IMPACTS OF REDUCING DIETARY CRUDE PROTEIN WITH CRYSTALLINE AMINO ACID SUPPLEMENTATION ON LACTATING SOW PERFORMANCE, NITROGEN UTILIZATION AND HEAT PRODUCTION

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ABSTRACT

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The inclusion of crystalline amino acids (CAA) has become a standard cost-saving practice to decrease total diet crude protein (CP), reduce ammonia emissions and decrease total heat production when fed to growing and finishing swine. Little research has been conducted, however, using low CP diets, supplemented with CAA, to reduce ammonia emissions and heat balance of the lactating sow and her litter. Consequently, this M.S. research was undertaken; entailing two experiments. The first, tested the hypothesis that lactation performance of sows would not differ if fed diets containing about 3 and 6 percent less CP than the standard corn/soy lactation diet and supplemented with crystalline amino acids (CAA) to meet the AA standardized ileal digestible (SID) requirement of a diet solely based on protein-bound AA. The second experiment tested the hypothesis that in a hot environment, feeding the LCP (low crude protein) from experiment 1 would reduce heat production by the sow, improve her utilization of N, and reduce short-term ammonia emission from her excreta without negative impacts on lactation performance. Results suggest that replacing protein-bound ingredients with CAA does not impact lactation performance of sows under either thermo-neutral or thermal heat stress environments, it optimizes dietary nitrogen utilization and lessens ammonia emission, but it does not reduce metabolic strain or total heat production of the lactating sow and litter.

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I. INTRODUCTION

The increase in global population drives the competition between humans and livestock, and thus the price, of complete protein feedstuffs. One solution for these problems may be in managing dietary crude protein (CP), through replacement with crystalline amino acids (CAAs). While soybean meal (SBM) has more than doubled in price in the last decade (Headey and Fan, 2008), CAAs have become a more prominent replacement for protein bound ingredients. Adding to the economic importance, crystalline lysine, methionine, threonine, and tryptophan have decreased over 40% in the price over the same time period. Utilizing CAA to reduce dietary CP provides more benefits than reduced price as it has been shown to decrease ammonia emissions (Kerr et al., 2003; Otto et al., 2003a; Li et al., 2011), and lessen energy which the animal must lose in its total heat production (Noblet et al. 1987). These added advantages may prove to be more valuable as the modern swine producer faces complex challenges as legislative and social pressures demand accountability for all waste nutrients leaving the farm (solid, liquid, and gas). Average global temperature is increasing and adding to the detrimental seasonal heat stress in many parts of the world. The swine industry is not immune to these financial and environmental challenges, but the opportunities in CAAs may reduce the impact.

Increasingly, farmers are challenged to reduce the loss of nutrients from their production systems. Nitrogen waste is a significant environmental concern due to risks associated with surface and ground water pollution, and ammonia (NH_3) volatilization. As climate change associated with greenhouse gas (GHG) emissions increases in importance, regulations on nutrient management and legislation written emphasizing whole farm balance may address nitrogenous emissions (Montes et al., 2013). Protein, the costliest dietary nutrient, is typically overfed to the sow when provided bound in whole ingredients. While essential for lactation,

uterine repair, and maintenance of the sow, excess protein nitrogen must be removed at an energy cost to the sow.

In growing pigs, crystalline amino acids (CAA) have been implemented without negatively impacting performance (Kerr et al., 2003; Shriver et al., 2003; Lordelo, 2008), decrease feed cost (Shriver et al., 2003) and reduce nitrogen loss to the environment (Otto et al., 2003a; Madrid et al., 2013). Crystalline amino acids have been used to meet the specific amino acid requirements of the growing pig with greater accuracy. The replacement of crude protein, with CAA has been extensively studied in growing swine. The order of limiting amino acids, stage of growth, and price determine how much whole-protein AA can be replaced by CAA in the diet. Although limited, the research available supports use CAA as a replacement for whole-protein sources in the lactating sow. In their study on the feasibility of aggressive CAA supplementation in diets of lactating sows, Manjarin et al. (2012) found that decreasing dietary CP by 4% and meeting the limiting AA requirement via CAA supplementation did not affect piglet ADG, and increased the sow's overall efficiency of AA utilization. Similar findings were recently reported by Huber et al. (2015).

In the current (November 2016) market ingredient prices for dietary ingredients find corn at $116.07/ton, soybean meal at $325/ton, L-Lysine at $1500/ton, DL-Methionine at $3,240/ton, L-Tryptophan at $7,900/ton, and L-Threonine at $2,060/ton. At these prices and using current NRC requirements, balancing sow lactation rations using least cost formulation will replace protein bound ingredients with commercially available CAAs. Utilizing only protein bound ingredients the diet price would be $185.29/ton with 19.0%CP, and with CAAs (L-Lysine, DL-Methionine, L-Tryptophan, and L-Threonine) it would be $177.86/ton with 15.5%CP. From a purely economic perspective, the 18.5% reduction in CP presently saves the producer $7.43. Use

of CAA also has the potential to reduce ammonia emissions from the manure of the sow and litter, and potentially decreasing the energetic cost of N elimination, theoretically improving the sow's ability to perform under heat stress.

II. LITERATURE REVIEW

Impact of Nitrogen Intake on Nitrogen Excretion and the Environment

Ammonia holds extreme social, economic, and biological importance as it can an excellent fertilizer or a pollutant, depending upon management. Ammonia (NH_3), may cause serious health impacts to humans and animals when in greater enough concentrations. It is also a precursor of fine particulate matter, an issue at the forefront of global pollution reduction. Lastly it holds the potential of becoming a greenhouse gas through the process of nitrification. Utilizing nutrition to lessen the impact of swine production on air quality has not been the primary focus of nutrient managers on the farm. Instead, nutrient management planning has made manure application and crop utilization of phosphorus (P) and nitrogen (N) to maintain agronomic balance and prevent leaching into ground water and runoff in surface water paramount. With additional scientific evidence a stronger case may develop for the whole farm balancing of N, including its gaseous emission and contribution to GHGs in the environment.

For growing swine, the reduction of in the dietary amount of CP by using supplemental crystalline amino acids (Kerr et al., 2003, Otto et al., 2003a) and the reduction of P concentration in the diet (Cromwell et al., 1995) decreased N and P concentrations in the manure, respectively. Otto and coworkers (2003a and 2003b) supplemented CAA and observed a 1.2 to 2 g reduction in urinary N excretion per day for each percentage unit reduction in dietary CP. With the replacement of CP with CAA, Li and others (2011) reported a 46% reduction in NH_3 emissions from growing pigs.

Dietary CP and the Lactating Sow

The sow does not have a specific crude protein requirement (NRC, 2012). Instead, the sow's requirements are based on daily amounts (g/day) of indispensable AA at given conditions: body weight, litter size, and minor corrections for environmental temperature. A typical lactating sow, over a 21-day lactation period, consumes about 23 kg of crude protein (CP) or 3.68 kg nitrogen (N) in a corn soy diet. The NRC (2012) indicates roughly 80% of the consumed CP is digested, hence 20% or 0.74 kg N is lost in feces. Assuming body weight remains constant, approximately 60% of the consumed CP or 67% of Lys is secreted into milk. Thus, as much as 40% of absorbed whole-protein N is not utilized and must be excreted in the urine, representing 178 g of N excreted per animal. Improving the efficiency of N utilization in lactating sow may be achieved by increasing CP digestibility, decreasing non-essential AA-N and increasing post-gut AA utilization. These steps offer the best nutritional strategy to reduce N excretion.

It has been previously reported that inclusion rate of crystalline l-Lys, l-Thr and dl-Met of 0.3, 0.1 and 0.088 %, respectively, in an 18.2% CP diet formulated to meet SID Lys of 1.07% does not compromise lactation performance of P1 sows as compared to a 21% CP diet formulated without supplemental CAA (Usry et al., 2009). In the same study, replacing CP with CAA also decreased the weaning-to-estrus interval from 7.1 to 5.5 d. Manjarín et al. (2009) found that feeding an optimum pattern of AA via CAA supplementation to lactation sows increased milk production when presented in a 13.5% CP diet compared to sows receiving a conventional diet containing 17.5% CP and the same AA profile as that of the 13.5% CP diet. The reduced-CP diet also led to increased mammary extraction efficiency of Lys, the first limiting AA in sows fed corn-soybean meal-based diets. In contrast, Perez Laspiur and others (2009) reported that feeding protein in excess of requirement (24 vs. 18% CP) decreased milk

and casein yield, and piglet average daily gain, and this change was associated with a reduction in the expression of a gene responsible for encoding one of the Lys transporter proteins. These studies suggest that excessive dietary N reduces lactation performance even when AA requirements are met, and that sows respond favorably to reduction in dietary N intake when the limiting AA requirements are met.

The biological mechanisms behind improvement in utilization of N and milk casein yield in sows fed reduced CP diets with CAA replacement are unclear. Although the studies by Manjarin et al. (2009) and Perez Laspiur et al. (2009) both indicated increased extraction efficiency of Lys by the mammary gland, Manjarin et al. (2009) and Huber et al. (2015) found no evidence that mammary AA transporter abundance played a role. Rather, competitive inhibition among AA may be a plausible mechanism. Whether reduced CP in lactating sow diets improves the efficiency of N utilization via decrease in energy expenditure remains to be tested. Buttery and Boorman (1976) reported excess CP intake reduced energy metabolism and increased energy expenditure in growing pigs. Buttery and Boorman's conclusions support the probability that the sow's loss in energetic efficiency as CP exceeds the requirement of the limiting AA of the sow may be due to the energy cost of excreting excess nitrogen, potentially increasing her total heat production. This proposition is supported by Noblet et al. (1987) observed that pigs provided a CP intake of 37.5 g protein/Mcal DE produced less heat when compared to pigs with a CP intake of 45 g protein/Mcal DE. In 2003, Kerr et al. observed growing pigs fed a 12% CP diet supplemented with crystalline amino acids had decreased heat production (HP) when compared to pigs fed a 16% CP non-supplemented diet. This was further quantified by Noblet et al. (1987) and Le Bellego et al. (2001), who reported one gram reduction in CP of growing pigs equated a reduction in total HP by 1.8 or 1.7 kcal, respectively.

In growing swine, the relationship between crude protein and HP has been studied under the assumption that decreasing dietary protein level at the same level of limiting essential amino acids, could allow an improved efficiency of metabolizable energy (ME) utilization, potentially increasing yield of the carcass (Noblet et al., 1987). Heat production is defined as the energy lost due to the physical, metabolic, and biological processes and includes feed consumption, maintenance, thermal regulation, and physical activity (NRC, 2012). Heat production can be calculated either via diet calculation or calorimetry. Previous research has determined that the HP of a lactating sow and litter ranges from 1033 to 1166 KJ/$BW^{-0.75}$/day, and from 514 to 692 KJ/$BW^{-0.75}$/day for the sow independent of litter (van den Brand et al., 2000 and Theil et al., 2004).

Considering the metabolic drain of excess N and the previous research in growing swine, reducing the sow's crude protein intake, while maintaining individual essential SID AA requirements, may reduce her HP. If true, this would be a valuable management tool in understanding the sow's energy requirements for least cost formulation and a way to avoid the financial loss of over-formulation with rich protein, during periods of heat stress. This concept was tested by Kerr et al. in 1998 with crossbred barrows in a 2 × 3 factorial arrangement of two environmental temperature treatments (thermoneutral [23°C] or heat stress [33°C]) and three diets (control [16% CP], negative control [12% CP], and a 12% CP diet supplemented with crystalline Lys, Trp, and Thr). Pigs were subjected to indirect calorimetry using measurement of gaseous exchange to estimate total HP. Although the 12% diet supplemented with CAA produced a significantly lower HP when compared to control in the thermoneutral environment, under heat stress the HP of all dietary treatments was significantly reduced, resulting in no significant diet-by-environment interaction. Utilizing CAAs to reduce CP has consistently

reduced the HP in growing pigs. The work by Kerr et al. (1998) however, suggests that under heat stress pigs biologically-reduce total HP to an absolute minimum, regardless of CP concentration, and the influence of reduced CP is not additive. If this is true in sows, implementing CAAs, may beneficial to the environment, but may not improve her production values (piglet ADG, sow ADFI, and return to estrous) under heat stress. Because the metabolic demand of lactation is far greater than that of growth, and the N utilization has more opportunities to lose metabolic and energetic efficiency considering the added AA pools of the milk and mammary gland, reducing CP could be more complementary to the metabolism of the heat stressed sow.

The pig has no storage depot for excess N, so the body must remove it. Blood plasma urea nitrogen (PUN) and milk urea nitrogen (MUN) are indicators of metabolic clearance of excess N, which may reflect excess CP intake. Not all the N will be used when the pig ingests n in the form of protein, free amino acids, and a small percentage of urea; all of which are commonly in its diet. The excess essential amino acids and many of the non-essential amino acids will need to be degraded and the elements used elsewhere. Because it is entirely water soluble, urea N is the primary way the pig can rid itself of excess N. This is true for the sow also. Provided there is not a loss of performance, it can be assumed that the less urea present in the sow's body and products (PUN and MUN), the more accurately the amount of N fed is meeting the sow's need and the more efficient the sow is at metabolizing that amount of nitrogen. This concept was supported by Coma et al. in growing pigs (1995) and lactating sows (1996), who demonstrated that PUN was an effective indicator of lysine requirement. Titrating various Lys levels, the Lys concentration that maximized Lys retention in a full N balance, also minimized

PUN ($P < 0.10$). The metabolic efficiency of protein utilization can be determined by the amount of urea present in the sow and her products.

Sow's Response to Hot Environmental Temperatures

Sow performance is affected by seasonality, with high environmental temperature during the summer period negatively impacting feed intake by 20–55% (Quiniou and Noblet, 1995; Johnston et al., 1999; Pérez Laspiur and Trottier, 2001; Pérez Laspiur et al., 2006) The impaired lactation performance of sows exposed to high ambient temperatures is not only characterized by depressed feed intake, but also by decreased milk production as indicated by decreased litter weight gain, accelerated rate of body protein and fat mobilization, delayed post-weaning return to estrus and increased rebreeding intervals (Prunier et al., 1996; Johnston et al., 1999; Pérez Laspiur et al., 2006). The diminished sow performance has been estimated to cost producers $299 million annually in the U.S. (Malmkvist et al. 2012.)

When a sow experiences effective ambient temperatures higher than ~22°C, defined as the upper critical temperature, the animal must begin making a series of metabolic and physiological changes to minimize the production of more heat, facilitate convection, and (or) employ evaporative processes to lose heat. Initially, under high ambient temperatures a pig will stop activities that produce heat, by restricting movement and drastically decrease feed intake (Huynh et al., 2005). This was documented by Collin et al. in 2001, who subjected group-housed sows to various levels of heat stress from 23° to 33°C, and observed an average of 45 g per day per degree Celsius feed intake reduction in sows as the ambient temperature increased over 23°C. This led to a significant reduction of bodyweight. Sows on this study also made significantly fewer trips to the feeder and spent less time standing and eating. Once the pig has

minimized the heat energy being added to the system, it will significantly increase blood flow to the integument to facilitate heat loss by convection. This process is highly dependent upon hydration state and electrolyte status as the animal must increase body water and conserve dissolved salts to increase blood volume and pressure. If increased circulation and restriction of body heat production are not managing core temperature, panting and sweating, are implemented. In sweating body water is expelled from the skin and the evaporation of that moisture allows for heat energy to be removed from the body. Although the process of sweating is very effective in mammals, the few numbers of cutaneous glands in pigs make it an inefficient solution (Ingram, 1967). Sows, instead, must rely on panting, where air is forced in and out of the bronchi by the animal's respiratory tract and body water is expelled as vapor alleviating heat energy as it is exhaled. Heat loss through evaporation is dependent upon humidity and the animal's hydration state, if the air is too moist for evaporation to occur or the animal cannot afford to lose the fluids necessary, evaporation is ineffective (Huynh et al., 2005).

The above behavioral and physical observations, have a profound effect on the sow body tissues, organs, and their function. Post-mortem examination of muscle in pigs exposed to heat stress during growth showed greater amount of anaerobic glycolysis in heat stressed animals and a trend of decreased muscle color-structure scores, which is considered an indicator of muscle integrity (Addis et al., 1967). For a sow, this mobilization of body tissues can become detrimental to reproductive success. Tompkins et al. (1967), Edwards et al. (1968), Omtvedt et al. (1971), all found that heat-stressed female pigs had delayed signs of estrus by an average of 2 d, fewer viable embryos, and tended to have less corpora lutea. This decrease in viable embryos combined with the significantly lower concentration of LH is a probable cause for the tendency of decreased conception rate witnessed by Omtvedt et al. (1971). Barb et al. (1991) also reported

that LH is significantly decreased under heat stress, which decreases the amount of GnRH. Both, LH and GnRH effect both oxytocin and prolactin secretion, which decreases milk letdown, ultimately decreasing piglet performance (Barb, 1991).

The piglets of heat stressed sows fight two battles, as they may be both immunocompromised and under nourished. Piglets of heat stressed sows tend to be born smaller and less viable, which then makes them less aggressive in suckling and experience reduced colostrum intake and decreased passive immunity (Machado-Neto et al., 1987). Subsequently, increases in morbidity and mortality may occur. Sow milk production is significantly decreased under heat stress as demonstrated by Pérez Laspiur and Trottier (2001), who observed sow's piglets experience a significant decrease in ADG when the sow and litter were subjected to heat stress. Sows exposed to heat stress in late gestation consistently produced the greatest decrease in performance of both sows and piglets post parturition (Omtvedt et al., 1971).

Despite the vast amount of research on heat stress and the sow, little work has been done to quantify the total HP of the heat stressed sow. In growing pigs, it is known that HP decreases with heat stress by as much as 15% of fasting HP; coincident with reduced feed intake (Nienaber et al., 1987). Another research group reported that reduced physical movement accounted for as much as 10% of the pigs reduced fasting HP (van Milgen et al., 1998). This is complicated by Brown-Brandl et al. in 2000, who reported the HP in growing swine with heat stress induced reduction in intake of 13% to be significantly lower than that of pigs subjected to the same feed reduction in thermoneutral environment, suggesting that the activity level of the pigs subjected to heat stress plays a greater role than intake on HP. Brown-Brandl and coworkers (2000) also reported that the thermoneutral treatment groups spent significantly less time lying down than the heat stress treatment groups ($P < 0.05$), however, also suggested that the lower tissue

accretion may play an important role in the HP of heat stressed swine. Considering the lactating sow will most likely sustain a state of a negative energy balance, the impact of heat stress may, like in the growing pig, may result in a reduction in HP.

Conclusions

Previous research would suggest that a reduction in crude protein, while providing essential SID AA requirements would reduce the metabolic challenge heat stress places on the sow and lessen the environmental impact of the sow. Based on the knowledge discussed herein, the aims of the research were to:

- Understand the impact of reduced protein diets with CAA supplementation on the performance of the lactating sow
- Assess the impacts of reduced protein diets with CAA supplementation on nitrogen balance and ammonia emissions.
- Quantify the HP of sows under HS
- Determine if HP could be reduced by reduced protein-CAA diets, mitigating the negative effects of HS.

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III. LACTATION PERFORMANCE OF SOWS FED DIETS WITH GRADED LEVELS OF CRYSTALLINE AMINO ACIDS AS SUBSTITUTE FOR CRUDE PROTEIN AT LYSINE REQUIREMENT

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ABSTRACT

The objective of this study was to test the hypothesis that lactation performance of sows fed diets containing 3 and 6% less CP and supplemented with crystalline amino acids (CAA) to meet the AA standardized ileal digestible (SID) requirement, will not differ when compared to a diet solely based on protein-bound AA. Multiparous purebred Yorkshire sows (n=48) were allocated to one of three dietary treatments: 17.16% CP (Control), 14.48% (MCP) and 11.82% CP (LCP), in a randomized complete block design. Diets were formulated to meet an SID Lys requirement of 0.78%. Control diet did not contain CAA and exceeded all AA SID requirements. The MCP and LCP diets contained L-Lys, L-Thr, L-Trp, and L-Val, in addition to L-Ile and L-Phe for LCP diet only. Voluntary feed intake was measured daily. Sow and piglet BW were recorded on d 0, 3, 6, 9, 12, 15, 18 and 21, and milk samples collected on d 4 and 16. Data were analyzed using rep, parity, day and diets as fixed classification effects, and sow within block as random effect. Compared to Control, voluntary feed intake (kg) of sows fed MCP and LCP did not differ (P = 0.373) and was 5.81, 5.61, 5.65 kg/d (± 0.11) for Control, MCP and LCP, respectively.

Compared to Control, piglet ADG of MCP and LCP did not differ ($P = 0.757$) and were 262, 278 and 258 g/d (± 9) for Control, MCP, and LCP, respectively. Compared to Control, milk urea-nitrogen (MUN) (mg/dL) decreased ($P < 0.001$) for MCP and LCP, and was 8.57, 6.85 and, 2.94 mg/dL (± 0.93) for Control, MCP, and LCP, respectively. In conclusion, unchanged sow lactation performance and reduced MUN suggest that aggressive CAA supplementation in lieu of protein-bound AA's can be employed in the formulation of lactating sow diets to improve dietary N utilization.

INTRODUCTION

With rising prices of high quality protein feedstuffs, it is increasingly cost effective to replace a portion of protein-bound limiting AA with crystalline amino acids (CAA) in growing swine diets. This practice reduces N excretion and loss to the environment. Ammonia from livestock operations is regarded as a health and environmental concern as it is a precursor of fine particulate matter. This has sparked regulations on manure management and legislation emphasizing whole farm nutrient balance (Montes et al., 2013). Dietary reduction in CP has been shown to dramatically reduce ammonia emissions (Li et al., 2015; Powers et al., 2006; Panetta et al., 2006). In growing pigs, CAA have been used to optimize growth performance costs (Kerr et al.,2003; Shriver et al., 2003; Lordelo 2008), decrease feed cost (Shriver et al., 2003) and reduce N loss to the environment (Otto et al., 2003; Madrid et al., 2013). In addition, as the swine industry continues to compete with human food production for protein rich ingredients, emphasis on alternative ingredients has become paramount. Soybean meal (SBM), for instance, has more than doubled in price in the last decade (Headey and Fan, 2008). Manjarin et al. (2012) found that decreasing dietary CP by 4% (from 17.5% to 13.5%) and meeting the limiting AA

requirement via CAA supplementation increased the efficiency of Lys and Arg utilization by the mammary gland without affecting lactation performance. Huber et al. (2015) showed that reducing CP by 2.7% (from 16% to 13.2%) with supplemental CAA increased casein yield and global N utilization. In both studies, sows were fitted with either mammary vein and carotid catheters (Manjarin et al., 2012) or urinary catheters (Huber et al., 2015), which reduced sow feed intake and limited the number of sows per treatment. The goal of this study was to test similar diets on a larger number of sows that were kept in a commercial-like setting. In addition, the lowest CP diet tested by Huber et al. (2015) may have been limiting in Phe and was 0.49%, which was well above the minimum N recommended by NRC (2012). We hypothesized that lactation performance of sows fed diets containing 3 and 6% less in intact CP and supplemented with CAA to meet standardized ileal digestible (SID) requirement of limiting AA, would not differ from sows fed a diet solely based on protein-bound AA ingredients. The objectives were 1) to determine lactation performance of sows fed Control and reduced CP diets, and 2) to evaluate post-feeding AA and N utilization of sows fed Control and reduced CP diets.

MATERIALS AND METHODS

Animals were managed throughout the study in accordance with requirements of the Michigan State University All University Committee on Animal Use and Care.

Animals, Housing and Experimental Design

Forty-five, multiparous (parity range from 2 to 7) purebred Yorkshire sows, mated to PIC327 (PIC USA, Hendersonville, TN) were used in 2 replicates (rep) of 24 sows. Sows were allocated to 2 identical rooms (each containing 12 farrowing stalls) and 3 different diets according to parity (2, 3, 4+) and parentage. Sows were then randomly assigned to 4 blocks per

room and 3 sows per block, with block representing room location. Thus, the number of sows per treatment was balanced across 2 rooms and location within rooms. Sows were placed in farrowing stalls on day 106 (± 2) of gestation. Piglets were cross-fostered within the first 36 h of lactation, such that each litter was adjusted to 10 piglets of similar weight within a litter.

Sow Lactation Performance Data Collection

Sows and piglets were weighed on d 1, 3, 6, 9, 12, 15, 18, and 21 of lactation. Sows were weighed prior to feeding. Feed intake was assessed daily. Ultrasound back fat measurements (RENCO® LEAN-MEATER® Renco Corporation, Minneapolis, MN) were taken on d 1 and 21, at the same exact point on the loin indicated by a marked and shaven at the sow's last rib.

Experimental Diets

Three diets were formulated to contain 17.16 (Control), 14.48 (MCP), and 11.82 (LCP) % CP (Table 1). All diets met a minimum SID Lys requirement of 0.78%. This requirement was estimated using a sow weight of 225 kg post farrowing and a litter size of 10 piglets, with an ADG of 260 g over a 21-d lactation period (NRC, 2012). The Control diet was formulated using soybean meal, corn and pelleted soybean hulls as sole ingredient sources of AA. The MCP and LCP diets contained less SBM than Control diet and were supplemented with increasing concentrations of CAA, including L-Lys, L-Val, L-Thr and L-Trp. The LCP diet also included DL-Met, L-Ile and L-Phe. Diets were isocaloric and contained similar concentration of fermentable fiber (Tables 2 and 3). The calculated CP and AA concentrations of each diet was comparable to the analyzed values (Table 3). Titanium oxide was included as an indigestible marker for estimation of nutrient digestibility.

Diets were provided to sows beginning on day 106 (± 2) of gestation and fed throughout lactation. Sows were fed 1 kg of diet twice daily pre-partum. After parturition, sows were fed

equal portions 3 times daily (0700, 1500, and 2300). Daily feed intake was estimated using the NRC (2012) model (Table 4), and consisted of 2.301 kg on d 1 and slowly increasing to 7.520 kg on d 21 to achieve an average daily feed intake of 6 kg over the entire lactation period.

Milk Sampling

Milk samples were collected on d 4 (early lactation) and on d 16 (peak lactation) of lactation. Half of the piglets were removed from the sows for approximately 1 h, and sows were administered 1 mL of oxytocin IM (20 IU/mL oxytocin, sodium chloride 0.9% w/v, and chlorobutanol 0.5% w/v; VetTek™, Blue Springs, MO) prior to feeding. Once milk let down occurred, the sow was fed to keep her standing, and approximately 100 mL of milk was manually collected across all glands. Piglets were then returned to the sow and allowed to suckle. Approximately 50 mL was submitted to Universal Lab Services, LLC (Northstar Cooperative Inc., Lansing, MI) for determination of true protein, urea nitrogen, fat, and lactose concentrations via infrared spectroscopy. The remaining milk samples (approximately 50 mL) were frozen at -20°C for later determination of casein concentration.

Blood Sampling

Pre-prandial blood samples were collected on d 6 (early lactation) and on d 18 (peak lactation) of lactation. Blood was drawn from a subset of sows prior to their 0700 feeding. Only sows who had eaten all the feed provided in the previous feeding by 0500 were sampled. Of the samples collected, only the sows who had plasma samples from both early and peak lactation that were not contaminated by blood cells were analyzed for serum AA. This protocol was developed to provide a thorough sample of the plasma amino acid pool, while minimizing the disruption of the normal eating and lactating behavior of the sow associated with fasting, and allowed for blood sample at a relatively steady state of intake. Blood was collected from the

jugular vein in non-heparinized vacutainer tubes (Becton, Dickinson and Company, Franklin Lakes, New Jersey), centrifuged, and the serum removed, aliquoted and stored at -20°C until analyzed for physiological AA concentrations.

Plasma and Milk Analyses

Amino acid concentrations in serum were analyzed at the Agricultural Experiment Station Chemical Laboratories (University of Missouri-Columbia, Columbia, MO) per AOAC (Official Method 982.30, 2006). For determination of casein concentration, milk samples were thawed overnight in a refrigerator, gently mixed and defatted by centrifugation at 1500 × *g* for 30 min at 4 °C. The fat layer was removed by gentle aspiration using a Pasteur pipette attached to a tube fitted to a low-pressure pump. Once defatted, milk caseins were precipitated by decreasing the milk pH between 4.6 (±1) using 1M HCl. Once precipitated, milk samples were placed in a refrigerator overnight (1.6° C) and the pH was assessed the following morning. If needed, pH was readjusted between 4.7 and 4.5 with 1M HCl. Samples were centrifugated at 1500 × *g* for 15 min, and the supernatant removed and discarded. The casein pellet was re-suspended in distilled water and centrifugated at 1500 × g for 15 min, the supernatant removed and discarded. This step was repeated one more time and the casein pellets were frozen at -20°C and freeze-dried. The freeze-dried pellets were weighed immediately after removal and casein concentration was determined from the pellet weight relative to total milk weight.

Statistical Analysis

The experiment was designed as a randomized complete block design with 15 sows per treatment (45 sows in total). One sow was removed from the LCP treatment because she did to eat and subsequently failed to lactate. This was unrelated to the treatment as records indicated poor lactation in her previous parity. Performance data were analyzed using the MIXED

procedure of SAS (SAS Inst. Inc., Cary, NC). A full model with the fixed effects of diet, rep, sire of dam, initial sow body weight as a regression variable, and the random effects of sow within diet-by-block was used. The effects of sire of dam and initial body weight were not significant (minimum $P > 0.1$) and were removed from the model. The reduced model included diet and rep as a fixed effect and sow within diet-by-block as a random effect. Milk and blood serum data were analyzed using GLMMIX and linear contrast statement (-1, 0, 1) in SAS. The reduced model includes fixed effects diet, day of lactation (4 and 16), and diet by day of lactation, and sow within diet by block was included as random effect.

RESULTS

Sow and Piglet Performance

Piglet ADG ($P < 0.01$; rep 1: 280g/d, rep 2: 251g/d), litter growth ($P < 0.01$; rep 1: 2.758 kg/d, rep 2: 2.394 kg/d), sow ADFI ($P < 0.001$, rep 1: 5.97 kg/d, rep 2: 5.44 kg/d), change in sow backfat depth ($P < 0.001$; rep 1: -2.5mm/d, rep 2: 2.7 mm/d), sow average daily body weight change ($P < 0.001$; rep 1: -0.06 kg/d, rep 2: -0.6 kg/d), milk production ($P < 0.01$; rep 1: 9.38 kg/d, rep 2: 8.20 kg/d) in rep 1 and 2 differed. There was no interactive effect between rep and diet for any of the response variables ($P > 0.10$) and data were pooled across reps. Overall sow feed intake, weight change, and backfat depth, piglet ADG and litter growth did not differ among dietary treatments (Table 5). Daily sow feed intake (Figure 1) and litter gain (measured every 3 d; Figure 2) did not differ between dietary treatments.

Milk Composition

When compared to the Control treatment, milk fat, true protein and lactose on d 4 and 16 of lactation did not differ for sows fed MCP and LCP (Table 6). On d 4 (early) and 16 (peak) of

lactation, MUN decreased as dietary CP decreased on (Linear; $P < 0.0001$). Milk casein concentration on d 4 of lactation did not differ in MCP and LCP compared to Control and increased (Linear, $P = 0.03$) as CP decreased on day 16 of lactation.

Serum AA and Urea Concentrations

Serum concentrations of essential His Phe and Trp, and non-essential Ala and Pro, did not differ between dietary treatments or stage of lactation. Serum concentration of Asn, Gly, Tyr, and 3MH concentrations did not differ between stages of lactation. Serum concentrations of Arg, Ile, Lys, Met, Val and Cys were lower in peak lactation compared to early lactation ($P < 0.05$) for all treatments. Serum concentrations of Leu, Thr and Ser did not differ between early and peak lactation for the control diet, and were lower for MCP and LCP diets in peak lactation when compared to early ($P < 0.05$). Serum urea concentration was greater at peak lactation for the control diet and MCP, and lower at peak lactation for LCP, when compared to early lactation ($P < 0.05$). During early lactation, compared to control, serum urea concentration was lower in the LCP ($P < 0.01$), and did not differ for MCP diet. During peak lactation, MCP diet was less than control, and LCP diet was less than MCP and control diet. Serum Lys, Thr and Val increased ($P < 0.05$) as dietary CP decreased. Serum Met, Ile, and Leu decreased ($P < 0.05$) with decreasing dietary CP, while Arg, His, Phe, and Trp did not differ between diets (Table 7).

DISCUSSION

Feeding reduced CP diets containing 14.79 and 12.56% CP with CAA to meet the SID requirement of AA did not impact lactation performance; measured as piglet ADG and sow feed intake. Similar indices of performance were unchanged with CAA use as a feeding strategy in work by Manjarin et al. (2012) and Huber et al. (2015). In our study, CP was lower yet (12.98,

analyzed) than the lowest amount fed in these two previous studies, which affirms repeatability of the concept. Using CAA as a partial replacement for CP reduced MUN over 2-fold in early lactation and over 5-fold in peak lactation as well as increasing the casein fraction by 25% in peak lactation. This reduction in MUN is consistent with the results of Huber et al. (2015), who observed the same 2-fold difference in early lactation and an over 5-fold difference at peak lactation. In both studies, MUN did not change for the reduced CP diet from early to peak lactation, while the control diet nearly doubled over the course of lactation.

A similar trend was observed in plasma urea nitrogen (PUN). During early lactation, the PUN of the LCP diet was nearly half of that observed in Control sows, and at peak lactation the PUN increased in Control sows but decreased, to 1/3 the PUN of control, in sows receiving the LCP treatment. Considering the greater sow feed intake near peak lactation (Figure 1), it was expected that all treatments would experience increases in MUN and PUN at peak lactation, not just the Control and MCP diets. PUN and MUN are mostly waste pools for excess N, and are by-products of the urea cycle. The urea cycle is an energy expensive process, which consumes 2 molecules of ammonia, and 1 molecule of carbon dioxide, creating 1 molecule of urea ($(NH_2)_2CO$), and regenerates a molecule of ornithine. The reduction in MUN coupled with the reduction in PUN without a decrease in milk protein or casein concentration suggests these sows were either more efficient in using digested N and avoiding the use of the energy-demanding urea cycle, or they were catabolizing body stores to maintain milk composition when provided the lowest CP diet. If the sows were catabolizing lean protein, a significantly greater weight loss would be expected for the LCP, which was not observed.

Serum AA measurements complicate the explanation of our results. Serum Lys concentration more than doubled, as dietary CP decreased, despite all diets being formulated to

the same SID amount. Samples were collected when the sow was at a steady state of feed intake, thus the timing of sampling relative to a meal should not have been the reason for this difference. This suggests utilization of AA may have been impacted. Huber et al. (2015) reported similar changes in serum Lys concentrations when samples were obtained 15 h postprandial. Like serum Lys, the serum concentrations of the next two limiting AA, Thr and Val, the opinion of this author, also increased by over 30% as CP was decreased in early lactation. However, at peak lactation, Thr was not changed, while Val increased 30% as CP was decreased. In terms of intake, Thr and Val, only differ by 0.04% (SID calculated) from the Control to the LCP diet. Serum Ile concentration decreased by nearly 0.20% (SID calculated) as dietary CP was decreased, making it the most limiting amino acid to dramatically reduce with treatment. This suggests a higher dietary requirement for Ile than modeled. If Ile was limiting, the utilization and ultimately absorption of other essential amino acids would decrease, which would explain the increase in serum Lys and Val concentrations. Under the assumption that Ile is limiting, it is possible that there are performance and efficiency opportunities left on the table. The addition of more Ile per unit of intake by sows fed the LCP diet may improve efficiency and increase sow performance.

Although we question whether the LCP diet met the sows Ile requirement, the overall results of this study, particularly the maintenance of lactation performance, the desirable change in MUN and casein synthesis, suggests CAA can be used in lactating sow diets to optimize N utilization and reduce dietary costs. These findings have applicability on-farm, with the biological potential to take out a 20 to 40% of the soybean meal traditionally in the lactation diet. Economics or cost of CAA will determine exactly how much of the biological potential we be realized.

APPENDIX

Table 1. Composition of experimental diets.

Item	Control	MCP	LCP
Corn	61.64	63.92	65.57
Soybean meal, dehulled, solvent extracted	25.20	18.42	11.73
Choice white grease	3.82	4.26	4.77
Sugar food by-product[1]	5.00	5.00	5.00
Soybean hulls	-	3.65	7.50
L- Lys·HCl	-	0.21	0.41
L-Val	-	0.09	0.21
L-Thr	-	0.05	0.15
L-Trp	-	0.01	0.04
DL-Met	-	-	0.06
L-Phe	-	-	0.07
L-Ile	-	-	0.04
Limestone	1.45	1.40	1.38
Mono calcium phosphate	1.60	1.70	1.78
Vitamin premix[2]	0.25	0.25	0.25
Mineral premix[3]	0.125	0.125	0.125
Sow pack[4]	0.25	0.25	0.25
Se 270[5]	0.0675	0.0675	0.0675
Salt	0.50	0.50	0.50
Titanium oxide	0.10	0.10	0.10
Total	100.00	100.00	100.00

[1]National Ingredient Corporation: CP 1.00 %; NE = 2719 kcal/kg (estimated using ME equation of Noblet et al. (2003)); fermentable fiber 0.05 %.

[2]Vitamin Premix provided the following per kg of diet: 3,000 IU vitamin A, 300 IU vitamin D_3, 20 IU vitamin E, 1 mg menadione (vitamin K), 20 µg vitamin B_{12}, 4 mg riboflavin, 10 mg D-pantothenic acid, and 15 mg niacin.

[3]Mineral Premix provided the following per kg of diet: 640 mg Fe (as $FeCO_3$), 260 mg Zn (as ZnO), 36 mg Mn (as MnO_2), 20 mg Cu (as $CuCl_2$), and 0.58 mg I (as ethylenediamine dihydroiodide).

[4]Sow Pack provided the following per kg of diet: 0.10 mg biotin, 250 mg choline (as choline chloride), 0.75 mg folic acid, 2.3 mg vitamin B_6 (as pyridoxine·HCl), 10 IU vitamin E (as DL-tocophorol acetate), 90 µg chromium (as chromium picolinate), and 23 mg carnitine (as L-carnitine).

[5]Cargill: Se 270 mg premix, sodium selenide.

Table 2. Calculated nutrient composition of experimental diets[1].

Item	Control	MCP	LCP
Net energy (NE) kcal/kg	2582	2582	2582
CP, %	17.16	14.79	12.56
Fermentable fiber, %	10.58	10.37	10.22
Total P, %	0.68	0.67	0.65
Standardized total digestible P, %	0.74	0.44	0.44
Ca, %	0.89	0.88	0.89
SID[2] CP, %	14.57	12.15	9.74
SID AA			
Arg, %	1.02	0.82	0.63
His, %	0.41	0.35	0.28
Ile, %	0.62	0.51	0.43
Leu, %	1.32	1.14	0.96
Lys, %	0.78	0.78	0.78
Met, %	0.24	0.21	0.23
Met + Cys, %	0.48	0.42	0.41
Phe, %	0.74	0.61	0.56
Phe-Tyr, %	1.22	1.01	0.81
Thr, %	0.53	0.49	0.49
Trp, %	0.18	0.15	0.15
Val, %	0.68	0.66	0.66
SID Lys/NE, g/Mcal	3.013	3.033	3.03

[1]Nutrient values were calculated using NRC (2012).

[2]Standardized ileal digestible.

Table 3. Calculated total and analyzed CP and AA composition of experimental diets1, %.

AA	Control		MCP		LCP	
	Calculated	Analyzed	Calculated	Analyzed	Calculated	Analyzed
CP	17.16	17.55	14.48	15.25	11.82	12.98
Essential						
Arg	1.1	1.19	0.89	0.86	0.69	0.69
His	0.47	0.47	0.4	0.37	0.33	0.3
Ile	0.71	0.76	0.59	0.59	0.46	0.51
Leu	1.5	1.62	1.31	1.38	1.11	1.16
Lys	0.9	0.99	0.89	0.89	0.88	0.87
Met	0.28	0.27	0.24	0.22	0.27	0.22
Phe	0.85	0.89	0.71	0.71	0.64	0.63
Thr	0.64	0.69	0.59	0.59	0.58	0.56
Trp	0.2	0.22	0.17	0.17	0.16	0.16
Val	0.8	0.83	0.76	0.76	0.76	0.71
Total						
Non-essential						
Tau	-	0.18	-	0.16	-	0.18
Asp	-	1.83	-	1.36	-	1.07
Ser	-	0.82	-	0.66	-	0.54
Glu[2]	-	3.28	-	2.61	-	2.07
Pro	-	1.06	-	0.9	-	0.78
Gly	-	0.75	-	0.6	-	0.51
Ala	-	0.93	-	0.79	-	0.68
Cys	-	0.28	-	0.24	-	0.19
Tyr	-	0.63	-	0.47	-	0.41
Total						
EAA: NEAA						

[1]Calculated CP and essential AA values per NRC (2012).
[2]Glutamate + Glutamine.

Table 4. Sow and Litter Performance.

Item	Control	MCP	LCP	SEM	*P*-value Diet
No. of sows	15	15	14	-	-
Sow BW, day 1, kg	221	235	244	11	0.32
Sow ADFI, kg	5.81	5.61	5.65	0.11	0.38
Sow daily BW change, g/day	-270	-413	-358	193	0.44
Litter size	9.80	9.74	9.93	0.13	0.31
Litter growth rate, kg/d	2.53	2.64	2.56	0.11	0.75
Piglet ADG, g	262	278	258	9	0.19
Milk production kg[1]	8.63	8.97	8.78	0.35	0.76
Backfat depth, mm, d 1	13.4	14.4	14.7	1.23	0.67
Backfat depth, mm, d 21	14.4	14.0	14.4	0.8	0.93
Backfat depth change, mm, d 1-21	0.2	0.0	0.1	0.7	0.93

[1]Calculated by equation NRC 2012

Table 5. Effect of decreasing CP on milk nutrient composition[1].

Item	Control	MCP	LCP	SEM	*P* - value	
					Linear	Diet
Early Lactation						
Fat, %	9.41	9.23	8.55	0.5	0.23	0.44
True protein, %	4.84	4.97	4.75	0.13	0.7	0.41
Lactose, %	4.89	4.86	4.98	0.11	0.54	0.7
MUN, mg/dl	5.7[a]	4.64[a]	2.61[b]	0.85	0.02	0.04
Casein, %	3.7	3.25	3.76	0.45	0.82	0.27
Casein:true protein	0.78	0.66	0.79	0.01	0.88	0.16
Peak Lactation						
Fat[2], %	7.57	8.35	8.51	0.44	0.09	0.2
True protein[2], %	3.93	3.88	4.17	0.16	0.28	0.13
Lactose[2], %	5.89[a*]	5.74[ab*]	5.47[b*]	0.1	<0.01	<0.01
MUN[3], mg/dl	10.06[a*]	4.58[b*]	1.15[c*]	0.75	<0.01	<0.01
Casein[3], %	2.85[a]	3.15[a]	3.81[b]	0.37	<0.01	<0.01
Casein:true protein	0.67[a]	0.80[ab]	0.90[b]	0.08	<0.01	0.02

[1] All differences a vs b vs c are noted within phase of lactation.

[2] Sample effect of $P < 0.05$.

[3] Sample trend of $P < 0.01$.

[*] Diet-by-sample effect $P < 0.05$.

Table 6. Sow serum AA concentrations of sows fasted 2.5 (±1) h between meals provided every 8 h, µmole/L.

	Early lactation			Peak lactation			SEM	*P*-value	
AA	Control	MCP	MCP	Control	MCP	LCP		Diet	Diet × stage[3]
Urea	3024.13[a]	2514.74[ab]	1662.92[b]	4412.28[a,*]	3250.88[b,*]	1402.80[c,*]	264.86	<0.01 [L]	0.01
Essential									
Arg	219.70	243.42	191.36	171.69[*]	174.20[*]	142.11[*]	20.89	0.13	0.81
His	91.28	105.12	98.87	103.12	101.02	81.57	7.58	0.16	0.07
Ile	87.18	87.44	72.29	78.68[a,*]	63.79[ab,*]	51.15[b,*]	6.57	<0.01 [L]	0.38
Leu[2]	163.68[a]	191.89[a]	154.31[b]	169.51[d,*]	142.24 [de,*]	126.4 [e,*]	13.46	0.05	0.06
Lys[2]	126.43[a]	182.88[ab]	271.79[b]	63.42[a,*]	100.28 [ab,*]	165.6 [b,*]	31.99	<0.01 [L]	0.67
Met[2]	36.90[a]	38.41[a]	63.72[b]	29.93[a,*]	17.44 [b,*]	37.51[a,*]	5.55	<0.01 [L]	0.02
Phe	64.10	78.81	62.70	65.47	59.49	51.41	7.49	0.21	0.27
Thr[2]	140.91[d]	155.42[d]	218.26[e]	128.26[*]	98.9[*]	120.6[*]	19.27	0.05 [L]	0.05
Trp	31.97	40.15	38.98	36.58	32.84	30.82	4.14	0.77	0.09
Val[2]	201.19[a]	263.77[b]	322.96[c]	192.79[a,*]	198.60[a,*]	284.16 [b,*]	16.65	<0.01 [L]	0.17
Non-essential									
Ala[2]	449.21[a]	550.86[a]	730.66[b]	418.04[a]	482.59[a]	650.25[b]	52.03	<0.01 [L]	0.82
Asn	37.79	36.18	37.95	49.29	48.35	38.17	5.89	0.58	0.48
Cys	1.57	2.56	3.25	0.00[*]	0.41[*]	1.38[*]	1.21	0.40	0.96
Glu	160.67[a]	255.26[ab]	278.50[b]	157.22	155.04	256.88	46.56	0.04 [L]	0.38
Gln	528.31	505.58	581.84	464.62	501.74	472.46	50.46	0.79	0.52
Gly[2]	716.03[a]	864.56[ab]	1035.65[b]	818.64[a]	833.49[ab]	1018.39[b]	78.48	<0.01 [L]	0.55
Pro	222.60	288.70	275.02	226.16	242.73	235.90	21.44	0.10	0.37
Ser	111.09	121.51	116.32	118.68[*]	89.04[*]	89.93[*]	8.64	0.15	<0.01
Tyr	93.51	96.51	79.62	121.72[a]	84.46 [b]	51.97[c]	11.60	<0.01 [L]	0.03
3MH[2]	34.20[ab]	43.41[a]	29.14[b]	29.56[ab]	35.98[a]	24.24 [b]	3.67	<0.01	0.89

[1] Linear at $P < 0.05$.

[2] All differences are noted within phase of lactation.

[3] Stage of lactation (stage)

[4] Within phase of lactation, a vs b vs c: $P < 0.01$.

[5] Within phase of lactation, d vs e vs f: $P < 0.05$.

* Peak lactation differs from early lactation at $P < 0.05$

Figure 1. Litter ADG (kg) by treatment over a 21 d lactation period where ADG was calculated as an average between each weight taken on days 1, 3, 6, 9, 12, 15, 18, and 21 with no significant differences ($P > 0.10$).

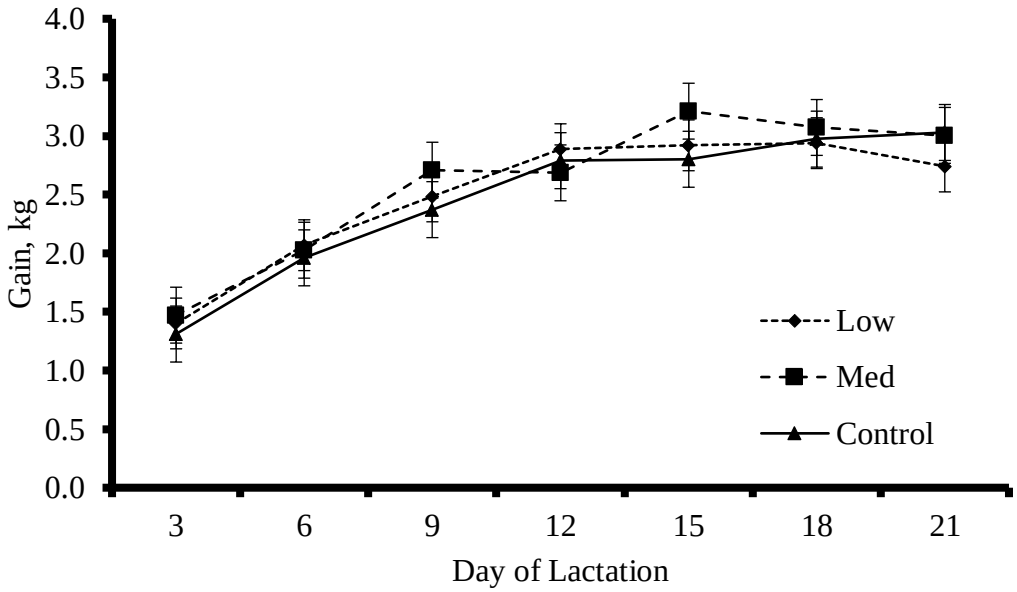
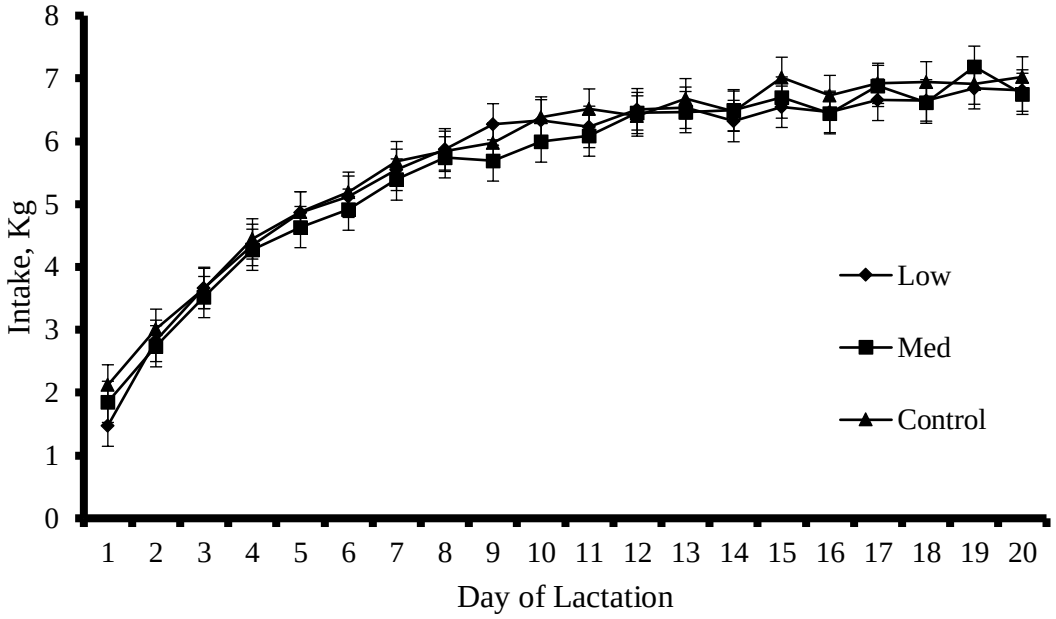


Figure 2. Average daily feed intake (ADFI) of the lactating sow (kg) by treatment over a 21 d lactation measured daily with no significant differences ($P > 0.10$).



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IV. IMPACT OF REDUCED DIETARY CRUDE PROTEIN CONCENTRATION WITH CRYSTALLINE AMINO ACID SUPPLEMENTATION ON LACTATION PERFORMANCE AND AMMONIA EMISSION OF SOWS HOUSED UNDER THERMO-NEUTRAL AND THERMAL HEAT STRESS ENVIRONMENTS

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ABSTRACT

The objective of this study was to test the hypothesis that feeding a diet containing lower CP and supplemental CAA compared to a diet meeting limiting AA requirement without CAA reduces heat production, improves N utilization, does not impact lactation performance and reduces ammonia emission of lactating sows in hot environments. Thirty-six multiparous sows were allocated to a 2 × 2 factorial arrangement of 2 temperatures (thermoneutral [21°C; TN] and heat stress [31.5°C; HS]), and 2 diets (17.16 [Control] and 11.82% CP [Low]), in a randomized complete block design. The HS sows were acclimated during late gestation to a temperature increase from 21 to 31.5 °C. During lactation, temperature for HS sows were incrementally changed (24 to 31.5°C and 31.5 to 24°C) from 0500 to 1500 and 1800 to 0500, respectively. Control diet met SID Lys requirement with no added CAA and Low diet contained added crystalline Lys, Thr, Trp, Val and Phe to meet NRC 2012 requirements. Piglet ADG, sow feed intake (FI), true milk protein (TMP), sow weight loss (ΔBW), change in sow backfat depth, sow body temp (BT), days from weaning to estrus, heart rate (HR), and respiration rate (RR) were

unaffected by diet. The Low treatment resulted in decreased ($P < 0.0001$) milk urea N (MUN) and ammonia emissions. Ammonia emissions were not affected by environment. The hot environment resulted in greater ($P < 0.05$) ΔBW, HR and RR, and less ($P < 0.05$) piglet ADG. Nitrogen retained as a percent of N intake was not impacted by environment. Nitrogen retained as a percent of N intake was greater ($P < 0.01$) for the LCP diet (71.62%) when compared to Control (59.12%). Oxygen, carbon dioxide exchange, respiratory quotient, and total heat production over the entire lactation were not impacted by reducing CP. Carbon dioxide emission was less ($P < 0.01$) for sows exposed to HS (6187.37 g/d) when compared to TN (7038.06 g/d). Oxygen intake was not impacted by diet or environment. Total heat production was greater ($P < 0.01$) in sows exposed to HS environment (1.92 mJ/$BW^{0.75}$•d) when compared to TN housed sows (2.04 mJ/$BW^{0.75}$•d). In conclusion, feeding reduced CP diet to lactating sows improved N utilization and did not alleviate heat stress. The reduction of dietary CP in conjunction with aggressive CAA supplementation may be implemented for lactating sows to mitigate ammonia emissions. The maintenance of lactation performance as observed previously was confirmed.

INTRODUCTION

Replacing a portion soybean meal with crystalline AA (CAA) to meet the requirements of growing swine has been used to reduce dietary cost since the 1960s. In more recent years, inclusion of CAA in reduced CP diets fed to growing and finishing pigs have been shown to reduce ammonia emissions (Li et al., 2011), and decrease heat production (Noblet et al., 1987, Kerr et al., 1998, and Le Bellego et al., 2001). The relationship between heat production and animal response to heat stress appears complex. In finishing pigs, a one gram reduction in dietary CP decreased daily HP by 1.8 kcal (Noblet et al. 1987) and 1.7 kcal (Le Bellego et al.

2001, respectively). But, Kerr et al. (1998) showed that in heat-stressed pigs, reduced CP diets did not further reduce total HP. These researchers suggested that under heat stress, pigs reduced HP to a minimum, such that the effect of dietary reduction in CP on HP becomes insignificant.

Lactating sows produce substantially more heat than growing and finishing pigs (~699 vs. ~1166 KJ/$BW^{0.75}$•d), and as such may be more responsive to reduction in dietary CP concentration. In a previous study (Chamberlin, Chapter 3), we showed that in sows fed a diet reduced by 6% CP relative to sows fed a non-reduced CP diet, serum and milk urea N (MUN) is decreased nearly 2-fold. In the study by Huber et al. (2015), similar diets let led to significant improvement in N utilization as well as reduction in serum urea-N. Together, these studies suggest that lactating sows receiving low CP diets have improved energetic efficiency, which may be beneficial, in particular, to sows under hot environmental conditions. With the notion that heat increment associated with protein digestion and metabolism is as much as 40% of ME intake, the goal of this study was to test the hypothesis that feeding a diet containing lower CP and supplemental CAA compared to a diet meeting limiting AA requirement without CAA reduces heat production, improves N utilization, does not impact lactation performance and reduces ammonia emission of lactating sows in hot environments. The objectives of this study were to determine 1) lactation performance and N utilization; 2) sow and litter heat production; and 3) gaseous emissions, of sows housed under thermo-neutral and thermal heat stress environments, and fed diet containing reduced CP or non-reduced CP.

MATERIALS AND METHODS

Animals were managed in accordance with requirements of the Michigan State University All University Committee on Animal Use and Care.

Animals, Experimental Design

Thirty-six second and third parity, purebred Yorkshire sows, mated to PIC327 (PIC USA, Hendersonville, TN), were used with 3 reps of 12 sows. Sows were allocated in a 2 × 2 factorial arrangement of 2 environmental temperature and 2 dietary treatments. The 2 environmental temperatures were 21°C and 31.5°C, respectively referred to as thermo-neutral (**TN**) and heat stress (**HS**). The 2 dietary treatments contained 17.16% (**Control**) and 11.82% CP (**Low**). Piglets were cross-fostered within the first 36 h of lactation, such that each litter was adjusted to 10 piglets of similar weight per sow.

Housing

Sows were individual housed in environmentally-controlled rooms (2.14×3.97×2.59 m) each equipped with an elevated farrowing stall on plastic coated steel flooring. Two separate steel manure collection pans (3.05 m × 1.52 m × 20.0 cm) were placed below each crate for separate collection of urine and waste water. Each of the rooms was adjacent forming a single row, with air inlet flow powered by 1 make-up air unit located in the center for all 12 chambers. Sows were blocked by parity, allocated to a treatment based on parentage. Sows were randomly assigned to one of 3 zones, then randomly allocated to one of 4 rooms within the zone, such that each treatment was present in the south-most 4 rooms, the north-most 4 rooms and the remaining 4 rooms in-between. Rubber mats were provided in creep area of the crate. To mimic industry standard, heat lamps were provided for piglet comfort up to 48 h post farrow, and lamps were managed as needed thereafter. Three thermostatically-controlled heaters were placed in the chambers allocated to HS sows to supplement heat needed to maintain room the temperature correlated to treatment. The thermostats monitored and recorded exhaust air temperature, which

controlled airflow, air conditioning, and heat supplementation for all chambers. The HS sows were progressively acclimated between d 107 and 114 of gestation to increasing daytime temperature from 21 to 31.5 °C. During lactation, temperature for HS sows gradually increased from 24 to 31.5 °C between 0500 and 1500 and gradually decreased from to 31.5 to 24 °C between 1800 and 0500.

Experimental Diets

Two diets were formulated to contain 17.16 (Control), and 11.82 (LCP) % CP using SBM, corn and soybean hulls as protein-bound ingredient sources (Table 7). All diets were formulated to meet standardized ileal digestible (SID) Lys minimum requirement of 0.78%. The requirement was estimated based on a sow weight of 225 kg post farrowing and nursing 10 piglets with an ADG of 260 g over a 21-d lactation period (NRC 2012). The Control diet was formulated using feed ingredients as sole source of AA. The LCP diet contained less soybean meal than Control diet and was supplemented with CAA to meet amino acid requirements; including L-Lys, L-Val, L-Thr, and L-Trp, DL-Met, L-Ile, and L-Phe (Table 7). Diets were isocaloric and contained similar concentration of fermentable fiber (Tables 8 and 9). A subsample was collected after mixing and shipped to the Agricultural Experiment Station Chemical Laboratories (University of Missouri-Columbia, Columbia, MO) for AA and N analysis (AOAC Official Method 982.30 E [a, b, c], 45.3.05, 2006 and AOAC Official Method 990.03, 2006, respectively) to verify accuracy of feed mixing. The calculated CP and AA concentrations of each diet was similar to the analyzed values (Table 9). Titanium oxide was included as an indigestible marker.

Diets were provided 8 d (± 2) prior to the expected parturition date and throughout the 21-day lactation period. Sows were fed 1 kg of allocated experimental diet twice daily pre-

partum. After parturition, sows were fed 3 times daily (0700, 1500, and 2300) in equal amounts. The daily feed allotment was according to NRC (2012) model, and consisted in 2.301 kg on d 1 and slowly increasing to 7.520 kg on d 21 to achieve an average daily feed intake of 6 kg over the entire lactation period.

Sow Lactation Performance Data Collection

Sows were weighed prior to the morning feeding on days 1, 6, 18, and 21. Piglet weights were recorded on d 1, 3, 6, 9, 12, 15, 18, and 21 of lactation. Feed intake (as disappearance) was determined daily. Ultrasound back fat P2 measurements (RENCO® LEAN-MEATER® Renco Corporation, Minneapolis, MN) were taken on d 1 and d 21.

Sow Health Observations

Twice daily (0500, 1400), sows and piglets were observed and sow rectal body temperature, heart rate, and respiration rate were manually collected and recorded.

Milk Sampling

Milk samples were collected on d 3 and 7 (Early Lactation) and on d 15 and 19 (Peak Lactation**).** Milk samples (100 mL) were manually collected from all glands on d 4 and 16. Approximately 50 mL was submitted to Universal Lab Services, LLC (Northstar Cooperative Inc., Lansing, MI) for determination of true protein, urea N, fat, and lactose concentrations via infrared spectroscopy. The remaining milk samples (approximately 50 mL) were frozen at -20°C for later determination of casein concentration.

For determination of casein concentration, milk samples were thawed overnight in a refrigerator, gently mixed and defatted by centrifuging at 1500 × *g* for 30 min at 4 °C. The fat layer was removed by gentle aspiration using a Pasteur pipette attached to a tube fitted to a low-pressure pump. Once defatted, milk caseins were precipitated by decreasing the milk pH of 4.6

(±1) using 1M HCl. Once precipitated, milk samples were placed in a refrigerator overnight (1.6 °C) and the pH was assessed the following morning. If needed, pH was readjusted between 4.7 and 4.5 with 1M HCl. Samples were centrifuged at 1500 × *g* for 15 min, and the supernatant removed and discarded. The casein pellet was re-suspended in distilled water and centrifuged at 1500 × g for 15 min, the supernatant removed and discarded. This step was repeated once more and the casein pellets were frozen at -20 °C and freeze-dried.

Nitrogen Balance

Nitrogen balance was conducted during early lactation (from d 3 and 7) and peak lactation (between d 15 and19) by total urine collection and fecal grab sampling. A screen-covered steel container was paced below each crate spanning 1.56 m from the back end of the stall to capture sow urine and minimize contribution from piglet urine. Urine was acidified to a pH of less than 3 using H_2SO_4. Twice daily, urine was collected and a subsample of 10% of the total weight of urine collected was obtained, pooled per sow and per day, and stored at 4°C. Fresh and uncontaminated feces were manually collected daily as described by Möhn and de Lange (1998) pooled per sow and per N balance period, and frozen at -20 °C until further analysis. Mass of feed refusals were recorded daily.

Fecal samples were pooled and mechanically homogenized after each N balance period, and a 400-g sample was freeze-dried and ground using a Cyclotec 1093 sample mill (Foss, Hillerød, Denmark). The DM and ash content of freeze-dried feces were determined after drying at 105 °C for greater than 8 h, followed by 5 h combustion at greater than 500 °C in a muffle furnace, respectively (Stocks and Allen, 2013). Urinary N was measured per the Hach method (Hach et al., 1987), and fecal N were measured by combustion method at the Agricultural Experiment Station Chemical Laboratories (University of Missouri-Columbia, Columbia, MO,

LECO; AOAC, 2006; Official Method 990.03). Titanium concentrations in feces and diets were quantified per standard AOAC procedures in duplicate (AOAC, 1997). Absorbance of standards and samples were measured by spectrophotometry (Beckman DU-7400; Beckman Instruments, Inc., Fullerton, CA) at 407 nm.

Nitrogen Balance Calculations

The following equations were used to calculate nitrogen efficiency, nitrogen retention (Möhn and de Lange, 1998), N absorbed (Möhn and de Lange, 1998), and fecal N output (Zhu et al., 2005):

N intake = [feed provided – feed refused] × analyzed N content of diet

N retention = N intake – N excreted in feces – N excreted in urine

Maternal N balance = N intake – N excreted in feces – N excreted in urine – N excreted in milk

N absorbed (Möhn and de Lange, 1998):

N intake – N excreted in feces.

Fecal N output (Zhu et al., 2005):

$$\text{N intake} * [\frac{\text{\% TiO in Feed}}{\text{\% TiO in Feces}} \times \frac{\text{\% N in Feces}}{\text{\% N in Feed}}]$$

Sow milk yield was estimated per NRC (2012) using 20-d litter growth rate, litter size, and a standard lactation curve (NRC, 2012, Eq. 8-71 and 8-72). Nitrogen output with true milk protein was estimated using the analyzed true milk protein concentration and estimated milk yield. Nitrogen utilization efficiency was expressed using two relationships; N retained as a percentage of N intake, and N retained as a percentage of N absorbed.

Measurement of Gases

Rooms were designed to continuously monitor incoming and exhaust concentrations of gases (oxygen, carbon dioxide, and ammonia). Heat production and respiratory quotient (RQ) were calculated from the O_2 and CO_2 exchange. Controlled by software (Lab-VIEW, National Instruments Corp., Austin, TX), sampling in each room occurred over a 15-minute period, the line was purged for 9.5 minutes and data retained for 5.5 minutes (measuring every 0.5 min and averaged over the 11 readings). Sampling time for 12 rooms and a back-ground sample totaled 195-min cycle (7-8 observations per room per day). Ammonia was measured using a chemiluminescence ammonia analyzer (Model 17i, Thermo Fisher, Franklin, MA; 0.001 ppm detection limit), which uses an ozone reaction, measures NO and NO_2 and calculates NH_3, NO and NO_2. CO_2 (2% lower detectable limit) limit and O_2 (1% lower detectable limit) concentrations where quantified through a combination of non-dispersive infrared, ultraviolet, thermal conductivity, paramagnetic oxygen, and electrochemical oxygen analysis (X-Stream X2GP, Rosemount Analytical Emerson Process Management GmbH & Co OHG, Hasselroth, Germany).

Although gas emissions were continuously monitored, only O_2 and CO_2 gaseous exchange were analyzed over the entire lactation. Ammonia concentrations were only analyzed from d 8 to 14 of lactation, as this was the only time frame manure could collect in the pans beneath each sow. All manure pans were emptied on d 3 and 15 to ensure adequate urinary collection for N balance. During the N balance periods, readings of NH_3 were within the error range of the instruments.

Heat Production

Heat production (HP) was determined by indirect calorimetry based on the two Brouwer equations (Brouwer, 1965), which quantifies total heat production (THP), or the energy lost due

to the physical, metabolic, and biological processes, and includes feed consumption, maintenance, thermal regulation and physical activity (NRC, 2012).

$THP = 16.18\ VO_2 + 5.02\ VCO_2$

Where:

VO_2 = Volume of oxygen consumption L/d.
CO_2 = Volume of carbon dioxide production L/d.

Statistical Analysis

Data were analyzed as a randomized complete block design using MIXED and GLM procedures of SAS (SAS Inst. Inc., Cary, NC). Performance data was analyzed using the MIXED procedure with diet, environment, and diet-by-environment as a fixed effects and sow within diet-by-environment-by block as a random effect. Milk data were analyzed using GLIMMIX procedures. The model included diet, phase of lactation, and diet-by-sample as fixed effects, and sow within diet-by-environment-by block as a random effect. All nitrogen balance, heat production, and gaseous exchange data were analyzed as a randomized complete block design using GLIMMIX procedure. The whole lactation model included fixed effects of rep, parity, diet, environment, day, diet-by-environment, parity-by-environment-by-day, and environment-by-day. Chamber, sire of dam, and sow nested within chamber-by-diet-by-environment-by-rep were included as random effects. The ammonia emissions (manure pans allowed to fill) model included rep, diet, environment, day, diet-by-environment, and environment-by-day as a fixed effects and chamber, sire of dam, and sow nested within chamber-by-diet-by-environment-by-rep as random effects. Nitrogen balance by phase of lactation model included rep, diet, environment, phase, diet-by-environment, environment-by-phase, diet-by-phase, diet-by-environment-by-phase as a fixed effects and chamber, sire of dam, sow nested

within chamber-by-diet-by environment-by-rep, and chamber-by-sire of dam-by-sow-by-diet-by-environment-by-rep-by-phase as random effects.

RESULTS

Sow Health Parameters under Heat Stress

Sow body temperature did not differ between TN and HS. Respiration rate was greater ($P < 0.01$) for sows under heat stress (62.1 and 86.9 breaths/min TN and HS, respectively). Heart rate was greater ($P < 0.01$) for HS sows than TN sows (90.4 vs 80.6 beats/min, respectively; Table 10).

Sow and Piglet Performance

Diet did not affect piglet ADG (255 and 259 g Control and LCP, respectively; Table 11). It was less ($P < 0.01$) in HS compared to TN (272 and 241g, respectively; Table 11). Piglet ADG was the only performance variable for which there was an interaction of diet and environment ($P < 0.04$), where the ADG of LCP/TN piglets was greater (P = 0.04) than Control/TN piglets, which was greater than that of the Control/HS and LCP/HS (Figure 3). Dietary treatment did not impact milk production (Table 13). Sows exposed to HS produced a similar amount of milk (7.45 kg/d) when compared to TN (7.09 kg/d) in early lactation, and significantly less milk in peak lactation (9.72 and 8.08 kg/d TN and HS, respectively). Sow body weight loss was similar for the two dietary treatment groups. (-570 and -618 g/d for LCP and Control, respectively; Table 11) Sows exposed to HS lost more weight ($P = 0.03$), compared to Control (-776 vs. -412 g/d respectively). Days for return to estrus was not impacted by diet. Dietary treatment did not affect sow feed intake, but environment did (Figure 4). Sow feed intake was less ($P < 0.01$) for HS sows (4.01 and 5.34 kg/d for HS and TN sows, respectively; Table 11).

Milk Composition

Housing in a hot environment did not affect milk composition, milk fat, true protein, casein, lactose, or MUN (Table 12). Likewise, milk fat, true protein, casein and lactose, during both early and peak lactation, were similar for both dietary treatments (Table 12). However, MUN was less ($P < 0.01$) when dietary CP was reduced (9.21mg/dL and 2.18 mg/dL for Control and LCP, respectively) and was greater in peak lactation ($P < 0.05$), resulting in a diet-by-stage of lactation effect ($P < 0.01$).

Sow Intake over Nitrogen Balance

During the period which N balance was assessed, sow feed intake did not differ by dietary treatment (Table 13). HS sows tended ($P = 0.08$) to eat less (4.24 kg/d) than TN sows (4.78 kg/d) in early lactation. During peak lactation, sow average daily feed decreased ($P < 0.01$) for the HS sows (5.01 kg/d) when compared to TN (6.40 kg/d). Sow total lysine intake was not different between dietary treatments. Total Lys intake tended ($P = 0.08$) to decrease for the HS sows (38.56 g/d) when compared to TN (43.53 g/d) in early lactation. During peak lactation, average Lys intake was significantly less ($P < 0.01$) for the HS sows (45.54 g/d) when compared to TN (58.25 g/d; Table 13).

Sow Nitrogen Utilization

Average daily N intake was less ($P < 0.01$) in the LCP diet (102.13 g/d) for the whole lactation when compared to Control (138.06 g/d; Table 13). When compared to TN, HS treated sow's N intake was reduced (131.56 and 108.63 g/d TN and HS, respectively). Nitrogen excretion was not impacted by environmental temperature and decreased ($P < 0.01$) for LCP sows when compared to control (55.23 and 27.55 g/d Control and LCP, respectively). Fecal N output was significantly decreased ($P < 0.01$) for the LCP diet (11.66 g/d) when compared to

Control (17.38 g/d). Increased environmental temperature reduced fecal N output ($P < 0.05$) for HS sows (12.94 g/d) when compared to control (16.10 g/d). Urinary N output was not impacted by environmental temperature. Urinary N decreased ($P < 0.01$) for the LCP fed sows (16.16 g/d), when compared to Control (37.91g/d). Reduction in dietary CP tended ($P = 0.06$) to decrease N retention for the LCP fed sows (73.94 g/d) when compared to control (83.25g/d) Sows exposed to HS retained less ($P < 0.01$) N (70.29 g/d) when compared to TN (86.903 g/d). Sows fed LCP absorbed less ($P < 0.01$) N (90.38 g/d) when compared to Control (120.62 g/d). Sows exposed to HS environment absorbed less ($P < 0.01$) N (95.62 g/d) than sows exposed to TN environment (115.37 g/d). Urine production was not impacted by diet or environmental temperature. Nitrogen retained as a percent of N intake was not impacted by environment. Nitrogen retained as a percent of N intake was greater ($P < 0.01$) for the LCP diet (71.62%) when compared to Control (59.12%). Nitrogen retained as a percentage of N absorbed was greater ($P < 0.05$) in the LCP diet (80.64%) when compared to Control (67.38%). Maternal N balance was not impacted by diet, and tended ($P = 0.06$) to be less in the sows exposed to HS (11.1g) when compared to TN (22.6 g; Table 13).

Whole Lactation Heat Production and Gaseous Exchange

Oxygen consumption, carbon dioxide elimination, respiratory quotient, and total heat production (HP) over the entire lactation were not impacted by reducing CP (Table 14). Carbon dioxide emission was less ($P < 0.01$) for sows exposed to HS (6187.37 g/d) when compared to sows exposed to TN (7038.06 g/d). Oxygen intake was not impacted by diet or environment. Airflow did not differ between dietary or environmental temperature treatments. Humidity was greater ($P < 0.01$) in the TN chambers (59.07%) when compared to the HS sow rooms (40.17%).

Respiratory quotient was greater ($P < 0.01$) for sows in a TN environment (0.55) when compared to HS (0.45). Total heat production was not impacted by environmental temperature (Table 13).

Ammonia Emissions

Environmental temperature did not impact ammonia emissions. Air in the chambers of sows fed the LCP diet had less ($P < 0.01$) ammonia concentration than the air in the chambers of sows fed the Control diet (11.79 g/d and 30.98 g/d, respectively; Table 15).

DISCUSSION

Sow Health Parameters under Heat Stress

The sows biological stress response to heat was evident in an increased respiration rate, nearly 30% greater. This was a similar observation to that reported previously by Pérez Laspiur et al. (2001). As the temperature increased throughout the day, the respiration rate increased significantly nearly an additional 20% in early lactation. The respiration rate of the heat stress sows was not greater than TN sows at peak lactation, as upwards of 90 breaths per minute seems to be a biological maximum, which the HS sows achieved in peak lactation around the clock. Pérez Laspiur et al. (2001) reported a nearly 1 °C increase in sow rectal body temperature, which was not observed in this study. Black et al. (1992) reported sharp increase in deep body temperature from 39.2 ± 0.1 °C to 40.6 ± 0.1 °C on the first day after ambient temperature changed from 18 °C to 28 °C, a return to 39.5 ± 0.1 °C on day two and remained constant thereafter and a similar response in growing pigs (Giles and Black, 1991). Biologically, this observation is supported Huynh et al. (2005), who concluded that when a sow experiences

effective ambient temperatures higher than ~22 °C, defined as the upper critical temperature, the animal must begin making a series of metabolic and physiological changes to minimize the production of more heat, facilitate convection, and (or) employ evaporative processes to lose heat and maintain homeostasis (body temperature). Chamber humidity was significantly less for the sows exposed to the hot environment when compared to a thermoneutral environment and was likely caused by electric supplemental heaters and similar air flow required to maintain the hot environment. The difference in humidity is not likely to have impacted the sow's response to her environment, as supported by Heitman et al. (1949) who reported little difference in the performance of hogs weighing over 200 pounds to relative humidities of 30 and 94 percent.

Sow and Piglet Performance

The sows exposed to the HS treatment exhibited typical signs of heat stress as shown by their higher respiration rate, lower feed intake and litter gain (300 g), and higher body weight loss. The nearly 24% feed intake depression in this study was similar to the differences reported by Quiniou and Noblet, 1995; Johnston et al., 1999; Pérez Laspiur and Trottier, 2001; Pérez Laspiur et al., 2006, who found high environmental temperatures negatively impacting feed intake from 20 to 55% in lactating sows. This decrease in sow intake likely contributed to the reduced milk yield of 1.64 kg/d and the reduced piglet ADG by 31g/d. The difference in ADG observed in this study was greater than the 12g/d piglet gain difference reported by et al. (2001) when comparing environmental temperatures. This discrepancy may be explained by the different environmental temperatures controlled in our studies. The Control treatment used by Pérez Laspiur and Trottier (2001) was 3 °C lower than the Thermoneutral treatment used in this study, and the hot environment was 1 °C less as well. As the HS sows utilized in this experiment were subjected to a hot environment prior to farrowing, the greater difference in piglet ADG

observed may also have been impacted by the hot environment in late lactation. Omtvedt et al. (1971) reported that piglets of gilts exposed to hot environments during gestation tended to be born smaller than those in a thermoneutral environment. Machado-Neto et al. (1987) reported increased cortisol and reduced antibody concentrations in piglets of sow's heat stressed 2 weeks prior to farrowing, suggesting the piglets receive decreased passive immunity, which would be compounded by the reduction in milk production in lactation.

The only performance variable impacted by diet was piglet ADG, where the LCP diet outperformed the Control by 14 g/d under the TN environment. Under HS, piglet ADG of LCP diet was not different from Control. This was somewhat surprising as we had expected differences between diets to be exacerbated under HS environment. This research group has previously reported a numerical increase in piglet ADG in sows fed reduced CP diet, not significant differences (Chapter 3 and Huber et al., 2015). Recognizing the small sample size and lack of previous research to support this difference, the improvement in ADG affirms there is opportunity in potentially reducing the diet to a lower CP, provided there are data to support clear requirements of next limiting amino acids.

One of the outcomes of this experimental design was the reduction of feed intake experienced by the HS treated sows during peak lactation that resulted in decreased Lys intake by nearly 5 g/d. Although the impact of heat stress on AA requirement is unclear, sows exposed to HS were fed below that recommended NRC (2012). The potential for an AA deficiency may be the driving factor behind some of the differences in performance and N balance. This makes it difficult to assess what may be an impact of heat stress and what may be an impact of moderate AA deficiency, and speaks to the radically different demands on the sow in peak lactation versus early lactation.

Milk Composition

In our previous work (Chapter 3 and Huber et al., 2015), using CAA as a partial replacement for CP reduced milk urea-N and increased the casein fraction of milk composition. Here, casein and true protein were not impacted by diet, however the remarkable reduction in MUN may have minimized the metabolic drain on piglets allowing more partitioning of energy to gain. The diet-by-stage of lactation effect observed in MUN was caused by an increase in MUN for the Control fed sows in peak lactation not observed in the LCP fed sows. Peak lactation demands more of the sow, resulting in increased feed intake, and the excess N of the control diet drives greater urea production.

Sow Nitrogen Utilization and Balance

Reducing dietary CP did not impact maternal N balance. Numerically the sows fed LCP diets and exposed to HS were the only treatment in a negative balance in early lactation, which could be attributed to the decreased feed intake. All treatments in peak lactation were in a positive N balance, despite average weight loss of over 0.5 kg/d across all treatments, and non-significant changes in backfat depth, are indicative of sows catabolizing some lean protein. Understanding the error in both mathematical estimation and empirical body measurements, these conflicting results suggest that one of the goals of this experiment was achieved: to feed sows at their amino acid requirement as defined by a N balance near zero.

Nitrogen retained after fecal and urinary output was over 9 g less for the LCP fed sows. They were 12% more efficient in retaining N as a percent of N consumed (138.06 and 102.31 g/d, Control and LCP, respectively). Sows fed reduced CP diets consumed less nitrogen, excreted less urinary and fecal N, and produced equivalent amounts of milk protein N, but were not different in maternal N balance. These results are supported by previous studies in our lab,

which suggests improvement in mammary AA utilization and casein yield in sows fed diets with improved AA balance profiles (Pérez Laspiur et al., 2009; Manjarín et al., 2012).

HS sows consumed less N, excreted the same amount of N, and produced the same amount of milk protein N, and tended to have a decreased maternal N balance. This suggests that the differences in N retention is an impact of the reduced feed intake, not a direct influence of heat the nitrogen utilization in the lactating sow. Both HS and TN sows remained in a positive N balance. Nitrogen efficiency was not different between environmental treatments.

Ammonia Emissions

The nearly 3-fold decrease in ammonia production when dietary CP was reduced, was expected as nitrogen excretion decreased, leaving less urea available for ammonia formation. It is well documented in growing swine that a reduction in the dietary CP by using supplemental crystalline amino acids (Kerr et al., 2003, Otto et al., 2003a) decreased N concentrations in the manure. Otto et al. (2003a and 2003b) supplemented CAA and observed a 1.2 to 2 g reduction in urinary N excretion per day for each percentage unit reduction in dietary CP. With the replacement of CP with CAA, Li and others (2011) reported a reduction in NH_3 and H_2S emissions from growing pigs.

Whole Lactation Heat Production and Gaseous Exchange

The heat production values observed in this experiment are well within the great range of HP values reported by others for sows and litters. Van den Brand et al. (2000) reported values range from 1.03 to 1.17 mJ/$BW^{0.75}$ • d, Theil et al. (2004) observed values from 0.679 to 0.692 mJ/$BW^{0.75}$ • d, and Stinn et al. (2014) documented values from 0.865 to 1.14 mJ/$BW^{0.75}$ • d. The lack of a diet-by-environment interaction of the total HP, although unanticipated, is supported by the growing pig calorimetry work of Kerr et al. (1998). In this study, total HP was evaluated for

crossbred barrows in a 2 × 3 factorial arrangement of two environmental temperature treatments (thermoneutral [23°C] or heat stress [33°C]) and three diets (control [16% CP], negative control [12% CP], and a 12% CP diet supplemented with crystalline Lys, Trp, and Thr). The reduced CP diet with supplemented with CAA produced a significantly lower HP when compared to control in the thermoneutral environment, under heat stress the HP of all dietary treatments was significantly reduced, resulting in no significant diet-by-environment interaction. Per the data collected for this paper, reduction in CP did not impact HP regardless of environmental temperature and supports the conclusions of Kerr et al. (1998) that under heat stress pigs biologically-reduce total HP to an absolute minimum, regardless of CP concentration, and the influence of reduced CP is not additive. Kerr's conclusions may also apply to the lactating sow, in part, as her metabolic demands may change the dynamics impacting HP. The lactating sow experiences metabolic and behavioral changes that are very different form growing swine with intakes rapidly approaching 9 kg, dramatically increased water intake, and providing the sole nourishment of 10 growing piglets for upwards of 3 weeks. These modifications increase HP and suggest that the sow cannot decrease her HP in this "overdrive" state. Another factor potentially impacting the lack of dietary effect on HP may be that the sows were limiting in an AA, reducing the uptake of other AA and adding to her metabolic load of waste N. Sows in experiment 1 (Chapter 3) were fed the same Control and LCP diets and the serum increase in Lys (first limiting AA) coupled with the reduction of serum Ile (next limiting AA), which suggests a higher dietary requirement for Ile than modeled. Under the assumption that Ile is limiting, it is possible that there are performance and efficiency opportunities that were not optimized.

Under heat stress, the lactating sows did not decrease HP, as expected, which is challenging to explain. In growing pigs, it is known that HP decreases with heat stress by as

much as 15% of fasting heat production with concurrent reduced feed intake (Nienaber et al., 1987; Huynh et al., 2005). van Milgen et al. (1998) reported that reduced physical movement accounted for as much as 10% of the pig's reduced fasting heat production. This was supported by Brown-Brandl et al. (2000), who reported the HP in growing swine with a HS induced reduction in intake of 13% to be significantly lower than that of pigs subjected to the same feed reduction in thermoneutral environment, suggesting that the activity level of the pigs subjected to HS plays a greater role than feed intake on HP. Brown-Brandl (2000) also reported that the thermoneutral treatment groups spent significantly less time lying than the heat stress treatment groups ($P < 0.05$), suggesting that the lower tissue accretion may play an important role in the HP of heat stressed swine. The HS sows failing to decrease total HP could be, in part, due to the lack of increase in urinary N of HS sows, as an increase in urinary N would decrease HP. The lack of difference between the environmental temperatures in total HP could also be related to the mobilization of body stores from the HS sows. It was observed that HS sows lost more weight, but not more backfat, that suggests lean protein catabolism, which would add to the heat production, however HP values have not been published for the heat stressed sow.

Although limited, reported lactating sow RQ's typically range from 0.8 to 1.0 (Theil et al. [2004] 1.01; Stinn et al. [2014] 0.96 to 1.1; Noblet and Etienne [1987] 0.88 to 1.03); substantially greater than those recorded here. We do not know why our RQ's are outside of this range. It is difficult to explain as none of the previously published calorimetry experimentation of the lactating sow (Theil et al., 2004; Stinn et al., 2014; Noblet and Etienne, 1987; Van den Brand et al., 2000) report all of these variables: CO_2 elimination, O_2 consumption, RQ, and total HP. Studying the various values from published works, the data collected in our experiment were similar to individual values, from different reports, of CO_2 elimination and total HP for the

lactating sow. The data collected in this study treated the sow and litter as one unit, which makes it difficult to confirm our values of calorimetry and gaseous exchange when most of data reported attempts to separate the sow and litter. In the present study, CO_2 elimination of the sow and piglets over the whole lactation ranged from 3599.85 to 3144.71 L/d, which is greater than the additive CO_2 production of a lactating sow (1705 L/d calculated from HP and RQ values of Noblet and Etienne, 1987) and of the 10 piglets (93.7 L/d of each piglet reported by Theil et al., 2007) of 2565 L/d. This discrepancy was expected as the data collected for this trial was collected over the whole lactation with sows and piglets of a greater mass of 260 (±10) while Noblet and Etienne (1987) sows weighed 174.5 to 175.7 kg and a litter of 10 piglets reported by Theil et al. (2007) weighed 33.2 to 48.2 kg, for a total of 207.7 to 222.7 kg. In our trial O_2 consumption of the sow and piglets over the whole lactation ranged from 6680.04 to 7311.28 L/d, which is greater than the additive O_2 production of a lactating sow (1656 L/d calculated from HP and RQ values of Noblet and Etienne, 1987) and of the 10 piglets (106.4 L/d of each piglet calculated from CO_2 production, assuming an RQ of 0.90, Theil et al., 2007) of 2697 L/d. The far greater O_2 utilization in this trial is difficult to explain with the very little published data available for the lactating sow and litter, but may be, in part, explained by greater mass of the sows and piglets in this trial. That there was no effect of diet on RQ is not a surprise as the diets did not differ greatly in the amount of lipid. The smaller RQ we observed in the hot environment was anticipated, as CO_2 elimination was decreased for the sows exposed to heat stress. This is likely due to the reduction observed in intake, as the greatest fraction of carbon eliminated in the form of CO_2 originated in the diet.

This experiment demonstrated that neither reduction in CP or HS impacted total HP, two responses that seem to be unique to the lactating sow. For the producer, this suggests more needs

to be done to alleviate heat from the lactating sow, while balancing the micro environments of the piglets to maintain piglet thermoneutrality. Much more data needs to be collected on a larger sample size to confirm if the principle that total HP of the lactating sow is increased in hot environments. In terms of dietary reduction in CP, more research is needed to better understand the order of limiting AA and their interactions in the lactating sow. The decrease in ammonia production has substantial environmental importance, and would support the improvement in N efficiency. The reduction in MUN and urinary N while maintaining the true milk protein fraction of the milk, also suggests the animal is more efficient as less N is wasted, but this efficiency must not be great enough to reduce the added heat production caused by heat stress.

APPENDIX

Table 7. Ingredient composition of diets.

Item	Control	LCP
Corn	61.64	65.57
Soybean meal, dehulled, solvent extracted	25.20	11.73
Choice white grease	3.82	4.77
Sugar food by-product[1]	5.00	5.00
Soybean Hulls	-	7.50
L- Lys·HCl	-	0.41
L-Val	-	0.21
L-Thr	-	0.15
L-Trp	-	0.04
DL-Met	-	0.06
L-Phe	-	0.07
L-Ile	-	0.04
Limestone	1.45	1.38
Mono calcium phosphate	1.60	1.78
Vitamin premix[2]	0.25	0.25
Mineral premix[3]	0.125	0.125
Sow pack[4]	0.25	0.25
Se 270[5]	0.0675	0.0675
Salt	0.50	0.50
Titanium oxide	0.10	0.10
Total	100.00	100.00

[1] National Ingredient Corporation: CP 1.00 %; NE = 2719 kcal/kg (estimated using ME equation of Noblet et al. (2003)); fermentable fiber 0.05 %.

[2] Vitamin Premix provided the following per kg of diet: 3,000 IU vitamin A, 300 IU vitamin D_3, 20 IU vitamin E, 1 mg menadione (vitamin K), 20 µg vitamin $B_{12,}$ 4 mg riboflavin, 10 mg D-pantothenic acid, 15 mg niacin.

[3] Mineral Premix provided the following per kg of diet: 640 mg Fe (as $FeCO_3$), 260 mg Zn (as ZnO), 36 mg Mn (as MnO_2), 20 mg Cu (as $CuCl_2$), and 0.58 mg I (as ethylenediamine dihydroiodide).

[4] Sow Pack provided the following per kg of diet: 0.10 mg biotin, 250 mg choline (as choline chloride), 0.75 mg folic acid, 2.3 mg vitamin B_6 (as pyridoxine·HCl), 10 IU vitamin E (as DL-tocophorol acetate), 90 µg chromium (as chromium picolinate), and 23 mg carnitine (as L-carnitine).

[5] Cargill: Se 270 mg premix, sodium selenide.

Table 8. Calculated nutrient composition of diets.

Item	Control	LCP
Net energy (NE) kcal/kg	2582	2582
CP, %	17.16	12.56
Fermentable fiber, %	10.58	10.22
Total P, %	0.68	0.65
Standardized total digestible P, %	0.74	0.44
Carbon, %	38.09	38.17
Ca, %	0.89	0.89
SID[2] CP, %	14.57	9.74
SID AA		
Arg, %	1.02	0.63
His, %	0.41	0.28
Ile, %	0.62	0.43
Leu, %	1.32	0.96
Lys, %	0.78	0.78
Met, %	0.24	0.23
Met + Cys, %	0.48	0.41
Phe, %	0.74	0.56
Phe-Tyr, %	1.22	0.81
Thr, %	0.53	0.49
Trp, %	0.18	0.15
Val, %	0.68	0.66
SID Lys/NE, g/Mcal	3.013	3.03

[1] Nutrient values were calculated using NRC (2012).

[2] Standardized Ileal Digestible.

Table 9. Calculated and analyzed AA composition of diets, %.

	Control		LCP	
	Calculated	Analyzed	Calculated	Analyzed
Essential				
Crude Protein	17.16	17.33	11.82	12.19
Arg	1.10	1.06	0.69	0.66
His	0.47	0.49	0.33	0.36
Ile	0.71	0.73	0.46	0.52
Leu	1.50	1.51	1.11	1.12
Lys	0.90	0.92	0.88	0.90
Met	0.28	0.25	0.27	0.23
Phe	0.85	0.86	0.64	0.66
Thr	0.64	0.65	0.58	0.57
Trp	0.20	0.21	0.16	0.16
Val	0.80	0.79	0.76	0.75
Non-essential				
Tau	-	0.15	-	0.16
Asp[2]	-	1.70	-	1.09
Ser	-	0.79	-	0.55
Glu[3]	-	2.96	-	2.03
Pro	-	1.01	-	0.77
Gly	-	0.69	-	0.51
Ala	-	0.86	-	0.65
Cys	-	0.26	-	0.19
Tyr	-	0.54	-	0.40

[1] Calculated CP and essential AA values per NRC (2012).
[2] Aspartate + Asparagine.
[3] Glutamate + Glutamine.

Table 10. Sow body temperature, respiration rate and heat rate sows fed reduced CP diets and housed under HS and TN conditions.

	Early lactation							Peak lactation							
	HS		TN		SEM	Env[1]	Time of Day	HS		TN		SEM	Env	Time of day	Stage[2]
	AM	PM	AM	PM				AM	PM	AM	PM				
Core body temp, °C	37	40	39	39	1.1	0.42	0.14	39	36	38	37	1.1	0.42	0.14	0.26
Respiration rate, #/min	76	100	49	68	5	<0.01	<0.01	83	88	53	70	5	<0.01	<0.01	0.13
Heart rate, #/min	85	86	89	66	6	0.02	<0.01	93	85	93	71	6	0.02	<0.01	0.03

[1] Environment (Env)
[2] Stage of lactation (Stage)
[3] Env × stage removed, not significant.
[4] Env × sample removed, not significant.
[5] Stage × sample removed, not significant.
[6] Env × sample × stage removed, not significant.

Table 11. Sow and litter performance of sows fed reduced CP diets and exposed to TN and HS environments.

	Thermoneutral		Heat Stress				
	Control	Low	Control	Low	SEM	Diet	Env[1]
Number of sows	9	9	9	8	-	-	-
Litter size at weaning	9.9	10.0	9.8	9.8	-	-	-
Litter growth, kg/d	2.6	2.8	2.4	2.3	0.1	0.51	<0.01
Piglet ADG[2], g	265[b]	279[a]	244[c]	238[c]	11	0.35	<0.01
Sow BW, kg/d	-0.5	-0.3	-0.7	-0.8	0.19	0.76	0.03
Backfat depth[3, 4], mm, d 1	16.7[a]	12.1[b]	13.4[b]	12.3[b]	0.9	<0.01	0.08
Backfat depth, mm, d 21	11.5	10.7	11.9	10.7	0.7	0.16	0.76
Change in backfat depth, mm, d 1-21	-1.4	-2.7	3.2	-2.1	0.99	0.89	0.51
Return to estrus, d	7.6	6.9	6.6	5.3	1.19	0.37	0.28
Sow feed intake, kg/d	5.17	5.51	3.67	4.33	0.39	0.15	<0.001

[1] Environment (Env)
[2] Diet × environment ($P < 0.04$), differences denoted by superscripts.
[3] Diet × environment ($P = 0.05$), differences denoted by superscripts.
[4] Differences are the result of randomization and allotment.

Table 12. Milk Composition.

Item	Early lactation				Peak lactation				SEM	Diet	Env[1]	Stage[2]
	Thermoneutral		Heat stress		Thermoneutral		Heat stress					
	Control	Low	Control	Low	Control	Low	Control	Low				
Fat, %	9	9.4	9.68	9.81	8.75	8.69	8.37	8.74	0.61	0.52	0.56	0.01
True Protein, %	4.87	4.69	5.13	4.96	4.21	4.08	4.23	4.19	0.16	0.2	0.11	<.0001
Lactose, %	4.95	4.99	4.87	4.92	5.38	5.58	5.61	5.46	0.16	0.77	0.95	<.0001
MUN[4], mg/dl	7.59	2.99	7.90	1.79	10.55	1.87	10.83	1.77	0.9	<.0001	0.77	0.02
Casein[3], %	2.77	2.81	3.50	3.14	2.65	2.45	2.57	2.64	0.36	0.62	0.19	0.04
Casein, % of True Protein	69.13	73.90	81.09	75.46	75.82	73.37	74.95	78.26	8.60	1.00	0.37	0.89

[1] Environment (Env)
[2] Stage of lactation (Stage)
[3] Analyzed on defatted milk and corrected using milk fat concentration
[4] Diet × stage of lactation ($P < 0.01$)

Table 13. Sow nitrogen utilization.

	Early Lactation				Peak Lactation								
	TN		HS		TN		HS		SEM	Stage[2]	Diet	Env[1]	Stage* Env
	Control	LCP	Control	LCP	Control	LCP	Control	LCP					
No. of sows	9	9	9	8	9	9	9	8	-	-	-	-	-
Litter size	9.9	10.0	9.8	9.8	9.9	10.0	9.8	9.8	0.1	1	0.63	0.1	1
Litter gain, kg	2.19	2.12	2.06	2.02	2.88	2.83	2.40	2.30	0.19	<0.01	0.54	0.01	<0.01
Feed intake, kg/d	4.66	4.68	4.00	4.29	6.11	6.46	4.82	4.99	0.35	<0.01	0.46	<0.01	<0.01
Lys intake[3], g/d	42.88	42.11	36.80	38.57	56.19	58.13	44.30	44.91	3.14	<0.01	0.73	<0.01	<0.01
N intake, g/d	129.0	91.0	110.8	83.7	169.1	125.7	133.4	97.4	8.6	<0.01	<0.01	<0.01	0.01
N excretion, g/d	50.8	31.0	49.4	28.1	65.3	27.2	54.4	22.8	4.4	0.32	<0.01	0.12	0.3
Fecal N[7, 9], g/d	14.7	10.0	13.1	11.4	24.9	14.0	16.0	10.4	2.1	0.01	<0.01	0.05	0.03
Urinary N[8], g/d	36.2	21.2	35.8	16.7	40.6	13.5	38.4	12.5	3.3	0.53	<0.01	0.39	0.83
N retention[4], g/d	78.0	60.2	65.2	55.3	103.5	98.7	78.9	74.4	6.5	<0.01	0.08	<0.01	0.05
N absorbed[5], g/d	114.4	81.1	97.6	72.4	144.3	111.8	117.4	87.1	7.5	<0.01	<0.01	<0.01	0.05
Milk, kg/d	7.5	7.3	7.1	7.0	9.8	9.6	8.2	7.9	0.6	<0.01	0.56	0.01	<0.01
True milk protein N, g/d	58.7	56.3	58.9	58.0	59.0	63.3	55.0	53.6	4.1	0.87	0.97	0.27	0.02
Urine weight[8], kg/d	5.8	5.7	5.3	6.3	7.9	6.3	7.2	6.1	0.9	0.01	0.5	0.77	0.62
N retained[4, 8,] % of intake	60.4	66.2	55.7	65.7	60.8	78.8	59.4	75.8	3.2	<0.01	<0.01	0.3	0.93
N retained % of absorbed[5, 9]	68.1	73.9	63.1	75.7	71.0	88.4	67.5	85.0	3.1	<0.01	<0.01	0.26	0.68
Maternal balance, g	19.4	5.0	1.9	-3.7	35.6	20.3	33.4	22.6	0.8	<0.01	0.28	0.06	0.67

[1] Environment (Env)
[2] Stage of lactation (Stage)
[3] Total Lys intake calculated with analyzed total Lys content (%) and sow intake.
[4] N intake – N excreted in feces – N excreted in urine.
[5] N intake – N excreted in feces.
[6] Diet by environment interactions, and diet by phase of lactation by environment were not significant.
[7] Significant effect of rep, $P = 0.01$
[8] Significant stage × diet effect, $P < 0.05$
[9] Tendency of a stage × diet effect, $P = 0.07$

Table 14. Whole lactation air emissions [2, 3, 4].

	TN		HS		SEM	Diet	Environment	Environment *Day
	HCP	LCP	HCP	LCP				
CO_2, g/d	7066.72[a]	7009.43[a]	6201.49[c]	6173.25[bc]	279.42	0.8	< 0.01	< 0.01
O_2, g/d	-9537	-9636.6	-10440	-9988.7	257.64	0.65	0.11	0.23
Air Flow, L/min	8993.63	8540.57	8942.01	9193.96	257.64	0.6	0.19	0.4
Humidity, %	57.87[a]	60.28[a]	40.53[b]	39.82[b]	1.72	0.46	< 0.01	0.05
VCO_2, L/d	3599.85[a]	3570.67[a]	3159.09[b]	3144.71[b]	142.34	0.8	< 0.01	< 0.01
VO_2[5], L/d	-6680	-6749.8	-7311.3	-6996.5	430.14	0.65	0.11	0.23
RQ	0.56[a]	0.55[a]	0.44[b]	0.46[b]	0.02	0.64	< 0.01	0.02
HP[5, 6], KJ/d	126050	126975	134133	128890	7629	0.65	0.3	0.19
HP[6], KJ/$BW^{0.75}$•d	1854.6	1987.1	2081.3	1991.3	115.08	0.8	0.17	0.33
CO_2, L/$BW^{0.75}$•d	53.15[ac]	55.99[a]	49.13[a]	48.94[bc]	1.93	0.38	< 0.01	0.02
VO_2, L/$BW^{0.75}$•d	98.2723	105.66	113.45	108.03	6.53	0.84	0.07	0.33
$BW^{0.75}$, kg[1]	68.52	64.65	64.43	65.6	-	-	-	-

[1] Total metabolic weight of entire chamber, sows and piglets
[2] Day of lactation was significant for all variables.
[3] Parity was not significant for any variables.
[4] Interaction diet × environment was not significant.
[5] Significant effect of rep, $P < 0.05$
[6] Brower (1965) equation = $16.18VO_2 + 5.02VCO_2$

Table 15. Day 8-14 emissions of sows fed reduced CP diets and exposed to TN and HS environments.

	TN		HS		SEM	Replicate	Diet	Environment	Day	Diet* Environment
	HCP	LCP	HCP	LCP						
CO_2, g/d	7318.93	7461.62	6674.31	6388.67	329.11	0.02	0.69	<.0001	<.0001	0.24
O_2, g/d	-9828.77	-10440.00	-11260.00	-10170.00	686.40	<0.01	0.54	0.13	<.0001	0.03
NH_3, g/d	34.29	11.44	27.69	12.13	2.78	0.03	<.0001	0.28	<.0001	0.19

Figure 3. Litter ADG (kg) by treatment over a 21 d lactation period where ADG was calculated as an average between each weight taken on days 1,3,6,9,12,15, 18, and 21, where diet did not impact piglet ADG ($P = 0.35$), environmental impacts of HS decreased ADG ($P < 0.01$), and the ADG of LCP/TN piglets was greater (P = 0.04) than Control/TN piglets, which was greater than piglets exposed to HS, resulting in an interaction of diet and environment ($P < 0.04$).

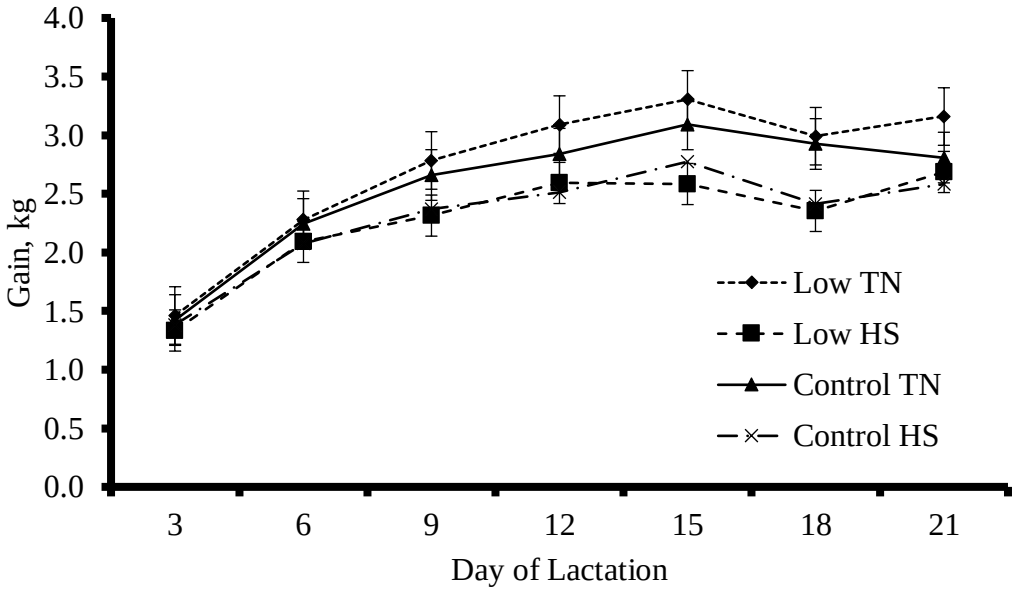
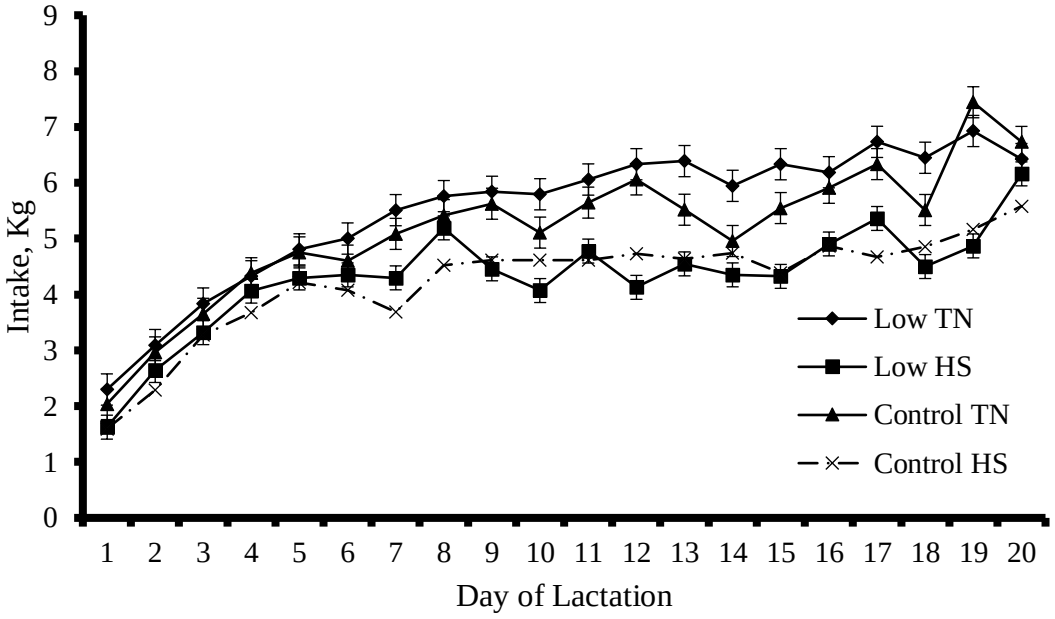


Figure 4. Average daily feed intake (ADFI) of the lactating sow (kg) by treatment over a 21 d lactation measured daily, where dietary treatment did not impact ADFI, and ADFI was less ($P < 0.01$) for HS sows.



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V. CONCLUSIONS

The goals of this thesis were to:

1. Understand the impact of reduced protein diets with CAA supplementation on the performance of the lactating sow.
2. Assess the impacts of reduced protein diets with CAA supplementation on nitrogen balance and ammonia emissions.
3. Quantify the HP of sows under HS
4. Determine if HP could be reduced by reduced protein-CAA diets, mitigating the negative effects of HS.

Feeding reduced CP diets containing CAA to meet the SID requirement of AA did not negatively impact lactation performance; piglet growth and sow feed intake. Milk fat, true protein, and lactose were not impacted by the reduction in CP. In its simplest terms, CAA substitution for CP still met the piglet's needs. Interestingly, CAA supplementation reduced MUN over 2-fold in early lactation and by over 5-fold in peak lactation. The reduced MUN may also hold the potential to ease the metabolic demands of the piglet. Urea is not a utilizable nutrient to the monogastric, and it is assumed the urea bound in milk must be excreted at the expense of energy by the piglet. More needs to be done to better understand the use or non-use of MUN by the nursing piglet.

The potential of reducing CP of diets may be limited by our understanding of AA and minimum N requirement. Serum Lys and the serum concentrations of the next two limiting AA, Thr and Val increased by over 30% as CP was decreased, while Ile was unchanged in early lactation. However, at peak lactation, Thr was not changed, while Val increased 30%, and Ile decreased nearly 35% as CP was decreased. This suggests a higher dietary requirement for Ile

than modeled at peak lactation. It is unknown if reducing N alters the sow's requirement, but it suggests an increased importance of accuracy in estimating the requirements of indispensable AA. Meeting the lactating sow's AA requirements with a standard corn and soybean meal, provides the luxury of overfeeding many essential AA that requirements are not well understood. As economics and nutrient management drive substitution of soybean meal, there needs to be well established AA requirements to meet the demands on the lactating sow.

Total heat production was not decreased with increase in environmental temperature or reduction in CP, suggesting the sow behaves differently than growing pigs. Although difficult to explain, this result emphasizes the extreme expectations placed upon the lactating sow; increase feed intake 70% in 3 to 7 days, dramatically increased water intake, and providing the sole nourishment of 10 growing piglets for upwards of 3 weeks. To the producer, this suggests greater importance on the management to alleviate heat form the lactating sow, while balancing the micro environments of the piglets to maintain piglet thermoneutrality. The negative effects of a hot environment were most clearly seen as a cascade of decreased sow feed intake, reduced milk production, and less piglet growth. The sow's efficiency in using dietary N and milk composition were unchanged. My work reiterates the recommendations of many others, that we must manage the sow's environment to keep her eating.

The decrease in ammonia production, has substantial environmental importance in the reduction of fine particulate matter. Reduction in nitrogen output impacts manure characteristics and environmental risks. I think producers need to better understand manure nitrogen concentration, its balance with phosphorus, and economic analysis of the fertilizer value of the manure when dietary CP is reduced.

I have learned that replacing the crude protein in the diet of the lactating sow, with CAA is a strategy that works well for the sow and litter, reduces cost, improves nitrogen utilization, and is positive for the environment.

I know that feeding CAA in a reduced CP lactation diet is being implemented by modern pork producers despite us not knowing precisely, the lactating sow's AA requirements, the order of limiting AA, and their interactions with one another.

I only saw the tip of iceberg regarding the sow's response to heat stress. There is so much that is unknown. The dietary approach I studied did not reduce the added heat production caused by heat stress.

The impact of feeding CAA in reduced CP lactation diets needs long-term evaluation. Its impact on lifetime productivity remains a significant question in my mind.