were fed a diet consisting of *ad libitum* access to bermudagrass (*Cynodon dactylon* L.) hay in Athens, Georgia, in January 2020. Steers were fed this diet throughout the duration of the trial.

Sealed bags containing NN, N/B, and BB WCS were soaked in 36°C water for 5 min before being placed inside the rumen for incubation for 0, 6, 12, 24, and 48 h in nylon mesh laundry bags. Bags were placed in the rumen in reverse order so that all bags were removed at the 48-h time point. Upon removal from the rumen, samples were immediately placed into an ice bath for 30 min to inhibit microbial activity. Bags were then rinsed by hand until the rinse water was clear, placed in an oven and dried at 60°C for 24 h, and weighed. Samples from 24 and 48 h were used to evaluate DM disappearance.

**Table 1.** Nutrient analysis<sup>1</sup> of heat-damaged whole cottonseed

Item	Whole cottonseed <sup>2</sup>		
	NN	N/B	BB
<b>Chemical analysis</b>			
CP, %	19.32	19.85	19.71
Crude fiber, %	1.43	0.98	1.11
ADF, %	33.80	35.12	49.00
NDF, %	47.89	46.15	51.45
Fat, %	14.64	16.64	16.48
Ash, %	10.64	5.95	3.77
<b>Mineral analysis</b>			
Calcium, %	1.42	1.59	2.28
Potassium, %	1.70	1.55	1.84
Magnesium, %	0.40	0.43	0.34
Sodium, %	0.17	0.21	0.24
Sulfur, %	0.44	0.45	0.43
Phosphorus, %	0.97	1.24	1.11
Chloride, %	0.38	0.40	0.50
Copper, mg/kg	11.53	12.02	13.03
Iron, mg/kg	66.56	—	—
Manganese, mg/kg	46.99	51.78	57.98
Zinc, mg/kg	41.62	36.59	35.06

<sup>1</sup>Analysis performed at Agricultural and Environmental Services Laboratory (Athens, GA).

<sup>2</sup>NN = 0% of whole cottonseed sample heat damaged, N/B = 50% of whole cottonseed sample heat damaged, and BB = 100% of whole cottonseed sample heat damaged.

Approximately 0.20 g of WCS residue from each bag was weighed in duplicate and wrapped in aluminum foil for nitrogen analysis. Samples were analyzed using a Leco Nitrogen Analyzer (Model FP268, LECO Corporation).

**Statistical Analysis.** Nonlinear regression was used to analyze N disappearance curves using the NLIN procedure in SAS (SAS Institute Inc.). Fractions of N were partitioned based on relative susceptibility to ruminal degradation as described by Ørskov and McDonald (1979). Cottonseed was partitioned into fractions A, B, and C with A representing the immediately soluble fraction, B describing the fraction that disappeared at a definable rate, and fraction C depicting the undegradable portion (NASEM, 2000). Disappearance rate ( $K_d$ ) was determined by the nonlinear regression model for fraction B. Fraction C was then calculated by difference [100 - (A + B)]. Dry matter disappearance and differences in A, B, and C fractions and disappearance rate were analyzed using the PROC GLM procedure in SAS (SAS Institute Inc.). Significance was declared at  $P \leq 0.05$ , and trends were discussed when  $P > 0.05$  or  $\leq 0.10$ .

### Exp. 2

Whole cottonseed samples following the 2018 growing season were collected for characterization of CP, unde-

gradable intake protein (UIP), degradable intake protein (DIP), and total and free gossypol concentrations. Cottonseed was harvested from 88 public breeding lines and 1 commercial cultivar Deltapine 1646 (Table 2). This material was planted, grown, and evaluated in field plots for its usefulness in the Auburn University breeding program with seed nutritional traits as a secondary objective. Seed samples for nutritive value analysis described herein were derived from a handpicked 25-boll sample before mechanical harvest and ginned using a 16-blade laboratory standing gin. Four replications were combined to generate a composite sample for analysis. Whole seeds were weighed, delinted, and then ground to pass a 2-mm screen (Wiley Mill, Thomas Scientific) for analysis.

**Protein Degradability.** Seed samples were analyzed in duplicate for total N concentration using the Kjeldahl Analyzer Unit Foss Tecator according to AOAC (1990) procedures. Crude protein concentration was calculated as N concentration  $\times$  6.25. Percentage of DIP in WCS samples was determined using a *Streptomyces griseus* protease procedure (Type XIV Bacterial; Sigma-Aldrich Co.) as described by Mathis et al. (2001). Based on this assay, each sample was weighed to obtain 15 mg of N based on N concentration of the sample and placed in 125-mL Erlenmeyer flasks. A total of 40 mL of a borate-phosphate buffer solution was added to each flask and incubated at 39°C for 1 h in a shaker water bath. After incubation, 10 mL of protease solution was added to each flask and incubated for 16 h at 39°C in a shaker water bath. Following the 16-h incubation, samples were filtered through Whatman #540 filter paper using a cone-shaped funnel and rinsed with 400 mL of distilled H<sub>2</sub>O to remove any incubation media. Samples were then dried in a 100°C oven for 24 h to obtain residual DM weight. Samples were analyzed for N using the Kjeldahl Analyzer Unit Foss Tecator. Percentage UIP was calculated by dividing the milligrams of residual N by the milligrams of initial N and multiplying by 100. Percentage DIP was calculated by subtracting percentage UIP from 100 for each sample.

**Gossypol and Seed Index.** Total and free gossypol concentrations in seed from cotton breeding lines were measured and determined according to AOCS Method Ba 7-58 (AOCS, 2020) by ATC Scientific Laboratory (North Little Rock, AR). Seed index, a measure of seed size, was reported for each breeding line as the weight (g) of 100 seeds from samples before delinting.

**Statistical Analysis.** Seed protein characteristics, gossypol concentration, and seed index were characterized using descriptive statistics to determine minimum, maximum, and mean values for each parameter in the genetic material screened and are reported in Table 2. A Jarque-Bera normality test was performed for seed protein measures, gossypol, and seed index to determine relative distribution of values across germplasm evaluated. Correlations of CP, DIP, UIP, and total and free gossypol with seed index were conducted to evaluate the relationship of

physical seed attributes with potential feed nutritive value parameters.

## RESULTS AND DISCUSSION

### Exp. 1

Dry matter disappearance for WCS was not different among seed treatments ( $P = 0.26$ ) at 24 h. However, DM disappearance was greater ( $P < 0.001$ ) for NN than all other treatments, and N/B was greater ( $P < 0.05$ ) than BB at 48 h (Figure 1). Arieli et al. (1989) reported that heating WCS at 180°C for 2 h decreased 48-h DM disappearance in the rumen compared with no or lower temperature exposure (140 and 160°C). This would suggest that the heat-damaged seed in the current experiment exceeded 180°C for at least 2 h, likely causing the degradation or binding of material that normally would be available to ruminal fermentation. Similarly, Dahlke (2013) reported in vitro true DM disappearance for dried distillers grains at 4 visually increasingly different levels of heat damage. These authors reported decreasing in vitro true DM disappearance of 98.4, 88.0, 75.7, and 54.2% for normal, mild heat damage, heat damage, and extreme heat damage, respectively.

The effect of heat damage on ruminal degradable CP fractions is presented in Table 3. The immediately degradable fraction (A) was not different ( $P = 0.58$ ) among WCS treatments; however, there was an effect ( $P = 0.03$ ) of heat damage on the fraction degradable at a definable rate (B) and a tendency ( $P = 0.06$ ) for the fraction unavailable to ruminal degradation (C). These results differ from those presented by Arieli et al. (1989), who reported that the A fraction of WCS heated for 1 or 2 h at 140, 160, and 180°C was decreased compared with the control, and the B fraction was increased. The amount of time or temperature in which WCS was exposed to in the present study is not known; however, it likely exceeded 180°C or was heated for longer than 2 h. Whole cottonseed in the NN treatment had a greater amount ( $P = 0.01$ ) of CP degradable at a measurable rate compared with the BB treatment but was not different ( $P = 0.11$ ) than the N/B treatment. Rate of CP degradation was not different ( $P = 0.34$ ) among treatments. In a review by Arieli (1998), it was estimated that about one-half of the rumen-degradable CP in WCS is contributed by the soluble fraction based on 3 in vitro experiments. This observation aligns with values for the A and B fractions of NN and N/B seed in the current experiment.

The fraction of CP unavailable to ruminal degradation (C) tended to be greater ( $P = 0.06$ ) for BB, compared with NN and N/B. The negative effect of heating on protein digestibility by reactions such as the Maillard reaction is well known (Goering et al., 1973). Pena et al. (1986) suggested that roasting or extrusion of WCS may decrease in vivo rumen ammonia level when WCS comprised 40% of the dietary DM in dairy cattle diets. Whole cottonseed

**Table 2.** A snapshot of the range of seed characteristics related to protein degradability, gossypol concentration, and seed index in a public cotton breeding program

Source <sup>1</sup>	Breeding line <sup>2</sup>	CP, %	UIP, <sup>3</sup> %	DIP, <sup>4</sup> %	Free gossypol, %	Total gossypol, %	Seed index, <sup>5</sup> g
AU	4051	21.3	50.3	49.7	1.1	1.2	9.7
AU	4079	19.8	44.2	55.8	1.2	1.3	9.9
AU	5315	21.5	49.6	50.4	1.1	1.2	10.7
AU	5346	19.9	57.4	42.6	1.0	1.1	9.8
AU	5418	23.9	45.9	54.1	—	—	10.9
AU	5428	22.3	44.4	55.6	0.9	1.0	10.7
AU	6001	21.6	47.5	52.5	1.2	1.2	10.7
AU	6126	22.1	53.1	46.9	1.1	1.2	10.7
AU	6202	20.2	49.5	50.5	1.2	1.4	10.9
AU	6252	23.1	49.5	50.5	—	—	10.7
AU	10090	27.0	34.7	65.3	1.0	1.1	9.9
AU	52021	23.3	50.0	50.0	1.2	1.3	11.3
AU	67059	19.9	46.5	53.5	0.7	1.0	9.9
AU	68088	21.9	41.2	58.8	1.1	1.2	9.8
AU	70001	20.9	38.6	61.4	1.0	1.2	9.9
AU	70049	23.2	41.1	58.9	0.9	1.2	10.5
AU	70062	21.4	39.7	60.3	1.2	1.5	10.6
AU	71069	20.7	39.7	60.3	1.1	1.4	10.2
AU	72021	25.1	33.1	66.9	1.0	1.3	10.3
AU	72028	24.8	56.7	43.3	0.8	1.2	10.5
AU	73055	20.7	36.2	63.8	0.9	1.2	10.9
AU	74044	23.3	41.6	58.4	—	—	9.6
AU	74088	27.2	57.4	42.6	0.8	1.0	10.1
AU	76008	23.4	41.8	58.2	0.9	1.1	10.0
AU	76036	21.1	38.1	61.9	1.1	1.3	11.1
AU	76038	23.2	46.9	53.1	0.9	1.1	12.4
AU	76074	21.9	47.7	52.3	0.7	0.9	10.9
AU	77009	21.9	49.7	50.3	0.7	1.1	9.9
AU	77053	22.9	45.8	54.2	0.9	1.4	11.0
AU	78080	21.5	44.0	56.0	1.1	1.3	11.2
AU	79056	16.1	42.2	57.8	0.9	1.2	10.1
AU	79085	25.2	41.6	58.4	0.9	1.3	10.4
AU	79094	27.3	49.7	50.3	0.8	1.1	8.9
AU	80003	25.1	37.0	63.0	0.9	1.0	9.1
AU	80006	23.4	42.8	57.2	0.8	1.0	11.0
AU	80030	21.8	44.2	55.8	0.9	1.4	11.7
AU	80065	22.7	49.6	50.4	—	—	9.8
AU	80098	23.1	57.4	42.6	—	—	11.4
AU	81019	23.3	45.9	54.1	1.0	1.1	11.2
AU	81025	23.7	38.1	61.9	1.1	1.4	10.0
AU	81043	23.0	46.5	53.5	1.3	1.3	10.9
AU	81071	24.1	49.7	50.3	1.1	1.1	10.9
AU	81097	25.2	52.3	47.7	1.0	1.1	10.3
AU	82028	21.2	36.7	63.3	0.8	1.0	10.7
AU	82074	22.9	50.9	49.1	—	—	11.3
AU	83046	24.1	38.6	61.4	1.0	1.1	10.3
AU	83060	25.5	36.9	63.1	0.9	1.0	9.7
AU	83100	25.3	39.7	60.3	0.8	0.9	10.5
AU	90098	25.0	49.9	50.1	—	—	10.0
AU	121036	25.0	49.9	50.1	0.8	1.0	9.6
AU	122034	21.8	50.4	49.6	—	—	10.1

*Continued*



**Table 2 (Continued).** A snapshot of the range of seed characteristics related to protein degradability, gossypol concentration, and seed index in a public cotton breeding program

Source <sup>1</sup>	Breeding line <sup>2</sup>	CP, %	UIP, <sup>3</sup> %	DIP, <sup>4</sup> %	Free gossypol, %	Total gossypol, %	Seed index, <sup>5</sup> g
AU	122045	23.4	48.7	51.3	1.0	1.1	10.5
AU	122048	24.5	49.7	50.3	1.0	1.2	11.1
AU	122072	22.7	52.3	47.7	1.1	1.1	11.0
AU	123037	24.5	48.5	51.5	0.9	1.0	11.7
AU	124016	23.6	45.8	54.2	1.0	1.3	9.4
AU	126069	23.2	52.5	47.5	1.0	1.2	12.2
AU	128071	26.3	49.1	50.9	0.7	1.0	10.4
AU	128089	25.1	51.4	48.6	0.9	1.0	10.4
AU	128092	27.7	49.0	51.0	1.0	1.0	9.2
AU	134079	25.6	51.4	48.6	1.1	1.1	11.3
Check	DP1646	23.7	52.3	47.7	0.5	0.7	8.0
Check	Red plant	25.0	49.9	50.1	—	—	9.4
RBTN	16-13P1115	25.2	47.9	52.1	0.2	0.2	13.4
RBTN	Ark 1004-38	24.4	47.9	52.1	0.8	1.2	10.1
RBTN	Ark 1005-35	22.8	50.3	49.7	0.9	1.4	12.0
RBTN	Ark 1005-41	24.4	56.7	43.3	0.8	1.4	11.9
RBTN	Ark 1007-15	26.6	36.7	63.3	0.9	1.3	11.1
RBTN	Ark 1015-42	24.5	50.3	49.7	0.8	1.2	11.6
RBTN	DP 393	23.3	46.7	53.3	1.0	1.2	11.7
RBTN	DP 493	24.6	38.6	61.4	0.8	0.8	9.6
RBTN	FM958	23.2	37.0	63.0	1.0	1.2	11.0
RBTN	GA2012141	24.8	52.3	47.7	0.7	1.2	11.4
RBTN	GA2015024	25.8	49.5	50.5	0.7	0.9	11.9
RBTN	LA 11309040	25.1	38.6	61.4	0.9	1.2	11.4
RBTN	LA14063075	27.5	38.6	61.4	0.8	1.2	12.0
RBTN	LA14063083	25.6	38.6	61.4	0.9	1.3	10.8
RBTN	MS 2010-875	23.9	45.8	54.2	1.0	1.1	12.5
RBTN	PD 2011026	25.7	45.8	54.2	0.9	1.1	11.2
RBTN	PD 2011081	22.5	45.8	54.2	1.0	1.2	12.0
RBTN	PD2011021	25.1	45.8	54.2	0.9	1.2	11.2
RBTN	SG105	22.5	56.2	43.8	1.4	1.5	9.4
RBTN	TAM 12J-39	25.5	39.7	60.3	1.0	1.2	14.6
RBTN	TAM LBB 150107	26.5	38.4	61.6	1.2	1.2	11.0
RBTN	TAM LBB 150824	25.3	38.3	61.7	1.1	1.3	11.1
RBTN	TAM LBB15092	24.2	45.8	54.2	1.3	1.4	11.0
RBTN	TAM13S-03	23.1	38.2	61.8	0.9	1.0	10.9
RBTN	UA222	20.8	40.3	59.7	1.1	1.4	11.0
Mean	—	23.6	45.7	54.3	1.0	1.2	10.7
SD <sup>6</sup>	—	2.0	6.0	6.0	0.2	0.2	1.0
Maximum	—	27.7	57.4	66.9	1.4	1.5	14.6
Minimum	—	16.1	33.1	42.6	0.2	0.2	8.0

<sup>1</sup>AU = Auburn University Cotton Breeding Program; RBTN = Regional Breeders Technical Network; mean, maximum, and minimum values for each parameter are presented across all lines.

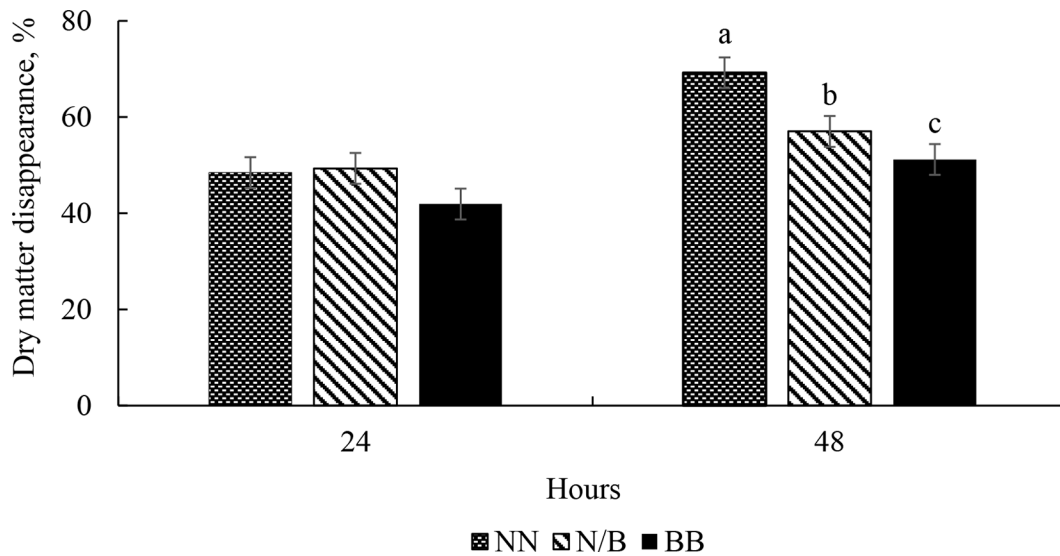
<sup>2</sup>Identifier code for individual cotton breeding lines.

<sup>3</sup>UIP = undegradable intake protein (% DM basis).

<sup>4</sup>DIP = degradable intake protein (% DM basis).

<sup>5</sup>Weight (g) of 100 non-delinted whole seeds.

<sup>6</sup>Standard deviation of the mean.



**Figure 1.** In situ DM disappearance of whole cottonseed heat damaged at different percentages (NN = 0% heat damaged, N/B = 50% heat damaged, and BB = 100% heat damaged). No differences in DM disappearance were observed at 24 h ( $P > 0.26$ ). At 48 h, columns with differing letters are different ( $P < 0.05$ ). Treatments were compared within time points. <sup>a-c</sup>Columns without a similar letter are different.

has been observed to be less sensitive to heat damage than other feedstuffs such as alfalfa, almond hulls, or safflower seeds; however, acid detergent insoluble nitrogen in WCS increased with time of heating exposure (Fadel, 1990). Differences in the C fraction of the present study indicated that heat-damaged WCS has more CP unavailable to ruminal degradation compared with WCS with no heat damage; however, the similarity between NN and N/B was unexpected. Heat treatment of feedstuffs may reduce rumen DIP value but increase the quantity of intestinal unavailable protein (Arieli, 1998), but this was not measured in the current study.

## Exp. 2

Characterization of protein fractions based on a range of germplasm derived from various cotton-growing regions of the United States may help refine beef or dairy cattle diet formulations for which WCS is the main contributor to protein supplementation in these systems. This information may help plant breeders couple feed nutritional value characteristics of seed with agronomic traits in plant breeding material. Crude protein concentration of cotton lines evaluated ranged from 16.1 to 27.7% CP (Table 2) with a mean CP of  $23.6 \pm 2.0\%$ , and values followed a normal distribution ( $P = 0.458$ ). The NASEM (2016) feed composition tables are widely used as reference values for nutritional characteristics of various feedstuffs and reports a CP percentage for WCS of  $22.87 \pm 2.53\%$  on a DM basis ( $n = 536$  samples), illustrating similar CP characteristics across a larger number of samples than the present study. Dowd et al. (2018) noted that CP concentration ranged from 16.7 to 24.6% across 24 cotton cultivars harvested over a 3-yr period and that shifts in average CP concentration in cottonseed appear to be relatively minimal due

to sustained breeding efforts over the years, which may be in part due to cotton selection for nitrogen use efficiency (Main et al., 2013).

Across the cotton lines evaluated in this study, seed contained a range of 42.6 to 66.9% DIP with a mean value of  $54.3 \pm 6.0\%$  (Table 2). Percentage of UIP ranged from 33.1 to 57.4% UIP with a mean value of  $45.7 \pm 6.0\%$ . Seed DIP and UIP concentration of cotton breeding lines followed a normal distribution ( $P = 0.827$ ), with the greatest number of observations between 50 and 55% DIP and 45 to 50% UIP ( $n = 37$  observations for each, respectively). There are few published reports of DIP or UIP characteristics of cottonseed. Degradable intake protein percentage is less and UIP percentage of WCS is greater in the present study than values reported in the review by Arieli (1998). A review by Arieli (1998) reported a DIP concentration of  $74 \pm 16\%$  (mean  $\pm$  SD) for WCS across 3 in vivo experiments with cattle. The author also noted that a similar rumen CP degradability of  $77 \pm 11\%$  was observed from in situ rumen incubation in 13 studies using sheep, beef steers, and dairy cows. The NASEM (2000) feed composition tables (p. 196; Table 1) indicate that WCS has an average of 69.6% DIP. However, this same value is also reported in previously published versions from 1996, and the number of samples from which this value is derived is not reported. There are no values for DIP or UIP of WCS in NASEM (2016) feed composition tables (p. 316, Table 18-1). Furthermore, cotton gins process cotton from surrounding farms where growers choose a respective cotton variety for their operation on a year-to-year basis. On average, cotton growers plant 1 to 2 cotton varieties per year, but a gin may process multiple varieties from individual farms within a season (Raper et al., 2019). Seed obtained from cotton gins represents mixing of various

**Table 3.** In situ CP degradation of whole cottonseed with varying proportions of heat damage

Item	Whole cottonseed <sup>1</sup>			SEM	P-value
	NN	N/B	BB		
Fraction, <sup>2</sup> % of CP					
A	21.6	18.3	24.1	3.6	0.58
B	70.0 <sup>a</sup>	61.9 <sup>ab</sup>	54.0 <sup>b</sup>	3.3	0.03
C	8.4 <sup>y</sup>	19.8 <sup>z</sup>	21.9 <sup>z</sup>	3.4	0.06
Rate of degradation, %/h	7.38	8.89	5.76	1.1	0.34

<sup>a,b</sup>Means with differing superscripts with a row are different ( $P < 0.05$ ).

<sup>y,z</sup>Means with differing superscripts with a row are different ( $P < 0.10$ ).

<sup>1</sup>NN = 0% of whole cottonseed sample heat damaged, N/B = 50% of whole cottonseed sample heat damaged, and BB = 100% of whole cottonseed sample heat damaged.

<sup>2</sup>Degradation fraction: A = fraction immediately degradable; B = fraction degradable at a measurable rate; C = fraction unavailable to rumen.

contribution levels of commercial cultivars, but is likely not composed of more than 3 to 5 cotton varieties based on grower variety selection criteria. Thus, CP in seed obtained from within a respective cotton gin may be a more stable parameter, but protein degradability characteristics may be more variable. Characterization of DIP and UIP for cottonseed may further refine ration development recommendations in beef cattle operations, especially for grower rations, and values reported herein begin to elucidate a range of values for Upland cotton.

Gossypol is a naturally occurring terpenoid found in pigment glands throughout the cotton plant that poses potential toxicity issues in ruminant and nonruminant animals (Wedegaertner and Rathore, 2015). Free gossypol is the more biologically active and toxic form for ruminants, whereas bound gossypol is attached to an AA and is considered nontoxic for ruminants (Gadelha et al., 2014). Both total and free gossypol concentration values followed a non-normal distribution ( $P < 0.0001$  for both parameters, respectively). The majority of values for free gossypol concentration were  $\geq 0.80$  and  $\geq 0.6\%$  for total gossypol ( $n = 77$  observations for each parameter, respectively; Table 2), but values were negatively skewed to as low as 0.2% for both variables. Gossypol concentration of seed can be influenced by weather, stand management, genetic selection pressures, and plant breeding goals. Bertrand et al. (2005) reported total gossypol values of 0.53 to 0.77% over 2 growing seasons in South Carolina. Nida et al. (1996) observed a total gossypol concentration range of 0.72 to 1.63% for cotton cultivars with RoundUp Ready technology. Additionally, the use of irrigation and other stand management practices can also influence seed gossypol concentration (Pettigrew and Dowd, 2014). These data illustrate the diversity in total and free gossypol concentrations in genetic material from a public cotton breeding program, which can be used as a reference point for plant breeders, while providing insight into ruminant livestock feed value.

Dowd et al. (2018) noted that cotton breeding efforts to increase fiber yield have led to shifts in seed and fiber compositional properties over the last several decades. Seed index is the accurate way to estimate seed size. In their evaluation of cotton cultivars, seed index averaged  $9.75 \pm 0.99$  g, with a range of 8.08 to 11.8 g. Seed index values followed a non-normal distribution ( $P < 0.0001$ ), where 71 of the 88 cotton lines evaluated had a seed index value of 9.5 to 11.8 g, and values were positively skewed to the right up to 14.6 g. In the present study, there was a low correlation between seed index and DIP ( $r = -0.08$ ;  $P = 0.007$ ), UIP ( $r = 0.08$ ;  $P = 0.007$ ), and total gossypol ( $r = -0.17$ ;  $P = 0.028$ ). A low positive correlation was observed for CP and seed index ( $r = 0.20$ ;  $P = 0.039$ ), although this may warrant further evaluation as protein is a characteristic of interest in agronomic and feed seed value traits. There was a strong positive correlation between free gossypol and seed index ( $r = 0.75$ ;  $P = 0.050$ ) of breeding lines in this study. This study suggests that a breeding program may already possess a range of seed nutritional value traits within their current germplasm. A more in-depth study of seed collected across various growing conditions and years is needed to determine the heritability of such traits to decide if this should be a future breeding objective. Incorporation of seed nutritional value information into cotton breeding programs along with production traits may enhance seed value and applications in beef cattle systems.

## APPLICATIONS

Whole cottonseed is a readily accessible byproduct feed source for beef cattle producers and can be easily incorporated into supplementation strategies in cotton-producing areas. The manner in which WCS is handled before feeding can affect the nutritional value. Results from the current study indicate that heat-damaged WCS had decreased DM degradation and amount of N that is degrad-

able at a definable rate and an increased amount of N that is unavailable to rumen microbial degradation, but it still maintains partial feed value. When WCS is heat damaged, the decreased amount of digestible DM and N available in the rumen should be considered when feeding livestock.

Inherent seed characteristics of cotton and seed storage conditions can influence seed nutritive value in livestock diets. In cotton lines screened from the Auburn University cotton breeding program, CP concentration was less variable than DIP and UIP. Observed concentrations of both free and bound gossypol varied widely among varieties. Future evaluation of nutritive value characteristics across a wider pool of seed sources that represent various harvest seasons and environments may further refine seed feed value characteristics that can be incorporated alongside other cotton breeding traits. Results from these studies can be used to tailor feeding recommendations in beef cattle diets for WCS based on pre- and postharvest cotton management.

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