

Energy costs of feeding excess protein from corn-based by-products to finishing cattle

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ABSTRACT: The increased use of by-products in finishing diets for cattle leads to diets that contain greater concentrations of crude protein (CP) and metabolizable protein (MP) than required. The hypothesis was that excess dietary CP and MP would increase maintenance energy requirements because of the energy costs of removing excess N as urea in urine. To evaluate the potential efficiency lost, two experiments were performed to determine the effects of feeding excess CP and MP to calves fed a finishing diet at 1 × maintenance energy intake (Exp. 1) and at 2 × maintenance intake (Exp. 2). In each experiment, eight crossbred Angus-based steers were assigned to two dietary treatments in a switchback design with three periods. Treatments were steam-flaked corn-based finishing diets with two dietary protein concentrations, 13.8% CP/9.63% MP (CON) or 19.5% CP/14.14% MP (dry matter basis; ECP), containing corn gluten meal to reflect a diet with excess CP and MP from corn by-products. Each period was 27 d in length with a 19-d dietary adaptation period in outdoor individual pens followed by a 4-d sample collection in one of four open circuit respiration chambers, 2-d fast in outdoor pen, and 2-d fast in one of four respiration chambers. Energy metabolism, diet digestibility, carbon (C) and nitrogen (N) balance, oxygen consumption,

and carbon dioxide and methane production were measured. At both levels of intake, digestible energy as a proportion of gross energy (GE) tended to be greater ($P < 0.06$) in ECP than in CON steers. Metabolizable energy (ME) as a proportion of GE tended to be greater ($P = 0.08$) in the ECP steers than in the CON steers at 2 × maintenance intake. At 1 × and 2 × maintenance intake, urinary N excretion (g/d) was greater ($P < 0.01$) in the ECP steers than the CON steers. Heat production as a proportion of ME intake at 1 × maintenance tended ($P = 0.06$) to be greater for CON than for ECP (90.9% vs. 87.0% for CON and ECP, respectively); however, at 2 × maintenance energy intake, it was not different (63.9% vs. 63.8%, respectively). At 1 × maintenance intake, fasting heat production (FHP) was similar ($P = 0.45$) for both treatments, whereas at 2 × maintenance intake, FHP tended to be greater ($P = 0.09$) by 6% in ECP than in CON steers. Maintenance energy requirements estimated from linear and quadratic regression of energy retention on ME intake were 4% to 6% greater for ECP than for CON. Results of these studies suggest that feeding excess CP and MP from a protein source that is high in ruminally undegradable protein and low in protein quality will increase maintenance energy requirements of finishing steers.

Key words: beef cattle, energy loss, excess protein

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INTRODUCTION

Alternative uses of agriculture feed grains to produce ethanol and other products have led to the use of new by-products, such as distiller's grains, in which crude protein (CP) values are much greater than the original feed grain (DiLorenzo and Galyean, 2010; May et al., 2010; Hales et al., 2012). The utilization of large quantities of these by-products in finishing diets leads to diets that contain greater concentrations of CP and metabolizable protein (MP) than are required by the animal. In the body, excess nitrogen (N) is converted to urea in the liver and then excreted in the urine. Huntington and Archibeque (2000) calculated that 2.5% to 5% of whole body oxygen consumption was attributable to ureagenesis in the liver. Martin and Blaxter (1965) estimated the energy cost of synthesis of urea from ammonia to be 3.80 ± 0.57 Kcal/g of N in sheep, and Tyrrell et al. (1970) estimated the cost to be 6.1 Kcal of digestible energy (DE) per gram of excess N fed in dairy cattle.

Hales et al. (2013) fed steers finishing diets containing 0% to 45% wet distiller's grains with solubles (WDGS) and noted that N excretion in urine increased with increasing dietary WDGS and CP; however, CP concentration did not significantly affect N retention. Heat production (HP), as a percent of metabolizable energy (ME) intake (MEI), increased and energy retention (RE) as a percent of MEI decreased as dietary WDGS and CP concentration increased, suggesting that overall efficiency of energy utilization decreased with increasing CP content. These losses in energetic efficiency may potentially have a negative impact on animal growth and production efficiency.

Our hypothesis was that feeding dietary CP and MP well above requirements would increase maintenance energy requirements because of the process of removing excess N as urea. Therefore, two experiments were performed to determine the effects of feeding excess CP and MP from corn by-products on the efficiency of energy utilization of calves fed a finishing diet at $1 \times$ or $2 \times$ maintenance energy intake.

MATERIALS AND METHODS

All procedures involving live animals were approved by the West Texas A&M University/CREET Institutional Animal Care and Use Committee (approval # 03-10-14).

Cattle and Dietary Treatments

Eight Angus-based crossbred steer calves were used in two respiration calorimetry experiments.

Steers were purchased from the West Texas A&M University Nance Ranch in Canyon, TX, at a weight of approximately 150 kg and transported 47 km to the Texas A&M AgriLife Research/USDA-ARS Feedlot in Bushland, TX. Over a 3-mo period, steers were trained to a lead and to the experimental procedures used in the respiration chambers. During halter and chamber training, steers received a standard growing diet (60% concentrate and 40% roughage, dry matter [DM] basis) fed at 2% of body weight (BW). In the first experiment, steers (263 ± 1.3 kg BW) were fed at their maintenance energy requirement. Maintenance intake was determined based on the NRC (1996) as 0.077 Mcal of net energy for maintenance (NEm) per kg of metabolic BW (MBW). Steer individual BW was monitored continuously throughout the experiment, and dry matter intake (DMI) was adjusted for changes in BW. The first experiment was followed by a second experiment where the same steers (374 ± 2.3 kg BW) were fed at $2 \times$ maintenance. Steers received a standard growing diet (60% concentrate and 40% roughage, DM basis) fed at 2% of BW for a 4-mo adjustment period between experiments. Cattle were not implanted during either experiment.

In each experiment, steers were assigned to one of two dietary treatments (Table 1) in a switchback design with three periods. Treatments were steam-flaked corn (SFC)-based finishing diets with two dietary protein concentrations, which included a control finishing diet containing 13.8% CP and 9.63% MP (DM basis; CON) and an excess protein finishing diet containing 19.5% CP and 14.14% MP (DM basis; ECP). The ruminally degradable protein (RDP) concentrations in the diets were similar (8.8% vs. 9.0% of DM) and met anticipated RDP requirements for steers fed SFC-based diets (Galyean, 1996; Cooper et al., 2002). All other nutrients were supplied in concentrations/quantities to meet or exceed nutrient requirements (NRC, 1996). Each period of the switchback was 27 d in length and consisted of a 19-d dietary adaptation phase in outdoor individual pens followed by a 4-d sample collection phase in one of four open circuit, indirect respiration calorimetry chambers. Following the 4-d chamber collection phase, steers were fasted for 2 d in their outdoor individual pens and then re-entered the respiration chambers for a 2-d phase to measure fasting heat production (FHP). During the first 4 d of each new period, steers were gradually stepped up to their designated level of feed intake to avoid digestive disturbances.

In each experiment, the same steers were randomized to two groups of four steers and assigned to the same chamber during each period (within

Table 1. Ingredient and nutrient composition (DM basis) of treatment diets for 1 × maintenance and 2 × maintenance intake trials

Ingredient, % DM	Dietary treatment ^a	
	CON	ECP
SFC	67.1	57.0
Corn gluten feed ^b	18.0	14.5
Corn stalks	9.0	9.0
Corn oil	0.9	—
Corn gluten meal ^c	—	14.5
Supplement ^d	5.0	—
Supplement ^e	—	5.0
Analyzed composition, DM basis (except DM)		
DM, %	80.2	81.8
CP, %	13.8	19.5
EE, %	4.6	3.5
NDF, %	19.8	19.4
Starch, %	49.9	44.8
GE, Mcal/kg	4.41	4.53
Calculated nutrient values		
Total RDP, %	8.80	9.05
Total soluble RDP, %	5.80	2.90
RUP, %	5.00	10.44
RDP, % of CP	63.7	46.4
Soluble protein, % of CP	42.0	14.9
MP, %	9.63	14.1
ME, Mcal/kg	3.22	3.22
NEm, ^f Mcal/kg	2.20	2.19
NEg, ^g Mcal/kg	1.51	1.51

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bCargill Sweet Bran, Dalhart, TX.

^cCorn gluten meal (69.9% CP; DM basis).

^dSupplement formulated to contain 19.83% urea (0.99% of the diet) and 33 mg of monensin/kg of DM (Elanco Animal Health, Greenfield, IN) and 8.7 mg of tylosin/kg DM (Elanco Animal Health) and vitamins and minerals to meet or exceed the [NRC \(1996\)](#) requirements in the complete mixed diet.

^eSupplement did not contain urea and was formulated to contain 33 mg of monensin/kg DM (Elanco Animal Health) and 8.7 mg of tylosin/kg DM (Elanco Animal Health) and vitamins and minerals to meet or exceed the [NRC \(1996\)](#) requirements in the complete mixed diet.

^fNet energy for maintenance.

^gNet energy for gain.

experiment) to assure that chambers were equally represented in each treatment. The sampling periods for the two groups of steers were staggered by 8 d to accommodate the two groups entering the four available respiratory chambers. Steers were individually weighed before feeding (Trojan Livestock Equipment, Weatherford, TX; set on four electronic weight bars [Tru-Test Inc., Mineral Wells, TX]; readability of ± 0.45 kg; scale calibrated with 454 kg of certified weights before use) at the start and end of each period.

Steers were fed once daily at 0800 h except during the fasting period. All steers (fed and fasted) had ad libitum access to fresh water. Diets were mixed in a mixer wagon (Roto-Mix IV 84–8, Roto-Mix, Dodge City, KS) mounted on load cells (Digi-Star, Fort Atkinson, WI, readability ± 0.45 kg) and were stored in a walk-in refrigerator in 132-L plastic barrels. Before mixing, each ingredient was weighed on a platform scale (90-kg capacity and 0.45-kg readability; Ohaus Corp., Pine Brook, NJ). Ingredients were added to the mixer, and each diet was mixed for approximately 3 min. Between diets, the mixer was cleaned to minimize cross-contamination of diets. Feed offered to each animal was weighed daily using a platform scale (22.7-kg capacity and 0.05-kg readability; Ohaus Corp.). Individual ingredients and mixed diets were sampled weekly throughout each experiment, composited by treatment across sampling periods, and frozen for later chemical analyses.

A consistent DMI was maintained during both adaptation and sampling periods of the experiments. In Exp. 1 (1 × maintenance intake), all feed was consumed during a short amount of time, and during Exp. 2 (2 × maintenance intake), steers were managed to leave only crumbs in the feed bunk the following morning. Therefore, no orts were observed or included in the dietary nutrient composition analyses or subtracted from DMI.

Digestion Collections and Gas Exchange Measurements

The fecal and urine collection methods and gas exchange measurement methods used were similar to those reported by [Hales et al. \(2012, 2013\)](#) and [Shreck et al. \(2017\)](#). Briefly, total urine output was collected daily by vacuum aspiration from a preacidified pan HCl solution (150 mL of a 20% vol/vol) to ensure that the pH of urine was less than 6 to prevent N volatilization losses. Total fecal output was collected from each steer using a nylon bag with a harness as described by [Tolleson and Erlinger \(1989\)](#). Diets, orts, urine, and feces were weighed daily on a top-loading analytical balance (1.0-g readability; Sartorius L2200, Data Weighing Systems Inc., Elk Grove, IL). Aliquots (10%) of urine and feces were collected daily and stored at 4 °C until completion of the collection period. After the completion of the 4-d collection period, the daily aliquots from each steer were thoroughly mixed, and subsamples were stored in plastic bags (feces) or polyethylene bottles (urine) at -5 °C for subsequent analyses.

Before gas measurements, the gas sampling system (described by Hales et al., 2012, 2013; Shreck et al., 2017) was calibrated for O₂ consumption and CO₂ production by burning propane as recommended by the manufacturer (Lighton, 2008). Oxygen and CO₂ recoveries were determined for each chamber and averaged 99.2% for O₂ and 99.4% for CO₂. Daily, before each gas exchange collection period, each gas analyzer (CO₂, CH₄, and O₂) was calibrated with commercially prepared gas standards (Matheson Tri-Gas, Pasadena, TX). Purity of gas standards was determined using gas chromatography (Shreck et al., 2017).

Respiration chambers were sealed approximately 23 h each day. The remaining hour was used for feeding and collection of feces, orts, and urine. Chambers were maintained between 20 and 21 °C, and relative humidity was maintained at approximately 35%. Four 23-h runs were used to determine gaseous exchanges for each animal, and HP was determined from O₂ consumption, CO₂ and CH₄ production, and urinary N excretion using the equation of Brouwer (1965) as follows:

$$\text{HP} = (3.866 \times \text{O}_2) + (1.2 \times \text{CO}_2) - (0.518 \times \text{CH}_4) - (1.413 \times \text{UN}),$$

where HP = heat production (Kcal/d), O₂ = oxygen consumption (L/d), CO₂ = carbon dioxide production (L/d), CH₄ = methane production (L/d), and UN = urinary N excretion (g/d). In addition, HP was also determined using the Brouwer (1965) equation without the correction for urinary nitrogen as suggested by Reed et al. (2017). Twenty-three-hour gas production was corrected to 24 h by dividing liters of gas consumed or produced by the exact hours recorded, then multiplied by 24. Similarly, because the animals were out of the chamber for 1 h each day, urinary excretion was corrected to 24 h.

Fasting Heat Production

Steers were fasted for 4 d with ad libitum access to fresh water. Gas exchange and urine and feces excretion (and chemical composition) were determined during the last 2 d of the fasting period in the same respiration chamber they occupied during the feeding stage of the period. FHP was calculated using the equation of Brouwer (1965) with and without the urinary N correction.

Laboratory Analysis

Dietary ingredients and ort samples were dried at 55 °C for 48 h, and fecal aliquots were lyophilized (Labconco, Kansas City, MO) before laboratory analyses. Diet, ort, and fecal samples were ground to pass through a 1-mm screen using a Wiley mill (Model 4, Thomas Scientific, Swedesboro, NJ), and a subsample of this was ground through a Cyclotec mill (Cyclotec CT 193, Foss, Hoganas, Sweden) to pass through a 0.5-mm screen for starch analysis. Laboratory DM of diet, ort, and fecal samples was determined by drying at 100 °C for 24 h, and organic matter was determined by ashing samples at 500 °C for 6 h. Ether extract (EE) of diets, orts, and feces was determined using petroleum ether in an automated EE extraction system (Ankom XT15 Extraction System, Ankom Technology, Fairport, NY). Concentrations of neutral detergent fiber (NDF) (using heat-stable α-amylase and sodium sulfite) and acid detergent fiber were determined using a semiautomated fiber analyzer (Model 2000, Ankom Technology). Gross energy (GE) of ingredients, diets, orts, and feces was determined using a bomb calorimeter (Model 6400, Parr Instrument Company, Moline, IL). Nitrogen and carbon (C) contents of urine were determined using a total N and total C analyzer with an ASI-L autosampler (Shimadzu Corp., Kyoto, Japan). Nitrogen and C contents of diets, ingredients, orts, and feces were determined using a C and N Analyzer (Elementar VarioMax, Americas Inc., Mt. Laurel, NJ). Starch content of diets, orts, and feces was determined using a PowerWave-XS Spectrometer (BioTek US, Winooski, VT) after converting starch to glucose with an enzyme kit (Megazyme International Ireland Ltd., Wicklow, Ireland).

Calculations

MP concentration of the diets was calculated using the equation of NRC (1996):

$$\text{MP} (\%) = (\% \text{RDP} \times 0.64) + (\% \text{RUP} \times 0.80)$$

in which RUP was rumen undegradable protein assuming all RDP was converted to microbial CP.

Urine GE excretion was calculated from urine C excretion using the equation of Blaxter et al. (1966). This equation was used because the caloric value per gram of C in different urinary components (i.e., urea, hippuric acid, etc.) is similar (9.2 to 12.6 Kcal/g), whereas the caloric value per gram of N is highly variable (5.4 to 72.3 Kcal/g; Blaxter

et al., 1966). In addition, the caloric value per gram of N excretion also increases with increased feed intake and increased dietary roughage inclusion (Blaxter, 1962). Urinary energy loss equations were as follows:

$$\text{Urinary energy loss (fed state), Kcal} = \\ ((10.33 \times \text{urinary C excreted, g}) - 38) / 1000;$$

$$\text{Urinary energy loss (fasted state), Kcal} = \\ ((10.04 \times \text{urinary C excreted, g}) - 3) / 1000.$$

Dietary MEI was calculated as GE intake minus energy in feces, urine, and enteric methane. Retained energy was calculated as ME minus HP. The proportion of ME lost as heat and/or retained was used as measures of the efficiency of energy utilization. FHP was considered the maintenance net energy requirement for each steer.

Fat retained was calculated using C–N balance from the noncarbon protein retained (Maynard and Loosli, 1969) as follows:

$$\text{C retained (in protein), g} = \\ \text{Protein retained, g} \times 0.5254,$$

$$\text{C retained, g} = \text{C intake, g} - \\ (\text{C in feces, g} + \text{C in urine, g} + \\ \text{C in CO}_2, \text{ g} + \text{C in CH}_4, \text{ g}),$$

$$\text{C retained (nonprotein), g} = \\ \text{C retained, g} - \text{C retained (in protein), g},$$

$$\text{Fat retained, Mcal} = \\ \text{C retained (nonprotein), g} / 0.765.$$

The partial efficiency of utilization of ME for maintenance (k_m) and gain (k_g) was determined by regressing energy loss or gain per unit of MBW ($\text{kg}^{0.75}$) against ME intake per unit of MBW of cattle fed at fasting, 1 \times maintenance and 2 \times maintenance. The slope of the linear regression line below maintenance was equivalent to k_m , and the slope of the linear regression line above maintenance was equivalent to k_g . The maintenance energy requirements were determined from the linear regression lines above and below maintenance, as well as quadratic equations that were developed using all data from both experiments.

Blood Sampling

A blood sample was collected at 0700 h (before feeding) from each individual steer at the middle

(day 14; 14 days from the beginning of adaptation) and end (day 19; upon entry of respiratory chambers) during each period. Blood was collected into tubes via jugular venipuncture, allowed to clot at ambient temperature for 1 h and then centrifuged at 4 °C for 30 min at 1,100 \times g. Serum was separated into 1-mL aliquots for subsequent analyses of serum urea nitrogen (SUN) and nonesterified fatty acid (NEFA). SUN concentration was determined via colorimetric assay using a commercial kit (urea nitrogen procedure No. 0580; Stanbio Laboratories, Boerne, TX) using a 96-well microplate and read at 520 nm using a microplate reader (BioTek US). NEFA concentration was determined via a colorimetric assay using a commercial kit (Wako Diagnostics, Mountain View, CA) using a 96-well microplate and read at 550 nm using a microplate reader (BioTek US).

Statistical Analysis

Digestibility and energetic data in each trial were statistically analyzed as a switchback design with three periods using the PROC MIXED procedures of SAS (SAS Inst. Inc., Cary, NC). Degrees of freedom were approximated using the Kenward–Roger method (ddfm=kr). Random effects were steer/chamber, period, and group, whereas diet was included as a fixed effect in the model. SUN and NEFA data were analyzed using the PROC MIXED of SAS (SAS Inst. Inc., Cary, NC) as a repeated measures (Tempelman, 2004) with steer \times diet as the subject and a compound symmetry covariant structure (type=CS). Random effects were steer/chamber, period, and steer \times diet, whereas day, diet, and day \times diet were included as fixed effects in the model. Effects were considered significant at $P \leq 0.05$ with tendencies at $0.05 < P \leq 0.10$.

RESULTS AND DISCUSSION

Experiments 1 and 2 were not statistically analyzed together as time was a confounding factor. However, numerical differences and differences/similarities in trends between experiments may be discussed.

Ingredient and Diet Nutrient Composition

Analyzed compositions of the dietary treatments are presented in Table 1. To achieve excess dietary CP and MP, in the ECP diet, corn gluten meal (69.9% CP, DM basis) replaced a portion of SFC, corn gluten feed (Cargill Sweet Bran, Dalhart,

TX), and all the urea and corn oil in the CON diet. Thus, the ECP diet had greater RUP (10.4%; as a proportion of DM) than the CON diet (5.0%; as a proportion of DM). The CON and ECP diets had similar RDP concentrations; however, as a proportion of total CP, the CON diet had greater RDP (63.7%) than the ECP diet (46.4%), because in part of inclusion of 0.99% urea as a portion of diet DM in the CON diet. The RDP concentration in both diets was adequate for cattle fed SFC-based finishing diets (Galyean, 1996; Cooper et al., 2002). GE was slightly greater in ECP than in CON; however, as designed, calculated ME and NE values were similar for both diets. EE and starch concentration were greater and MP concentration was less in CON than ECP. Dietary NDF concentrations were similar for ECP and CON diets.

DM and MP Intake and BW

Within each experiment, DMI and BW were similar for CON and ECP steers (Tables 2 and 3, respectively). By design, in Exp. 1, BW did not change (262.5 vs. 260.1 ± 1.4 kg for initial and final BW, respectively), whereas in Exp. 2, cattle gained, on average, 1 kg/d throughout the experiment (excluding the fasting periods). The estimated MP requirement for maintenance in Exp. 1 was 257 g/d, and MP intakes averaged 221 and 325 g/d for the CON and ECP steers, respectively. The estimated MP requirement for maintenance in Exp. 2 was 324 g/d, and the MP requirement for gain was 298 g/d for a total MP requirement of 622 g/d (NRC, 1996). MP intake for CON averaged 596 g/d and for ECP averaged 875 g/d. Thus, in both experiments, the CON steers had MP intakes at or slightly below requirements, and ECP steers had excess MP intakes of 68 and 253 g/d in Exp. 1 and 2, respectively. Therefore, calculated excess N intakes were approximately 10.9 and 40.5 g/d in ECP steers in Exp. 1 and 2, respectively.

Nutrient Digestibility

Digestibility of DM, starch, NDF, and EE at 1 × maintenance and 2 × maintenance levels of intake is presented in Tables 2 and 3, respectively. Apparent DM digestibility tended ($P = 0.07$) to be lower in the CON steers than that in ECP steers at 1 × maintenance intake, whereas no differences were observed at 2 × maintenance intake. The apparent DM digestibility's observed in this study were similar to those reported by May et al. (2009) with steers having ad libitum access to SFC-based,

15% CP (DM basis) finishing diets using dried corn distiller's grains with solubles or soybean meal (SBM) as the protein source. Archibeque et al. (2007) reported that steers consuming diets with 12% or 14% CP (DM basis) or consuming diets that oscillated between 9% and 14% CP (DM) also had DM digestibility similar to that observed in the present experiments. However, Archibeque et al. (2007) reported that steers continuously fed a 9% CP (DM basis) diet had a lower DM digestibility than steers fed 12% or 14% CP (DM basis) diets when SBM was the supplemental protein source.

Apparent starch digestion exceeded 99% in both treatments and experiments. At both 1 × and 2 × maintenance intake, NDF apparent digestibility was lower ($P = 0.03$) in the CON steers than in the ECP steers. This could possibly be due to greater digestibility of NDF in corn gluten meal than in SFC or to the greater fat content of the CON diet. In contrast, EE apparent digestibility (uncorrected for fecal calcium soaps) was greater ($P < 0.01$) in the CON steers than in the ECP steers at both intake levels. Hales et al. (2013) reported decreasing apparent NDF digestibility as CP increased from 13.5% to 20% of the diet (DM basis) because of increasing WDGS in SFC-based finishing diets, whereas they noted no effect of CP or WDGS concentration on apparent EE digestibility. The apparent DM, starch, NDF, and EE digestibilities were numerically similar across experiments at 1 × and 2 × maintenance intake. This, to some extent, contrasts with Reynolds et al. (1991a) in which heifers fed a 75% concentrate diet at a high intake had numerically lower DM, EE, and NDF digestibility than heifers fed at a low intake.

Nitrogen Balance

Nitrogen balance for 1 × maintenance and 2 × maintenance intake is presented in Tables 4 and 5, respectively. By design, intake of N (g/d) was greater ($P < 0.01$) in the ECP steers than in the CON steers, at both levels of intake. At both 1 × and 2 × maintenance intake, fecal N excretion was not affected by diet ($P \geq 0.17$); however, urinary N excretion (g/d) was greater ($P < 0.01$) in the ECP steers than in the CON steers. The lack of a diet effect on fecal N excretion (g/d) tends to contrast with previous research (Cole et al., 2006; Archibeque et al., 2007; Vasconcelos et al., 2009; Hales et al., 2013). In Exp. 1 and 2, respectively, 68% and 55% of the increased N intake (i.e., ECP vs. CON) was excreted in the urine. Blaxter and Martin (1962) reported that 83% to 86% of casein infused into the rumen or

Table 2. Effects of excess dietary CP on nutrient intake, excretion, and digestibility in steers fed at 1 × maintenance energy intake

Item	Dietary treatment ^a			
	CON	ECP	SEM ^b	P value
Observations, <i>n</i>	12	12	—	—
Initial BW, kg	263	262	1.3	0.30
Final BW, kg	260	260	1.5	0.85
DM				
Intake, g/d	2,294	2,300	1.3	<0.01
Fecal excretion, g/d	455	397	34.4	0.11
Apparent digestibility, % of intake	80.0	82.8	1.5	0.07
Starch				
Intake, g/d	1,110	1,000	1.3	<0.01
Fecal excretion, g/d	1.7	1.4	0.1	0.09
Apparent digestibility, % of intake	99.8	99.8	0.01	0.51
NDF				
Intake, g/d	436.8	441.6	0.3	<0.01
Fecal excretion, g/d	219.2	183.7	18.2	0.06
Apparent digestibility, % of intake	49.4	58.6	4.0	0.03
EE				
Intake, g/d	73.2	48.4	0.4	<0.01
Fecal excretion, g/d	16.3	15.9	1.6	0.79
Apparent digestibility, % of intake	77.7	67.1	2.9	<0.01

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

Table 3. Effects of excess dietary CP on nutrient intake, excretion, and digestibility in steers fed at 2 × maintenance energy intake

Item	Dietary treatment ^a			
	CON	ECP	SEM ^b	P value
Observations, <i>n</i>	12	12	—	—
Initial BW, kg	375	373	2.3	0.34
Final BW, kg	407	409	2.0	0.34
DM				
Intake, g/d	6,145.2	6,230.6	41.5	0.06
Fecal excretion, g/d	1,070.4	1,011.9	43.0	0.20
Apparent digestibility, % of intake	82.5	83.7	0.7	0.11
Starch				
Intake, g/d	3,156.9	2,875.3	20.8	<0.01
Fecal excretion, g/d	6.1	5.4	0.6	0.30
Apparent digestibility, % of intake	99.8	99.8	0.02	0.87
NDF				
Intake, g/d	1,199.2	1,222.7	8.6	0.02
Fecal excretion, g/d	503.4	462.6	21.8	0.08
Apparent digestibility, % of intake	57.8	62.2	1.8	0.03
EE				
Intake, g/d	199.5	138.6	1.3	<0.01
Fecal excretion, g/d	32.6	48.3	4.3	<0.01
Apparent digestibility, % of intake	83.6	65.0	2.9	<0.01

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

abomasum was excreted in the urine. In this study, most of the excess CP fed would be in the form of RUP and equate to an abomasal infusion of CP. The apparent postruminal digestibility of casein

should approach 100% (Blaxter and Martin, 1962), whereas postruminal degradability of RUP in corn gluten meal would most likely not exceed 90% (NRC, 2001). Hales et al. (2013) reported similar

Table 4. Effects of excess dietary CP on nitrogen (N) balance in steers fed at 1 × maintenance intake

Item	Dietary treatment ^a			P value
	CON	ECP	SEM ^b	
N intake, g/d	50.1	69.3	0.3	<0.01
N excretion, g/d				
Urine	35.9	48.9	2.5	<0.01
Feces	12.0	11.4	0.7	0.42
N digestibility, %	76.0	83.6	1.1	<0.01
Digestible N intake, g/d	37.9	58.1	0.8	<0.01
N retained				
Total g/d	2.0	9.3	2.4	<0.01
% of digested N	5.3	16.5	4.1	0.02
Urine N, % of digested N	94.7	83.6	4.1	0.02

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

Table 5. Effects of excess dietary CP on nitrogen (N) balance in steers fed at 2 × maintenance intake

Item	Dietary treatment ^a			P value
	CON	ECP	SEM ^b	
N intake, g/d	130.8	192.0	1.7	<0.01
N excretion, g/d				
Urine	64.2	97.8	3.7	<0.01
Feces	31.8	34.0	1.6	0.17
N digestibility, %	75.6	82.2	0.9	<0.01
Digestible N intake, g/d	99.3	157.7	1.4	<0.01
N retained				
Total g/d	35.1	60.0	3.3	<0.01
% of digested N	35.4	38.5	2.5	0.24
Urine N, % of digested N	64.6	61.5	2.5	0.24

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

urinary N excretion results, although they noted that fecal N excretion increased as CP of diets fed a 2 × maintenance increased from 13% to 20% and WDGS concentration increased from 0% to 45% of diet DM. The differences in fecal N excretion and N digestibility between our study and Hales et al. (2013) could be explained by the digestibility of the RUP in the by-product ingredients used by Hales et al. (2013) compared with our study. WDGS and wet corn gluten feed have a similar CP digestibility; however, the RUP fraction of WDGS is more digestible than wet corn gluten feed (Kelzer et al., 2010). The postruminal digestibility of the RUP in corn gluten meal (92%) is estimated to be greater than both corn gluten feed (85%) and distillers grains (80%; NRC, 2001). Archibeque et al. (2007) observed an increase in urinary N excretion of steers as dietary CP increased from 9% to 14% CP (DM basis) and steers were fed at ad libitum intake. Cole et al. (2006) performed a study using

cattle fed diets containing 10%, 11.5%, or 13.0% CP (DM basis) from finishing diets with cotton seed meal (CSM) and(or) urea as the supplemental protein sources. Urinary N excretion increased as dietary CP (DM basis) increased throughout the feeding period (Cole et al., 2006). However, fecal N excretion did not differ between treatments until the last 56 d of the feeding period, where fecal N excretion was greatest in steers consuming 13% CP (DM basis; Cole et al., 2006).

Nitrogen digestibility and digestible N intake (g/d) were greater ($P < 0.01$) in the ECP steers than in the CON steers at both intake levels. Similarly, steers consuming diets containing 11.5%, 13%, and 14.5% CP (DM basis) at ad libitum intake, with various levels of CSM or urea, had increasing apparent N absorption as dietary CP increased (Vasconcelos et al., 2009). The lack of a dietary protein effect on fecal N excretion and a dietary effect on protein digestibility may be due in part to differences in metabolic fecal N excretion. If metabolic fecal N is estimated from indigestible DMI (i.e., fecal DM excretion) using the conversion factor of 0.068 g of metabolic fecal N/g of fecal DM (NRC, 1985), estimated true protein digestibilities were 86.0% and 89.7% for CON and ECP, respectively, in Exp. 1 and 82.5 and 85.3% for CON and ECP, respectively, in Exp. 2.

Total N retained (g/d) was greater ($P < 0.01$) in the ECP steers than in the CON steers at both intake levels. Nitrogen retained as a proportion of digested N was also greater ($P = 0.02$) in the ECP steers than in the CON steers; therefore, urinary N excreted as proportion of digested N was greater ($P = 0.02$) in CON steers than in ECP steers at 1 × maintenance intake. There were no differences ($P = 0.24$) in N retained or urinary N excreted as a proportion of digested N in steers fed at 2 × maintenance.

The greater total N retention in the ECP steers than the CON steers could be because the CON steers had not yet met their MP requirement and the ECP steers retained more N because it was available. Hales et al. (2013), Cole et al. (2006), and Vasconcelos et al. (2009) reported no differences in N retention as dietary CP increased from approximately 9.5% to 20.0%, whereas Archibeque et al. (2007) observed an increase in N retention in steers as dietary CP increased from 10% to 12% CP (DM basis) although N retention plateaued at 14% CP (DM basis) and then fell below that retained by steers receiving the 12% CP (DM basis) treatment diets at ad libitum intake (Archibeque et al., 2007). The 1 × maintenance intake during the first

experiment limited weight gain and delayed growth; therefore, mature BW to reach a common empty body composition could have increased, modifying their growth curve and resulting in an increase in MP requirements going into the second experiment. This could explain the greater level of N retention in the ECP steers during 2 × maintenance intake.

Many studies have looked at the effect of ruminal and postruminal infusions of casein on N and energy metabolism of the whole body, portal drain viscera, and liver of sheep and cattle. In this study, the excess protein was primarily provided by RUP in corn gluten meal. Although corn gluten meal has a postruminal digestibility similar to casein (NRC, 2001), it has an appreciably lower protein quality than casein. It is not known if a potential postruminal amino acid (AA) deficiency or imbalance affected our results. However, if microbial CP did not correct an amino acid deficiency, the excess protein provided in corn gluten meal still may have provided postruminal amino acids that were deficient in the CON steers and could account for the greater N retention in the ECP steers than the CON steers.

Gaseous Exchange

Daily O₂ consumption and CO₂ and CH₄ production at 1 × and 2 × maintenance levels of intake are reported in Tables 6 and 7, respectively. There were no differences ($P = 0.91$) in O₂ consumption and CO₂ production between the CON and ECP steers in Exp. 1 when steers were fed at 1 × maintenance intake. Similar results were reported by Hales et al. (2012, 2013) where CO₂ production was similar regardless of dietary CP content, WDGS inclusion rate, or corn-processing method (dry rolled vs. steam flaked), and diets were fed at 2 × maintenance intake. In contrast, Archibeque et al. (2007) reported greater CO₂ production in steers consuming 12% and 14% CP (DM basis) diets than a 9% CP diet, with differences being attributed to DMI. In Exp. 2, total consumption of O₂ was 6.5% greater ($P = 0.04$) in the ECP steers than in the CON steers, and the respiratory quotient (RQ) in the ECP steers was lower ($P < 0.01$) than in the CON steers. A lower RQ would be expected with an increase in metabolism of protein at the expense of carbohydrate metabolism. In addition, O₂ consumption per kg of DMI tended to be 5.4% greater ($P < 0.06$) in ECP steers than in CON steers. One source of CO₂ production is from anaerobic fermentation in the gut. This may be of importance in our study because ruminal fermentation of corn

Table 6. Daily oxygen (O₂) consumption and methane (CH₄) and carbon dioxide (CO₂) production influenced by excess dietary CP in steers fed at 1 × maintenance intake.

Item	Dietary treatment ^a			P value
	CON	ECP	SEM ^b	
Intake				
DM basis, kg/d	2.29	2.30	0.001	0.01
% of BW	0.88	0.88	0.005	0.99
% of MBW	3.55	3.55	0.02	0.81
O ₂ consumption, L/steer	1,329.8	1,333.0	27.9	0.91
O ₂ consumption, L/kg DMI	578.9	578.4	12.2	0.97
CO ₂ production, L/steer	1,349.6	1,353.6	34.9	0.91
RQ ^c	1.03	1.02	0.01	0.70
CH ₄ production				
L/steer	66.9	70.0	3.4	0.38
% of GE	6.4	6.4	0.3	0.89
% of DE	7.6	7.5	0.4	0.75

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

^cRQ = respiratory quotient.

Table 7. Daily oxygen (O₂) consumption and methane (CH₄) and carbon dioxide (CO₂) production influenced by excess dietary protein in steers fed at 2 × maintenance intake

Item	Dietary treatment ^a			P value
	CON	ECP	SEM ^b	
Intake				
DM basis, kg/d	6.15	6.23	0.04	0.06
% of BW	1.51	1.52	0.01	0.36
% of MBW	6.79	6.84	0.05	0.30
O ₂ consumption, L/steer	2,647.2	2,819.5	75.4	0.04
O ₂ consumption, L/kg DMI	432.2	455.5	11.5	0.06
CO ₂ production, L/steer	2,732.6	2,800.5	84.6	0.43
RQ ^c	1.03	0.99	0.01	0.01
CH ₄ production				
L/steer	113.8	105.5	9.5	0.39
% of GE	4.0	3.5	0.3	0.21
% of DE	4.7	4.1	0.4	0.14

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

^cRQ = respiratory quotient.

gluten meal would be much less than SFC, which is another possible reason for the slightly lower RQ for the ECP steers than for the CON steers in Exp. 2.

Reynolds et al. (1991a, 1991b) noted that the portal drain viscera and liver accounted for 46% of whole body O₂ consumption and urea synthesis accounted for 13% to 16% of hepatic O₂

consumption. Reynolds et al. (1991b) also noted that the synthesis of urea-N from ammonia-N is an energetically expensive process. Milano et al. (2000) reported that infusion of NH_4HCO_3 increased hepatic O_2 consumption by 25% and that 54% of the increase in hepatic O_2 consumption was due to hepatic ureagenesis. However, if urea nitrogen is derived from AA without the involvement of glutamate dehydrogenase, the cost of urea synthesis can be considerably lower (Reynolds et al., 1991a, 1991b).

Ferrell et al. (2001) reported that O_2 uptake of the portal drain viscera of sheep fed a high-concentrate diet was not affected by dietary protein concentration (6.6 vs. 11.2% CP, DM basis) or supplemental protein source/ruminal degradability (urea, SBM, or blood meal). However, protein concentration and source significantly affected hepatic O_2 uptake. Hepatic O_2 uptake was significantly less when the supplement had a high RDP (urea) than a high RUP (blood meal). Heat production of the liver accounted for 19.4%, 20.1%, 23.2%, and 26.2% of total HP with control, urea, SBM, and blood meal diets (Ferrell et al., 2001). Furthermore, net O_2 uptake was 483, 502, 621, and 648 kg/d (or a 34% increase) in sheep fed a control diet or diets supplemented with urea, SBM, and blood meal, respectively. Thus, the effect of protein supplementation on total body HP was approximately 6.6% to 8.9%, which is similar to the 6.5% increase in O_2 consumption we noted in the 2 × maintenance study.

Bruckental et al. (1997) reported a nonsignificant 17% increase in hepatic O_2 uptake of steers abomasally infused with casein and fed a protein deficient high-concentrate diet. Similarly, Guerin et al. (1991) reported a 9% to 11% increase in hepatic and splanchnic O_2 flux in steers abomasally infused with casein. Therefore, numerous studies have shown that abomasal infusion of casein or feeding protein supplements high in RUP increase O_2 consumption by the liver, most likely because of an increase in ureagenesis.

No differences ($P \geq 0.38$) were observed in CH_4 production between CON and ECP steers at both intake levels. As would be expected, because of greater DMI, CH_4 production (L/d) was greater in Exp. 2 than in Exp. 1, whereas methane yield as a percent of GE intake or as a percent of DE intake was greater in steers fed at 1 × maintenance than in steers fed at 2 × maintenance (NASEM, 2016). Archibeque et al. (2007) also noted no effect of dietary CP on CH_4 production; however, Hales et al. (2013) observed a linear increase in CH_4 production (L/steer) as dietary WDGS and CP content

increased. However, this increase in CH_4 was attributable to an increase in NDF content, rather than an increase in CP content of the diets.

Energy Losses/Utilization

GE intake was greater ($P < 0.01$) in the ECP steers than in the CON steers in both experiments (Tables 8 and 9, respectively) because of greater GE content of the ECP diet. At both intake levels, there were no differences ($P \geq 0.12$) in daily fecal energy loss (Mcal/d) between treatments; however, CON steers tended to have greater ($P \leq 0.06$) fecal energy loss as a proportion of GE than the ECP steers. At 1 × maintenance intake, DE as a proportion of GE was greater ($P \leq 0.04$) in the ECP steers than in the CON steers, and at 2 × maintenance intake, DE as a proportion of GE tended to be greater ($P = 0.06$) in the ECP steers than in the CON steers. In both experiments, ECP steers had greater ($P < 0.01$) DE intakes (Mcal/d) than CON steers.

Fecal energy losses as a percent of GE intake were lower, and DE values as a percent of GE were higher than values noted by Reynolds et al. (1991) and by Ferrell et al. (2001) who fed diets based on cracked or dry-rolled corn. However, values in this study were similar to other studies in which the diet was based on SFC (Hales et al., 2012, 2013). This was most likely due to the greater starch and energy digestibility of SFC than dry-rolled corn (Hales et al., 2012).

The ECP steers had greater ($P < 0.01$) daily urinary energy loss (Mcal/d), and urinary energy loss as a proportion of GE ($P < 0.01$), than CON steers at 1 × and 2 × maintenance intake. The urinary energy losses as a percent of GE for CON steers were similar to values of Reynolds et al. (1991a), which ranged from 3.8% to 4%. Greater urinary energy loss was expected with the ECP diet because a primary energetic constituent in urine is urea, which probably increased with increased dietary CP/MP content (Blaxter and Martin, 1962). Blaxter and Martin (1962) noted that urinary energy losses increased 15.3 Kcal per 100 Kcal of casein infused. Both Archibeque et al. (2007) and Hales et al. (2013) reported increases in urinary energy losses as dietary CP increased in steers consuming diets at ad libitum intake and at 2 × maintenance, respectively. Total methane production (Mcal/d) and methane production as a percent of GE were not significantly affected ($P > 0.21$) by treatment diet. These values are similar to other studies (Reynolds et al., 1991a; Hales et al., 2012, 2013).

At both 1 × and 2 × maintenance intake, ME intake (Mcal/d) was greater ($P < 0.01$) in ECP steers than in CON steers. ME as a proportion of GE intake was not significantly different ($P = 0.40$) across treatments in Exp. 1, but in Exp. 2, ME/GE tended ($P < 0.08$) to be greater in ECP steers than in CON steers. Blaxter and Martin (1962) noted that the ME of casein infused into the abomasum was 87.3% of GE, whereas when infused into the rumen, the ME of casein was 62.5% of GE. Thus, the feeding of a supplement high in digestible RUP could be expected to increase dietary ME. The greater ME of the ECP diet can also partially be explained by the greater GE content of the ECP diet. In steers fed at 1 × maintenance, ME as a percent of DE was greater ($P < 0.02$) in CON than in ECP steers; however, when fed at 2 × maintenance, there was no dietary effect ($P = 0.60$) on ME as a percent of DE. The ME:DE in this study is higher than the customarily used value of 0.82 (NRC, 1996; NASEM, 2016) but is similar to values reported by Reynolds et al. (1991) and Hales et al. (2012, 2013) with high-concentrate diets. The recent review by Galyean et al. (2016) reported that ME:DE is higher in high-concentrate diets than high-forage diets.

The heat of fermentation within the gut can account for 5% to 10% of GE in ruminants (Blaxter, 1962). Blaxter (1962) used a “corrected ME” that was adjusted for the heat of fermentation in the gut. In that equation, the heat of combustion of methane was multiplied by 1.8 to correct for the heat of fermentation in the gut. Using the methane correction factor decreased the ME values in Exp. 1 and 2 but did not significantly change the magnitude of difference in ME between CON and ECP diets. The uncorrected ME values were 3.21, 3.32, 3.41, and 3.57 Mcal/kg for 1 × maintenance CON, 1 × maintenance ECP, 2 × maintenance CON, and 2 × maintenance ECP, respectively. The ME values corrected for heat of fermentation were 2.99, 3.08, 3.28, and 3.43 Mcal/kg, respectively. The ECP steers fed at the 1 × maintenance level of intake could have been partitioning excess protein as an energy substrate for greater RE than CON steers, whereas RE was similar for both diets at 2 × maintenance.

Heat production (Mcal/d and percent of GE intake) was not affected ($P > 0.18$) by dietary CP/MP concentration at 1 × maintenance intake. In contrast, at 2 × maintenance intake, heat production (Mcal/d) tended ($P = 0.09$) to be greater in ECP than in CON steers, although heat production as a proportion of GE intake was not affected by diet. At 1 × maintenance, heat production as a

Table 8. Effects of excess dietary CP fed at 1 × maintenance intake on energy intake and losses of steers

Item	Dietary treatment ^a			
	CON	ECP	SEM ^b	<i>P</i> value
GE, Mcal/d	9.92	10.23	0.001	<0.01
Fecal energy, Mcal/d	1.52	1.37	0.09	0.12
Fecal energy, % of GE	15.40	13.36	0.89	0.04
DE, Mcal/d	8.38	8.87	0.09	<0.01
DE, % of GE	84.60	86.64	0.89	0.04
Urinary energy, Mcal/d	0.42	0.57	0.03	<0.01
Urinary energy, % of GE	4.25	5.57	0.29	<0.01
CH ₄ , Mcal/d	0.63	0.66	0.03	0.38
CH ₄ , % of GE	6.39	6.44	0.32	0.89
ME, Mcal/kg of DM	3.21	3.33	0.04	<0.01
ME, Mcal/d	7.32	7.65	0.09	<0.01
ME, % of GE	73.94	74.65	0.88	0.40
ME, % of DE	87.36	86.15	0.42	0.02
Heat production, Mcal/d	6.67	6.67	0.14	0.99
Heat production, % of GE	67.17	65.13	1.43	0.18
Heat production, % of ME	91.86	88.22	1.90	0.06
Retained energy, % of ME	8.14	11.78	1.91	0.08

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

Table 9. Effects of excess dietary CP fed at 2 × maintenance intake on energy intake and losses of steers

Item	Dietary treatment ^a			
	CON	ECP	SEM ^b	<i>P</i> value
GE, Mcal/d	27.10	28.20	0.19	<0.01
Fecal energy, Mcal/d	4.19	3.98	0.19	0.27
Fecal energy, % of GE	15.54	14.12	0.69	0.06
DE, Mcal/d	22.94	24.19	0.22	<0.01
DE, % of GE	84.46	85.88	0.69	0.06
Urinary energy, Mcal/d	0.82	1.07	0.03	<0.01
Urinary energy, % of GE	3.01	3.79	0.13	<0.01
CH ₄ , Mcal/d	1.07	0.99	0.09	0.39
CH ₄ , % of GE	3.98	3.53	0.34	0.21
ME, Mcal/kg of DM	3.41	3.56	0.03	<0.01
ME, Mcal/d	21.05	22.13	0.22	<0.01
ME, % of GE	77.40	78.63	0.67	0.08
ME, % of DE	91.73	91.48	0.45	0.60
Heat production, Mcal/d	13.36	14.06	0.39	0.09
Heat production, % of GE	49.45	50.16	1.32	0.60
Heat production, % of ME	64.01	63.87	1.79	0.93
Retained energy, % of ME	35.99	36.13	1.79	0.98
Retained energy as protein, %	14.8	23.0	1.23	<0.01
Retained energy as fat, %	85.2	77.0	1.23	<0.01

^aCON = control (13.8% CP), ECP = excess CP (19.5% CP).

^bSEM = standard error of the mean.

proportion of ME intake tended ($P < 0.08$) to be greater in CON than in ECP. In contrast, at 2 × maintenance, HP as a proportion of ME intake was not affected by dietary CP/MP concentration. Our HP:GE values were greater than those reported

by Reynolds et al. (1991a), most likely because of lower digestibility of their diets compared with ours. However, as noted in the present experiments, Reynolds et al. (1991a) reported that HP as percent of GE was greater in heifers fed at low DMI than at high DMI (56% vs. 45%, respectively). The HP as percent of GE values in Exp. 2 were similar to those reported by Hales et al. (2012, 2013) who fed SFC-based diets similar to ours.

Heat production calculated with the Brouwer (1965) equation without the urinary N correction factor was slightly greater than values with the correction factor. In Exp. 1, uncorrected heat productions were 6.73 and 6.74 Mcal/d for CON and ECP, respectively, whereas heat production calculated using the urinary N correction factor was 6.67 and 6.67, respectively. Similarly, in Exp. 2, uncorrected heat productions were 13.45 and 14.21 for CON and ECP, and corrected values were 13.36 and 14.06 Mcal/d. Thus, eliminating the urinary N correction factor had little or no effect on energy metabolism values.

Hales et al. (2013) noted that HP as a percent of MEI increased (68.2%, 73.2%, 75.2%, and 82.8%) as dietary CP content increased from 13.3% to 14.3% to 18.3% and to 20.3%, although these values were somewhat confounded by changes in dietary starch and fiber content. Our hypothesis was that an increase in maintenance energy requirement caused by an excess of dietary N would be reflected by an increase in HP as a percent of MEI. However, that was not seen in the present studies. This may be due to the higher ME values of the ECP diet caused primarily by a higher GE content and higher digestibility of the energy in the ECP diet. Using a data set with over 1,100 values, Reed et al. (2017) noted that feeding greater quantities of RDP increased heat production of dairy cows by 1.03 to 1.22 Mcal/kg of RDP fed, whereas feeding extra RUP increased heat production only by 0.78 Mcal/kg RUP fed. Thus, a greater response might have been noted if we had fed excess RDP, rather than excess RUP. Reed et al. (2017) also noted that excess N intake had a significant effect on energy retention when N intakes exceeded requirements; however, when protein intakes were below requirements, increased protein intake had a positive effect on energy balance. In Exp. 1, when cattle were fed at 1 × maintenance, the excess N intake (10.9 g/d) may have been too small to elicit a measurable change in maintenance requirements. In Exp. 2, when cattle were fed at 2 × maintenance, although there was no significant difference in HP, cattle fed the ECP diet had 6% greater ($P < 0.04$) total O₂ consumption and tended ($P < 0.06$) to have a 5.4%

greater O₂ consumption per kg of DMI (432.2 vs. 455.5 ± 11.5 L/kg DMI).

In Exp. 1, daily RE as a proportion of MEI tended ($P < 0.08$) to be greater in ECP steers than in CON steers, whereas in Exp. 2, RE as a proportion of ME intake was similar ($P = 0.98$) between treatments. In contrast, Hales et al. (2013) reported that RE as a proportion of ME decreased as dietary CP increased (because of increased WDGS content) in diets fed at 2 × maintenance. Also, Archibeque et al. (2007) observed a decrease in RE (Mcal/d) as dietary CP increased from 9% to 14% of diet DM.

In Exp. 2, the proportion of energy retained as protein was greater ($P < 0.01$) for ECP steers than for CON steers, and, thus, the proportion of energy retained as fat was greater ($P < 0.01$) for CON steers than for ECP steers. These values were similar to results of Reynolds et al. (1991a) who noted that protein accounted for 12.6% to 19.7% of total RE in cattle fed high-concentrate diets containing 16% to 17% CP. The greater protein retention in ECP steers was most likely because steers on the CON diet were in a slight MP deficiency, whereas in ECP steers, MP was adequate or excessive. Because there were a number of animals that had negative protein, fat, and(or) energy retentions in Exp. 1 when fed at 1 × maintenance, it was not possible to statistically compare the proportion of energy retained as protein or fat.

Gleghorn et al. (2004) reported that average daily gain (ADG) of steers increased linearly with increasing dietary CP concentration (11.5%, 13.0%, and 14.5%, DM basis) during the first 112 d of the feeding period, because in part of increasing DMI. However, over the entire feeding period (140 to 185 d), feeding CP in excess of 13% appeared to decrease performance in steers fed finishing diets based on SFC. The optimal RDP concentration appeared to be approximately 8.2% of diet DM. The estimated MP requirements were 369 g for maintenance and 433 g for gain (total 802 g/d). Among the nine experimental diets they used, a 13% CP diet containing 1% urea (8.2% RDP and 4.8% RUP) was closest to meeting the RDP and MP requirements. In a similar study to ours, Samuelson et al. (2017) noted no effect of dietary CP concentration (20.9% vs. 13.9%, DM basis) or source (urea vs. corn gluten meal) on ADG, DMI, or gain:feed of finishing heifers during the last 35 d of the feeding period. In their study, heifers had energy intakes of approximately 2.15 × maintenance; however, estimated MP intakes were not reported. In another study, Samuelson et al. (2016) fed finishing diets that contained 0%, 0.5%, or 1.0% urea (13.7%, 14.4%, or

15.8% CP and 9.7%, 10.0%, and 10.1% MP, respectively) during the last 27 d on feed and noted a lower ADG and gain:feed in steers fed the 1.0% urea diet. Energy intakes were approximately $1.85 \times$ maintenance, and MP intakes in all treatments were 243 to 275 g greater than MP requirements (NRC, 1996), which would correspond to an excess N intake of 38.9 to 44.0, similar to the surplus we noted in Exp. 2. Hales et al. (2016) fed dry-rolled corn-based finishing diets that contained 13.5% or 17.5% CP, with the protein supplement being SBM. Steers fed the 17.5% CP diet had lower DMI but had greater gain:feed than steers fed the 13.5% CP diet, possibly because the 13.5% CP diet was calculated to be limiting in MP. Estimated energy intakes were approximately $2 \times$ maintenance. In short, a number of studies have noted variable effects of excess CP on performance of finishing beef cattle; however, the results are somewhat inconsistent because of differences in energy intake, grain processing, stage of growth, and supplemental protein source.

Fasting Heat Production

In Exp. 1, FHP of ECP and CON steers was not different ($P = 0.50$; 4.85 vs. 4.94 ± 0.13 Mcal/d) as was FHP per kg MBW ($P = 0.42$; 75.6 vs. 77.6 ± 0.2 Kcal, respectively). In Exp. 2, FHP of ECP steers tended to be greater ($P < 0.08$) than of CON steers (8.54 vs. 8.02 ± 0.3 Mcal/d, respectively), as was FHP per kg MBW ($P < 0.08$; 97.9 vs. 92.2 ± 0.34 Kcal, respectively). The FHP was greater in Exp. 2 than in Exp. 1 as would be expected as Blaxter (1962) noted that FHP varies with the level of pre-fast feed intake. As previously noted, the lack of an effect of excess protein on FHP in Exp. 1 may be because there was minimal intake of excess protein. However, the greater FHP in steers fed at $2 \times$ maintenance suggests that feeding of excess CP/MP increased the maintenance energy requirement by approximately 6.1% to 6.5%. This is similar to the 6% greater O_2 consumption and 5.4% greater O_2 consumption per kg of DMI noted in ECP steers than in CON steers in Exp. 2.

Utilization of ME

As previously noted, k_m and k_g were determined by regressing RE per kg MBW against MEI per kg MBW below and above maintenance, respectively. The linear regression lines below maintenance were as follows:

$$\text{for CON; RE} = 0.8544\text{MEI} - 83.74; R^2 = 0.97;$$

$$\text{for ECP; RE} = 0.8456\text{MEI} - 86.78; R^2 = 0.94,$$

where RE = retained energy (Kcal/kg MBW daily) and MEI = ME intake (Kcal/kg MBW daily). Thus, the k_m for CON steers (0.854) was similar to the k_m for ECP steers (0.845). These values were slightly greater than k_m values reported by Blaxter and Wainman (1964) for steers fed 20% forage diets (0.81). However, they noted that k_m increased as portion of roughage in the diet decreased. The calculated MEI at RE = 0 was 98.0 for CON and 102.6 for ECP, a difference of 4.7%.

The linear regression lines above maintenance were as follows:

$$\text{for CON; RE} = 0.6187\text{MEI} - 58.05; R^2 = 0.94;$$

$$\text{for ECP; RE} = 0.6123\text{MEI} - 59.81; R^2 = 0.94,$$

where RE = retained energy (Kcal/kg MBW daily) and MEI = ME intake (Kcal/kg MBW daily). Thus, the k_g for CON steers (0.619) was similar to the k_g for ECP steers (0.612). These values were similar to k_g values reported by Blaxter and Wainman (1964) for steers fed 5% forage diets (0.64). They also noted that k_g increased as portion of roughage in the diet decreased. Blaxter and Martin (1962) noted that the net availability of ME for gain of casein averaged 50.2% when it was infused into the rumen of sheep and 64.7% when it was infused into the abomasum. Because the supplemental protein in our study was predominantly RUP from corn gluten meal, the expected k_g of the protein in corn gluten meal would be similar to values reported by Blaxter and Martin (1962) when casein was infused into the abomasum. The calculated MEI at RE = 0 was 93.7 for CON and 97.7 for ECP, a difference of 4.3%.

The RE vs. MEI quadratic equations of each treatment developed using all the data from both experiments were as follows:

$$\text{for CON; RE} = -0.0009\text{MEI}^2 + 0.9389\text{MEI} - 83.49; R^2 = 0.98;$$

$$\text{for ECP; RE} = -0.0008\text{MEI}^2 + 0.9315\text{MEI} - 86.59; R^2 = 0.98.$$

Values for MEI at RE = 0 were calculated as the first derivative of the quadratic equations. For CON steers, MEI at RE = 0 was 98.2 and for ECP steers was 101.9, a 3.8% increase.

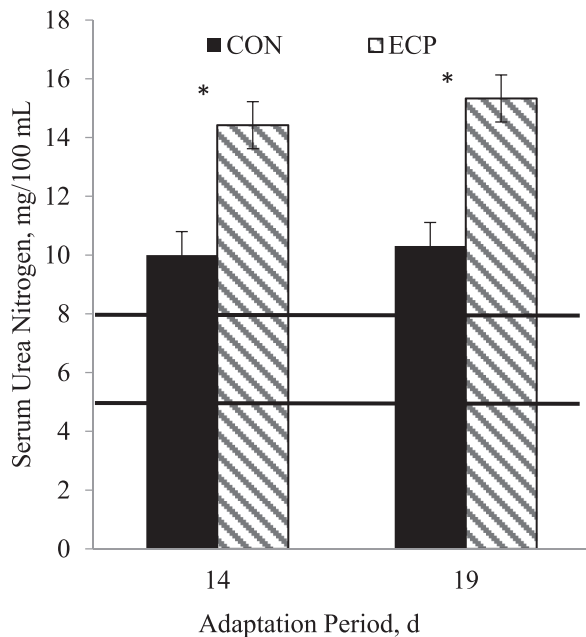


Figure 1. SUN concentration in steers fed a control (CON) and excess CP (ECP) diet at $1 \times$ maintenance intake. Horizontal lines at a SUN (mg/100 mL) of 5 and 8 are in reference to [Johnson and Preston \(1995\)](#) who suggested that excessive nitrogen (N) intake and N wastage would be indicated by blood urea-N concentrations greater than 5 to 8 mg/100 mL. N = 12 and 12 for CON and ECP, respectively. Day ($P = 0.24$) standard error of the mean (SEM) = 0.50, diet ($P \leq 0.01$) SEM = 0.47, and day \times diet ($P < 0.01$) SEM = 0.69. * indicates dietary treatment effect, $P \leq 0.01$.

Using these varied regression methods, although k_m and k_g were not apparently affected by excess protein intake, estimated maintenance energy requirements (i.e., MEI at RE = 0) were consistently 3.8% to 4.7% greater in ECP steers than in CON steers. [Garrett and Johnson \(1983\)](#) noted that the energy cost of both protein and fat deposition was approximately 12 to 13 Kcal/g. These convert to an efficiency of ME utilization of 45% to 50% for protein retention and 70% to 77% for fat retention, respectively. Thus, the greater protein deposition in the ECP steers could account for all, or a portion of, the higher maintenance requirements in ECP steers than in CON steers.

A number of articles have discussed the probable negative effect of excess protein intake on retained energy, and it has been expertly reviewed by [Reed et al. \(2017\)](#). As noted by [Reed et al. \(2017\)](#), the actual biochemical pathways potentially involved in a loss of energetic efficiency are difficult to determine because of multiple physiological factors such as urea recycling and transport of ammonia and urea across the gut wall. [Tyrrell et al. \(1970\)](#) postulated that the energy cost of excreting excess N was 7.2 Kcal/g. Using a larger data set, [Reed et al. \(2017\)](#) noted a smaller value of 1.2 or less Kcal/g of excess N intake. However, it appears that the energy required

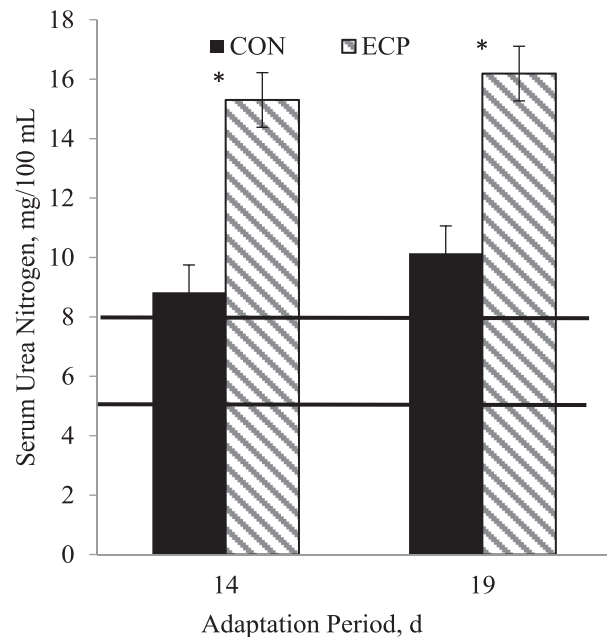


Figure 2. SUN concentration in steers fed a control (CON) and excess CP (ECP) diet at $2 \times$ maintenance intake. Horizontal lines at a SUN (mg/100 mL) of 5 and 8 are in reference to [Johnson and Preston \(1995\)](#) who suggested that excessive nitrogen (N) intake and N wastage would be indicated by blood urea-N concentrations greater than 5 to 8 mg/100 mL. N = 12 and 12 for CON and ECP, respectively. Day ($P = 0.04$) standard error of the mean (SEM) = 0.52, diet ($P \leq 0.01$) SEM = 0.74, and day \times diet ($P < 0.01$) SEM = 0.90. * indicates dietary treatment effect, $P \leq 0.01$.

to metabolize N compounds, when measured by HP or O_2 consumption, is highly variable and dependent upon the source of N ([Reynolds et al., 1991a, 1991b; Ferrell et al., 2001; Reed et al., 2017](#)).

Blood Metabolites

SUN concentrations for $1 \times$ maintenance and $2 \times$ maintenance intake are presented in [Figures 1 and 2](#), respectively. As would be expected, the ECP steers had greater ($P \leq 0.01$) SUN concentration than CON on days 14 and 19 of the adaptation period in both experiments. [Vasconcelos et al. \(2006\)](#) and [Gleghorn et al. \(2004\)](#) also noted greater SUN concentrations as dietary CP content increased. [Johnson and Preston \(1995\)](#) suggested that plasma urea nitrogen concentrations greater than 5 to 8 mg/100 mL indicated excessive N intake and N wastage; however, [Cole et al. \(2003, 2006\)](#) suggested that the borderline was slightly higher, in the range of 9 to 12 mg/100 mL. In this study, SUN concentrations averaged approximately 10 mg/100 mL in the CON steers.

Serum NEFA concentrations for $1 \times$ maintenance and $2 \times$ maintenance intake are presented in [Figures 3 and 4](#), respectively. In Exp. 1, the CON steers had greater ($P \leq 0.02$) serum NEFA

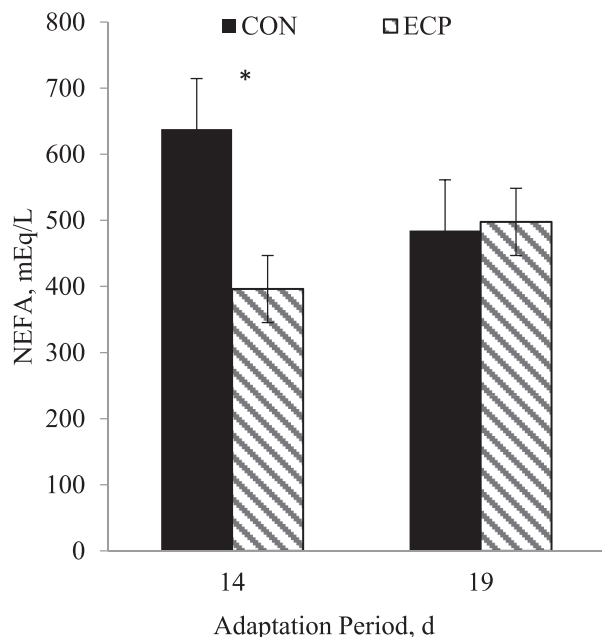


Figure 3. Concentration of NEFA in serum of steers fed a control (CON) and excessive CP (ECP) treatment diet at $1 \times$ maintenance intake. $N = 12$ and 12 for CON and ECP, respectively. Day ($P = 0.68$) standard error of the mean (SEM) = 63.18, diet ($P = 0.11$) SEM = 61.47, and day \times diet ($P \leq 0.03$) SEM = 88.14. * indicates dietary treatment effect, $P \leq 0.02$.

concentration than the ECP steers on day 14 of the adaptation period but not on day 19. No significant effect of diet was detected for serum NEFA concentrations in steers fed at $2 \times$ maintenance. The greater serum concentration of NEFA in the CON steers on day 14 in Exp. 1 could indicate a greater mobilization of fat than in the ECP steers, possibly because of lower ME intake in the CON steers, or an adaptation to the experimental diet. [Trenkle and Barrett \(2005\)](#) noted that animal ADG could be depressed for 14 d or more after a change in dietary protein concentration or source. This could account for the significant difference in serum NEFA concentrations on day 14 of the adaptation period in Exp. 1. Although not statistically compared, serum NEFA concentrations of steers fed at $1 \times$ maintenance were 2 to 3 times greater than steers fed $2 \times$ maintenance.

CONCLUSIONS

The objective of these studies was to evaluate the effects of feeding excess protein from corn by-products on the efficiency of energy utilization of calves fed a finishing diet at $1 \times$ maintenance or at $2 \times$ maintenance energy intake. We hypothesized that this would be demonstrated by an increase in HP per unit of MEI; however, this did not occur in either experiment. However, the

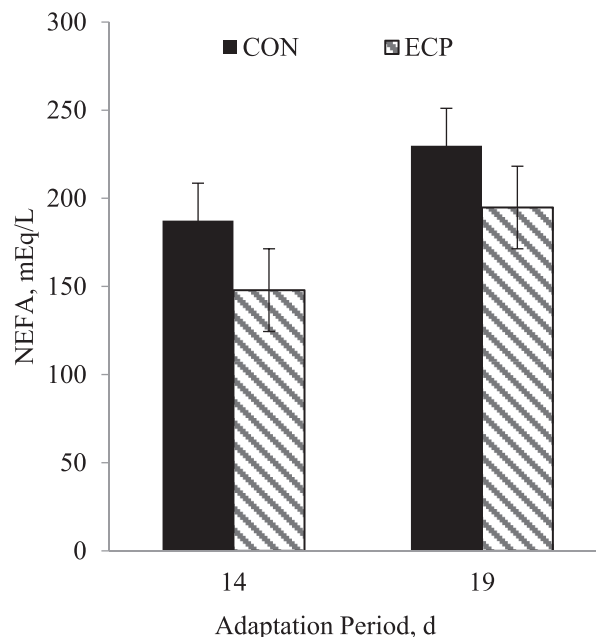


Figure 4. Concentration of NEFA in serum of steers fed a control (CON) and excessive CP (ECP) treatment diet at $2 \times$ maintenance intake. $N = 12$ and 12 for CON and ECP, respectively. Day ($P = 0.08$) standard error of the mean (SEM) = 24.58, diet ($P = 0.18$) SEM = 24.23, and day \times diet ($P = 0.24$) SEM = 34.51.

greater GE and DE content of the ECP diet than CON diet somewhat complicates interpretation of the energy metabolism results. The lack of any effect on HP when steers were fed at $1 \times$ maintenance is most likely due to the small amount of excess N fed as well as a possible MP deficiency in steers fed the CON diet. However, when cattle were fed at $2 \times$ maintenance and excess MP, N exceeded 40 g/d, O_2 consumption per kg of DMI was approximately 5.4% greater in cattle fed excess protein N, and FHP was approximately 6.5% greater. Similarly, when RE was regressed against MEI, calculated maintenance energy requirements were 3% to 4.7% greater in ECP steers than those in CON steers. The poor quality of the supplemental proteins fed in the ECP diet (mostly corn-based) could potentially affect the postruminal amino acid supply. Therefore, additional studies should evaluate the effect of feeding excess RDP and in feeding excess quantities of a higher quality protein sources (i.e., SBM). However, these results should apply to more practical situations where finishing diets contain a high proportion of corn and corn by-products with their inherently poor-quality protein.

Conflict of interest statement. The mention of trade names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation

or endorsement by the U.S. Department of Agriculture.

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