RESEARCH REPORT
ANIMAL SCIENCE

Title: Improving energy utilization in high by-product diets with copper – **NPB #14-070**

Investigators: Drs. Bob Goodband, Kyle Coble, Derris Burnett, John Gonzales, James Usry, Mike Tokach, Joel DeRouchey, Jason Woodworth, Steve Dritz, Josh Flohr, and Matt Vaughn

Institution: Kansas State University

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Industry Summary:

The overriding objective is to determine if energy utilization from diets containing high levels of by-products and differing in energy content can be improved with supplemental Cu in commercial finishing pigs. In order to accomplish the overall objective, an intensive field experiment was conducted to determine the effects of added Cu and diet type (energy and fiber) on growth performance, carcass characteristics, economics, energy digestibility, gut morphology, and mucosal mRNA expression of finishing pigs.

Although these data did not demonstrate the large increase in feed intake and gain from feeding added Cu observed in previous experiments, it did provide data on possible modes of action for Cu. Furthermore, it was successful at demonstrating the differences in growth performance when high-fiber, high-energy diets are fed to finishing pigs compared to a simple corn-soybean meal-based diet. It is important to note, that the original proposal intended to have an additional diet type that was high in fiber and low energy; however, due to technical difficulties in the production facility, data integrity for those treatments could not be maintained. Therefore, those treatments were not included in the data set.

Treatments included the main effects of diet type, a corn-soybean meal-based diet (corn-soybean meal-based) or a high byproduct diet (byproduct) with 30% distillers dried grains with solubles (DDGS) and 15% bakery meal, and with or without added Cu (0 or 150 ppm added Cu). Overall, neither added Cu nor diet type influenced growth performance. However, caloric efficiency was decreased for pigs fed the byproduct diet compared to the corn-soybean meal-based diet. One potential reason for this is accurate energy values for bakery meal does not exist and is a potential area for future research needed by producers. Furthermore, pigs fed the byproduct diet had decreased carcass yield and HCW F/G and tended to have decreased HCW and HCW ADG compared to pigs fed the corn-soybean meal-based diet. This is consistent with previous NPB Project #12-167.

Dry matter and gross energy digestibility (GE) during the early finishing period were improved when Cu was added to the corn-soybean meal-based diet, but not in the byproduct diet. During the late finishing period, added Cu increased DM digestibility by nearly 1% and GE digestibility by 3% while pigs fed the byproduct diet had decreased DM and GE digestibility compared to those fed the corn-soybean meal-based diet. This could

potentially explain why Cu may have provided a late finishing response in some previous studies. For gut morphology, pigs fed added Cu had decreased crypt depth in the distal small intestine compared to those fed no added Cu. This could potentially be a sign of improved gut health with reduced turnover intestinal cells over time. Furthermore, relative mRNA expression of intestinal fatty acid binding protein (iFABP) was decreased in pigs fed added Cu compared to those fed no added Cu. A decrease in iFABP of the distal small intestine mucosa of pigs fed added Cu would suggest that the gene responsible for iFABP transcription is possibly down regulated with added Cu. If fat digestibility is truly increased with Cu supplementation, we would expect increased iFABP expression to accommodate the additional end products of fat digestion (Fatty Acids). This makes for an interesting finding demonstrating that Cu does impact intestinal metabolism of dietary fats at the molecular level, unfortunately there is currently very little data available for comparison and further research in this area is warranted

From an economic prospective when measured as income over feed cost (IOFC = gain value/pig – feed cost/pig), statistically there were no differences between diet type nor added Cu. Economics were calculate on a carcass basis because of the known negative impact that high fiber diets have on carcass yield and producer profit. Numerically, when calculated on a constant time basis pigs fed added Cu had a $1.75 advantage over pigs not feed added Cu in the corn-soy diet. For pigs fed the byproduct diet, the advantage for feeding Cu was $0.96/pig. When calculated on a constant weight basis, only a small numerical improvement was observed in the corn-soy diet with added Cu.

Producer bottom line:

- Adding 150 ppm Cu to the diet during the early finishing period tended to increase in ADG, but growth performance for the overall growth study was not influenced by added Cu.
- Pigs fed the byproduct diet compared to the corn-soybean meal-based diet had decreased ADG and ADFI during the early finishing period, but diet type did not affect overall growth performance even though pigs fed the byproduct diet had a reduction in carcass yield and HCW.
- Dry matter and energy digestibility are influence by diet type and Cu may provide a means for improving that; however, more research is need to determine the typical amount of improvement that can be observed
- Added Cu provided a numerical improvement in IOFC on both a constant weight and time basis.

Dr. Bob Goodband
Kansas State University
Department of Animal Science and Industry
242 Weber Hall
Manhattan, KS 66506
785-532-1228
Goodband@ksu.edu

Dr. Kyle Coble
New Fashion Pork
Jackson, MN 56143
507-847-4610
kcoble@nfpinc.com

Dr. Derris Burnett
Mississippi State University
Department of Animal and Dairy Sciences
334-552-0881
ddb362@msstate.edu

Key words:

Copper, energy digestibility, byproduct, yield, growth, morphology, gene expression

Scientific Abstract:

A total of 757 pigs (PIC 337 × 1050; initially 60.8 lb BW) were used in a 117-d experiment to determine the effects of added Cu (TBCC; tribasic copper chloride, IntelliBond C; Micronutrients, Inc., Indianapolis, IN) and diet type on growth performance, carcass characteristics, energy digestibility, gut morphology, and mucosal mRNA expression of finishing pigs. Pens of pigs were allotted to 1 of 4 dietary treatments, balanced on average pen weight in a randomized-complete block design with 26 to 28 pigs per pen and 7 replications per treatment. Treatments were arranged as a 2 × 2 factorial with main effects of diet type, a corn-soybean meal-based diet (corn-soybean meal-based) or a high byproduct diet (byproduct) with 30% distillers dried grains with solubles (DDGS) and 15% bakery meal, and with or without added Cu (0 or 150 ppm added Cu).

There were no Cu × diet type interactions for growth performance. Overall, neither added Cu nor diet type influenced growth performance. However, caloric efficiency was improved ($P = 0.001$) for pigs fed the byproduct diet compared to the corn-soybean meal-based diet. Pigs fed the byproduct diet had decreased carcass yield ($P = 0.007$) and HCW F/G ($P = 0.013$), and tended to have decreased HCW ($P = 0.067$) and HCW ADG ($P = 0.056$) compared to pigs fed the corn-soybean meal-based diet.

A Cu × diet type interaction ($P < 0.05$) existed for DM and GE digestibility during the early finishing period as added Cu improved digestibility of DM and GE in the corn-soybean meal-based diet, but not in the byproduct diet. During the late finishing period, added Cu increased DM and GE digestibility ($P = 0.060$) while pigs fed the byproduct diet had decreased DM and GE digestibility ($P = 0.001$) compared to those fed the corn-soybean meal-based diet. For gut morphology, pigs fed added Cu had decreased crypt depth ($P = 0.017$) in the distal small intestine compared to those fed no added Cu. Furthermore, relative mRNA expression of intestinal fatty acid binding protein (iFABP) was decreased ($P = 0.032$) in pigs fed added Cu compared to those fed no added Cu.

In summary, adding 150 ppm added Cu or including 30% DDGS and 15% bakery meal into a corn-soybean meal-based diet did not influence growth performance. However, HCW ADG and HCW G/F was reduced in pigs fed the byproduct diet compared to those fed the corn-soybean meal-based diet. Only minor differences in gut morphology or mRNA expression were observed from pigs fed diets with high levels of Cu or byproducts compared to those fed a corn-soybean meal-based diet.

Introduction

For many years, copper (Cu) has been supplemented in nursery and early finishing diets to improve growth performance. While feeding high levels of Cu has been shown to improve growth, the duration and degree of response has not always been consistent. Research has typically shown that added Cu impacts growth the most during the early finishing period but not late finishing (Davis et al., 2002[1]; Hastad, 2002[2]). Recently, Coble et

[1] Davis, M.E., C. V. Maxwell, D. C. Brown, B. Z. de Rodas, Z. B. Johnson, E. B. Kegley, D. H. Hellwig, and R. A. Dvorak. 2002. Effect of dietary mannan oligosaccharides and (or) pharmalogical additions of copper sulfate on growth performance and immunocompetence of weanling and growing/finishing pigs. J. Anim. Sci. 80:2887-2894.

[2] Hastad et al., Swine Day 2001, Report of Progress 880, Pages 111-117.

al. (2014[3]) reported that adding 150 ppm Cu in finishing diets tended to increase ADFI and improve F/G during the late finishing period.

It has been postulated that the growth-promoting effects of Cu are partly due to its impact on tissue repair in the small intestine and its ability to stimulate the synthesis of digestive enzymes, resulting in a better digestion and absorption of nutrients (Hedemann et al., 2006[4]). Lou and Dove (1996[5]) report that nursery pigs fed 250 ppm Cu had improved fat digestibility. Rochell et al. (2015[6]) reported an improvement in AA digestibility in low Lys diets with added Cu in chicks, and Gonzales-Eguia et al. (2009[7]) reported an improvement in fat digestibility with added Cu in 66 to 132 lb growing pigs. Although the strategies for using Cu have not changed much over the years, the types of diets that are used in commercial production are different in ingredient composition from diets utilized in the original research with Cu. It has yet to be investigated if ingredient usage and diet formulation are an important factor to consider when adding Cu to improve growth performance. As a result, this study sought to investigate possible response criteria to potentially explain addition ways Cu can improve growth.

Objectives:

1) Determine the effects of added Cu and diet type on growth performance, carcass characteristics, energy digestibility, gut morphology, and mucosal gene expression of finishing pigs.

Materials & Methods

The Kansas State University Institutional Animal Care and Use Committee approved the protocol used in this experiment. The experiment was conducted in a commercial research facility in southwestern Minnesota. The facility was double-curtain sided with completely slatted concrete flooring. The barn contained 48 pens with 26 to 28 pigs (similar number of barrows and gilts) in each, equipped with a 4-hole conventional dry self-feeder (Thorp Equipment, Thorp, WI) and a cup waterer providing ad libitum access to feed and water. A computerized feeding system (FeedPro; Feedlogic Corp., Willmar, MN) delivered and recorded daily feed additions of specific diets to each pen.

A total of 757 pigs (PIC 337 × 1050; PIC, Hendersonville, TN; initially 60.8 lb BW) were used in a 117-d experiment. Before d 0, all pigs were fed a common diet with 205 ppm Cu from tribasic copper chloride (TBCC, Intellibond C; Micronutrients, Inc., Indianapolis, IN). On d 0, pens of pigs were weighed, ranked by average pen BW, and allotted to 1 of 4 dietary treatments in a 2 × 2 factorial arrangement with average pig BW balanced across each treatment. There were 7 replications per treatment. Treatments included 2 diet types, a corn-soybean meal-based-based diet (corn-soybean meal-based) or a high byproduct diet with 30% distillers grain with solubles (DDGS) and 15% bakery meal (byproduct), with or without added Cu (0 or 150 ppm; Tables 1 to 3).

All diets contained a basal level of 17 ppm added Cu from CuSO4 provided by the trace mineral premix. Treatment diets were fed in 5 dietary phases in meal form and formulated on a standardized ileal digestible (SID) Lys basis to meet or exceed requirements (NRC, 2012). Diets were balanced on a SID Lys:NE ratio

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[3] Coble et al., Swine Day 2013, Report of Progress 1092, pages 168-180.
[4] Hedemann, M. S., B. B. Jensen, and H. D. Poulsen. 2006. Influence of dietary zinc and copper on digestive enzyme activity and intestinal morphology in weaned pigs. J, Anim. Sci. 84:3310-3320.
[5] Lou, X. G., and C. R. Dove. 1996. Effect of dietary copper and fat on nutrient utilization, digestive enzyme activities, and tissue mineral levels in weanling pigs. J. Anim. Sci. 74: 1888-1896.
[6] Rochell, S., T. Parr, J. Usry, C. Parson, and R. Dilger. 2015. Effects of dietary amino density and tribasic copper chloride supplementation in *Eimeria acervulina*-infected chicks. International Poultry Science Forum. M106 (Abstr.)
[7] Gonzales-Equia, A., C. Fu, F. Lu, and T. Lien. 2009. Effects of nanocopper on copper availability and nutrients digestibility, growth performance and serum traits of piglets. Livestock Sci. 126:122-129.

across all treatments within phase to insure Lys was not a limiting factor for growth. Nutrient values for the ingredients were based on the NRC (2012), with the exception of the DDGS. The NE value (1,194 kcal/lb) for DDGS were calculated based upon the oil content, as described by Graham et al. (2014[8]). Samples of each diet were collected during each phase from multiple feeders 2 d after the beginning of a phase and 2 d before ending a phase. The 2 samples were combined to form a composite sample for each treatment within each phase and analyzed, in duplicate (Table 4).

Pens of pigs were weighed and feed disappearance was recorded approximately every 3 wk to determine ADG, ADFI, F/G, and caloric efficiency on and ME and NE basis. Caloric efficiency was calculated by dividing the sum of total feed intake and diet calorie content, by total gain. On d 94, the 3 heaviest pigs in each pen were weighed and sold according to standard farm procedures. These pigs were used in calculation of pen growth performance, but not carcass characteristics. Prior to marketing, the remaining pigs in the barn were individually tattooed with a pen identification number to allow for carcass measurements to be recorded on an individual basis. On d 117, final pen weights were taken and feed disappearance was recorded. A subsample of two gilts per pen, representing the mean individual weight of the pen, were transported 67 miles to a commercial packing plant for processing, intestinal sampling, and data collection (Packing Plant #1; Natural Foods Holdings, Sioux Center, IA). All remaining pigs were transported 59 miles on d 118 to a commercial packing plant (Packing Plant #2; JBS Swift and Company, Worthington, MN) for processing and carcass data collection. Hot carcass weight was measured immediately after evisceration and each carcass evaluated for carcass yield, backfat depth, loin depth, and percentage lean.

Carcass yield was calculated by dividing the HCW at the plant, by the live weight at the farm before transport. Fat depth and loin depth were measured with an optical probe inserted between the third and fourth last rib (counting from the ham end of the carcass) at a distance approximately 7 cm from the dorsal midline. An assumed yield of 75% was used to calculate initial HCW at the beginning of the experiment. Hot carcass weight ADG was calculated by subtracting initial HCW from the final HCW obtained at the plant, then divided by 117 d on test. Hot carcass weight F/G was calculated by dividing HCW gain by feed intake over the 117 d experiment.

Feed and fecal grab samples were collected from each pen over a 2 d period during phases 2 (d 25 to 26) and 4 (d 74 and 75) to determine DE content of the experimental diets. Acid insoluble ash (Celite 545, Univar Inc., Redmond, WA) was included in the diet at 1.0% for 9 d at the beginning of the phase 2 and 4 to allow for a 7 d adaptation period and 2 d collection period. Feed and fecal samples from each pen and day were dried at both 50°C and 100°C, using a two-step drying process. Samples across days within pen were then pooled together for a composite sample for analysis. To determine the acid insoluble ash content of the feed and feces, samples were heated to 600°C for 18 h, digested in 2 N HCL for 5 min, filtered, and heated again to 600°C for 18 h, as explained by Atkinson et al. (1984[9]). Gross energy values for the feed and feces were determined by oxygen bomb calorimetry (Model 1341EB, Parr Instrument Company, Moline, IL), according to Galyean (2010[10]). Dry matter and GE digestibilites were calculated using the index method, according to Adeola (2001[11]), using the following equation where AIA is acid insoluble ash:

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[8] Graham, A. B., R. D. Goodband, M. D. Tokach, S. S. Dritz, J. M. DeRouchey, S. Nitikanchana, and J. J. Updike. 2014a. The effects of low-, medium-, and high-oil distillers dried grains with solubles on growth performance, nutrient digestibility, and fat quality in finishing pigs. J. Anim. Sci. 92:3610-3623.

[9] Atkinson, J. L., J. W. Hilton, and S. J. Slinger. 1984. Evaluation of acid-insoluble ash as an indicator of feed digestibility in rainbow trout (Salmo gairdneri). Can. J. Fish. Aquat. Sci. 41:1384-1386.

[10] Galyean, M. L. 2010. Laboratory Procedures in Animal Nutrition Research. Accessed December 2014. http://www.depts.ttu.edu/afs/home/mgalyean/lab_man.pdf.

[11] Adeola, O. 2001. Digestion and balance techniques in pigs. In: A. J. Lewis and L. L. Southern, editors. Swine nutrition, 2nd ed. CRC Press, Boca Raton, FL. p. 903–906.

$$Digestibility, \% = 100 - \left[100 \times \left(\frac{\% \, AIA \, in \, feed \, \times \% \, component \, in \, feces}{\% \, AIA \, in \, feces \, \times \% \, component \, in \, feed}\right)\right]$$

Prior to transportation to the packing plant #1, blood was collected from the 2 gilts identified to be subsampled for intestinal collection that represented the mean weight of the pen. Samples were collected via jugular venipuncture into sterile vacutainer tubes (Tyco Health Care Group LP, Mansfield, MA) and immediately placed on ice until processed. Whole blood was centrifuged (2,000 × g for 15 min at 4°C) and the serum removed and frozen at -80°C until analyzed. Mammalian specific ELISA kits (EMD Millipore Corp., Billerica, MA) were used to determine serum concentrations of glucagon-like peptide 1 (GLP-1; Cat. # EZGLP1T-36K) and glucagon-like peptide 2 (GLP-2;Cat. # EZGLP2-37-K).

On d 117, tissue samples and mucosal scrapings were collected from the two, pre-identified gilts per pen that were identified for small intestinal (SI) mucosal gene expression and gut morphology at packing plant #1. Approximately 15 min after the pigs were slaughtered, the entire viscera was collected and segregated. The small intestine was dissected from the stomach 2 cm distal from the pyloric sphincter of the stomach and 2 cm proximal the ileocaecal junction. From each intestine, two, 5 cm samples were collected from the proximal (2 m from the proximal end of the SI-duodenum) and distal (2 m from the distal end of the SI-ileum) sections of the SI. A mucosal scrape was collected from one of the samples by using a sterile plastic slide to scrape the intestinal cells off the lining of the lumen. Scrapings were placed in a sterile Whirl-Pak bag (Fisher Scientific), and snap chilled and stored in liquid N until all samples were collected. These samples were utilized for mRNA analysis and were maintained at -80°C until analysis. To preserve samples for histological analysis, samples were placed in 4% formaldehyde solution in a 50 mL conical tube for transport back to Kansas State University. Approximately 48 h after samples were placed in the formaldehyde solution, samples were embedded in paraffin and 4 µm cross sections were cut and stained with hematoxalin and eosin (H & E) for histological examination of gut morphology, as described by Hedemann et al. (2006[12]). Measurements included villus height, crypt depth, and villus height to crypt depth ratio.

Ileal mucosal RNA was isolated and transcribed to cDNA as described by Paulk et al. (2015[13]) to measure the relative mRNA gene expression of intestinal fatty acid binding protein (iFABP), copper transporter 1 (CTR1), and glucagon-like peptide 1 (GLP-1R). Five nanogram equivalents of total RNA were amplified with gene-specific primers (Table 5), DNA polymerase, and SYBR green chemistry (Perfecta Sybr fast mix; Quanta Biosciences, Gaithersburg, MD) in a Realplex PCR System (Eppendorf North America, Hauppauge, NY).

At the conclusion of the study, an economic analysis was calculated on both a constant days on feed or constant market weight basis to determine the value of feeding TBCC in the two diet types by the two difference scenarios. Because of the negative impact that high-fiber ingredients have on carcass yield and that producers are paid for their pigs on a carcass basis, economics were calculated on a carcass basis. For calculating economics on a constant days on feed basis, economics were calculated using the treatment means from the experiment. To determine economics on a constant carcass weight basis, carcass feed efficiency was adjusted to a common carcass weight by a factor of 0.005 per lb of final weight, also accounting for the change in carcass yield.

For the constant days on feed and constant weight economic evaluation, total feed cost per pig, cost per lb of gain, gain value, and income over feed cost (IOFC) were calculated. Feed cost was calculated by multiplying

[12] Hedemann, M. S., B. B. Jensen, and H. D. Poulsen. 2006. Influence of dietary zinc and copper on digestive enzymes activity and intestinal morphology in weaned pigs. J. Anim. Sci. 84:3310-3320.

[13] Paulk, C. B., D. D. Burnett, M. D. Tokach, J. L. Nelssen, S. S. Dritz, J. M. DeRouchey, R. D. Goodband, G. M. Hill, K. D. Haydon, and J. M. Gonzales. 2015. Effect of added zinc in diets with ractopamine hydrochloride on growth performance, carcass characteristics, and ileal mucosal inflammation mRNA expression of finishing pigs. J. Anim. Sci. 93:185-196.

ADFI by the feed cost per lb and the number of days in each respective period, then taking the sum of those values for each period calculated the total feed cost per pig. Cost per lb of gain was calculated by dividing the total feed cost per pig by the total carcass pounds gained overall. The value of the carcass weight gained during the experiment (gain value) was calculated by subtracting the product of initial carcass weight from the final HCW, times $88.44/LCWT. Income over feed cost was calculated by subtracting total feed cost from gain value. The income over feed and facilities cost (IOFFC) was calculated for the constant market weight evaluation because pigs with faster growth rates will reach a 210 lb carcass sooner, therefore decreasing the cost of housing the pigs. Facility cost was calculated by multiplying the number of overall days the pigs need to reach a 210 lb carcass based on their respective growth rate by $0.10 per-day facility cost.

Experimental data were analyzed in a randomized complete-block design using the PROC MIXED procedure in SAS (SAS Institute Inc., Cary, NC) with pen serving as the experimental unit and initial BW serving as the blocking factor. The random effect of pen within treatment was included in the model when multiple observations were collected within an experimental unit (pen). Contrasts were used to evaluate the interaction between added Cu and diet type and main effects of added Cu or diet type. Residual assumptions were checked using standard diagnostics on studentized residuals. The assumptions were reasonable met with the exception of gene expression data. For the gene expression criteria, the values were ranked using the PROC RANK procedure prior to analysis. Degrees of freedom were estimated using the Kenward-Roger's approach. Backfat depth, loin depth, and lean percentage were adjusted to a common hot carcass weight. Results from the experiment were considered significant and $P \leq 0.05$ and a tendency between $P > 0.05$ and $P \leq 0.10$.

Results and Discussion

The chemical analyses of the complete diets were similar to the intended formulation (Table 4). The addition of 30% DDGS and 15% bakery meal increased the CP, NDF, crude fiber, ether extract, and ash concentrations in the byproduct diet compared to the corn-soybean meal-based diet as anticipated. Total Ca and P levels were similar between diet types across each dietary phase. The total analyzed Cu levels ranged from 30 to 58 ppm in the diets without added Cu, and ranged from 159 to 211 ppm for the diets with 150 ppm added Cu. These values are within the acceptable analytical limits according to the Association of American Feed Control Officials (AFFCO, 2014[14]), given 17 ppm of Cu was provided by the trace mineral premix and the Cu provided by that of the ingredients used in formulation. For diet characteristics, the byproduct diet decreased bulk density of the diet by an average of 7.4% compared to the corn-soybean meal-based diet.

During the early finishing period (d 0 to 45), there were no Cu × diet type interactions. Feeding pigs 150 mg/kg added Cu tended to increase ADG ($P = 0.076$) by 2.4% compared to pigs fed no added Cu (Table 6). During the late finishing period (d 45 to 117), diet type tended to influence the response to Cu for F/G (Cu × diet type interaction, $P = 0.060$). This was the result of a decrease in F/G for pigs fed the byproduct diet compared to the corn-soybean meal-based diet when added Cu was fed, while pigs fed no added Cu had a slight increase in F/G when fed the byproduct diet compared to the corn-soybean meal-based diet. Overall (d 0 to 117) added Cu did not influence growth performance.

From d 0 to 45, pigs fed the byproduct diet had decreased BW on d 45 ($P = 0.004$) in response to a 3.5% decrease in ADG ($P = 0.001$) and 7.5% decrease in ADFI ($P = 0.009$) compared to the corn-soybean meal-based diet. However, from d 45 to 117 and overall, growth performance and final BW were not influenced by diet type. The reduction in growth performance during the early finishing period was not surprising and is consistent with others whom have fed high-fiber, byproduct diets not equalized for dietary energy. Although in our study, overall growth performance was not affected by diet type, caloric efficiency was worse ($P < 0.05$) for pigs fed the byproduct diet compared to the corn-soybean meal-based diet as they required more Mcal of energy per lb of gain on both an ME and NE basis. This is partly due to both the numerical reduction in F/G for pigs

[14] Association of American Feed Control Officials (AAFCO). 2014. Official Publication. Assoc. Am. Feed Cont. Off., Champaign, IL.

fed the byproduct diet compared to the corn-soybean meal-based diet, as well as the potential overvaluing of the energy content of either the DDGS, bakery meal, or both, during formulation.

Due to the differences in growth response previously seen from added Cu in early and late finishing, the two different time points for GE and DM digestibility were measured in this study. In understanding how Cu can affect diet digestibility, research has suggested that Cu potentially improves fat digestibility (Dove and Haydon, 1992[15]). Diet type influenced the response to Cu (Cu × diet type interaction, $P < 0.05$) for both DM and GE digestibility during early finishing (Table 7). Pigs fed the byproduct diet had a greater decrease in DM and GE digestibility compared to the corn-soybean meal-based diet when Cu was added in the diet, compared to when Cu was not added to the diet. Despite the interaction, pigs fed the byproduct diet had decreased ($P < 0.05$) DM and GE digestibility compared to pigs fed the corn-soybean meal-based diet during the early and late finishing periods. However, adding Cu tended to increase the digestibility of DM ($P = 0.060$) and GE ($P = 0.003$) whereas adding Cu improved GE digestibility in pigs by 2.3% compared to pigs not fed added Cu. Although added Cu influenced the response for DM and GE digestibility and F/G between the two diet types, no biological explanation exists to describe the response. Importantly though, the current experiment was successful at demonstrating the potential for Cu to improve GE digestibility in a higher total dietary fat diet containing byproducts.

For carcass characteristics, pigs fed the byproduct diet compared to the corn-soybean meal-based diet tended to have decreased HCW ($P = 0.067$), and a significant reduction in carcass yield ($P = 0.007$; Table 8). As a result of the decrease in HCW, HCW ADG also tended to decreased ($P = 0.056$) for pigs fed the byproduct diet compared to the corn-soybean meal-based. The numerical reduction in F/G and significant reduction in carcass yield for pigs fed the byproduct diet compared to the corn-soybean meal-based diet also led to a decrease in HCW F/G ($P = 0.011$) for pigs fed the byproduct diet compared to the corn-soybean meal-based diet. Added Cu did not increase HCW or HCW ADG, which is not consistent with previous research completed by Coble et al. (2014). However, the reduction in HCW and carcass yield for pigs fed the byproduct diet compared to those fed the corn-soybean meal-based diet is consistent with most published literature (Asmus et al., 2014).

There were no Cu × diet type interactions for any of the calculated economic scenarios (Table 9). When economics were calculated on a constant days basis, pigs fed the byproduct diet had decreased ($P = 0.001$) feed cost and cost per lb of carcass gain, and a tendency ($P = 0.056$) for a decrease in carcass gain value compared to pigs fed the corn-soybean meal-based diet. Neither reduction resulted in a difference in IOFC. When economics were calculated on a constant carcass weight basis, the adjusted carcass F/G was increased ($P = 0.010$) for pigs fed the byproduct diet compared to the corn-soybean meal-based, as expected. However, facility cost tended to increase ($P = 0.069$) for pigs fed the byproduct compared to those fed the corn-soybean meal-based, as it would take those pigs longer to reach the common carcass weight of 210 lb.

As a result of the variability in proposed modes of action for Cu, other areas of research have evolved to suggest that Cu may potentially be improving growth performance by acting on specific neurological pathways shown to be associated with feed intake. Glucagon-like peptide 1 (GLP-1) is an incretin hormone that is released by the L-cells in the intestine in the response to food ingestion, stimulating insulin secretion. In addition to GLP-1 mediating glucose levels, it also signals the brain to the slow the rate of digestion, and has been shown to directly influence feed intake (Tang-Christensen et al., 1996[16]; Daily and Moran, 2013[17]). In addition, GLP-2 is a hormone that is secreted in a 1 to 1 ratio with GLP-1, shown to improve intestinal health through increasing the growth and functionality of mucosa in the small intestine (Janssen et al., 2013). In the current experiment,

[15] Dove, C. R., and K. D. Haydon. 1992. The effect of copper and fat addition to the diets of weanling swine on growth performance and serum fatty acids. J. Anim. Sci. 70:805-810.
[16] Tang-Christensen, M., P. J. Larsen, R. Goke, A. Fink-Jensen, D. Jessop, M. Moller, and S. P. Sheikh. 1996. Central administration of GLP-1-(7-36) amide inhibits food and water intake in rats. Am. J. Physiol. 274:848-856.
[17] Daily, M. J., and T. H. Moran. 2013. Glucagon-like peptide 1 and appetite. Trend. Endocr. Met. 24(2):85-91.

there was no evidence for a difference in serum concentrations of glucagon-like peptide-1 (GLP-1) or glucagon like peptide-2 (GLP-2; Table 10) between diet types or with added Cu. Factors that could have affected this could be that the current experiment measured the concentrations in serum versus plasma, the age of the pig, and time of collection. Both GLP-1 and GLP-2 have a relatively short half-life (7-17 min) after being released into the blood stream, and feed intake was not controlled across each animal since all pigs were allowed ab libitum intake therefore postprandial collection times may have varied from pig to pig.

Research has demonstrated that added Cu and diet type potentially impact gut morphology. In our study, in the proximal section of the small intestine, neither villus height, crypt depth, nor villus height to crypt depth ratio were not influenced by added Cu or diet type (Table 11). In contrast, in the distal section of the small intestine, crypt depth was decreased in pigs fed added Cu compared to those not fed added Cu ($P = 0.017$).

In order to further investigate the different modes of action for Cu, the relative ileal mucosal mRNA expression of proteins involved with digestion were measured. In combination with the GE digestibility measurements, intestinal fatty acid binding protein (iFABP) mRNA expression was measured because of the importance it has on fatty acid transport across cell membranes. In addition to the serum concentration, GLP-1R mRNA expression was measured for the reasons mentioned previously. Furthermore, mRNA expression of an important protein involved in Cu transport across the cell wall, copper transporter protein-1 (CTR1) was also measured. For relative mucosal mRNA expression, there was no evidence for any diet type × Cu interactions for the measured genes in the proximal or distal small intestine (Table 12). Furthermore, there was no evidence of a difference for the relative mRNA expression of iFABP, CTR1, or GLP-1R in the mucosal layer of the proximal small intestine. However, relative mRNA expression of iFABP in the mucosal layer of the distal small intestine was decreased ($P = 0.032$) in pigs fed added Cu compared to those not fed added Cu. A decrease in iFABP of the distal small intestine mucosa of pigs fed added Cu would suggest that the gene responsible for iFABP transcription is possibly down regulated with added Cu. If fat digestibility is truly increased, we would expect this to be upregulated.

In conclusion, adding 150 ppm Cu to the diet during the early finishing period tended to increase in ADG, but growth performance for the overall growth study was not influenced by added Cu. Pigs fed the byproduct diet compared to the corn-soybean meal-based diet had decreased ADG and ADFI during the early finishing period, but diet type did not affect overall growth performance even though pigs fed the byproduct diet had a reduction in carcass yield and HCW. The changes in growth performance typically observed in finishing pigs fed added Cu does not appear to be related to the changes in serum metabolite profile for GLP-1 and GLP-2 concentrations or relative mRNA expression of GLP-1R, or CTR1. However, more research is need to clarify the impacts that added Cu has on DM and GE digestibility, especially since we observed that Cu influenced energy digestibility during the late finishing period.

Table 1. Composition of diets for phases 1 and 2 (as-fed basis)[1]

	Phase 1		Phase 2	
Item	Corn-soy	Byproduct	Corn-soy	Byproduct
Ingredient, %				
Corn	69.43	37.91	75.51	44.30
Soybean meal-basedbean meal, 46.5% CP	27.83	14.19	21.88	7.92
Distillers dried grains with solubles	---	30.00	---	30.00
Bakery meal	---	15.00	---	15.00
Monocalcium P, 21% P	0.60	0.10	0.60	0.13
Limestone	1.25	1.55	1.13	1.40
Salt	0.35	0.35	0.35	0.35
Vitamin premix	0.08	0.08	0.08	0.08
Trace mineral premix[2]	0.10	0.10	0.10	0.10
L-Lys HCl	0.225	0.560	0.238	0.575
DL-Met	0.075	0.020	0.055	---
L-Thr	0.055	0.100	0.050	0.095
L-Trp	---	0.032	0.001	0.041
Phytase[2]	0.013	0.013	0.013	0.013
TBCC[4]	±	±	±	±
Total	100.0	100.0	100.0	100.0
Calculated analysis				
Standardized ileal digestible (SID) Lys:NE, g/Mcal	4.29	4.29	3.67	3.67
SID AA, %				
Lys	1.050	1.105	0.912	0.958
Ile:Lys	65	59	63	56
Met:Lys	31	29	31	28
Met + Cys:Lys	56	56	56	56
Thr:Lys	62	62	62	62
Trp:Lys	19.7	19	19.0	19.0
Val:Lys	69	69	69	69
Total Lys, %	1.19	1.29	1.03	1.13
ME, kcal/lb	1,492	1,520	1,497	1,525
NE, kcal/lb	1,111	1,166	1,128	1,184
CP, %	19.0	21.1	16.6	18.6
Ca, %	0.66	0.66	0.59	0.59
P, %	0.51	0.49	0.48	0.46
Available P, %	0.36	0.36	0.35	0.35
Base Diet Cost, [5]$/ton	228.14	216.57	215.43	204.49

[1] Phase 1 diet fed from d 0 to 21 and phase 2 fed from d 21 to 45; provided in meal form.
[2] Trace mineral premix provided 17 ppm Cu in the form of $CuSO_4$ to each diet.
[3] Optiphos 2000 (Huvepharma, Sofia, Bulgaria) provided 1,102 phytase units (FTU)/kg, with a release of 0.10% available P.
[4] Tribasic copper chloride (Intellibond C, Micronutrients, Inc., Indianapolis, IN) provided 150 ppm Cu at the expense of corn.
[5] Cost of corn = $4.14/bushel; soybean meal-basedbean meal = $355/ton; DDGS = $201/ton; bakery = $120/ton; L-Lys = $0.64/lb; TBCC = $3.85/lb. The diet cost including TBCC in the base diet is the base diet cost, plus $2.02/ton.

Table 2. Composition of diets for phases 3 and 4 (as-fed basis)[1]

	Phase 3		Phase 4	
Item	Corn-soy	Byproduct	Corn-soy	Byproduct
Ingredient, %				
Corn	79.76	47.27	83.32	47.36
Soybean meal-basedbean meal, 46.5% CP	17.70	5.15	14.17	5.32
Distillers dried grains with solubles	---	30.00	---	30.00
Bakery meal	---	15.00	---	15.00
Monocalcium P, 21% P	0.65	0.13	0.60	0.08
Limestone	1.03	1.30	1.03	1.28
Salt	0.35	0.35	0.35	0.35
Vitamin premix	0.08	0.08	0.08	0.08
Trace mineral premix[2]	0.10	0.10	0.10	0.10
L-Lys HCl	0.240	0.520	0.250	0.400
DL-Met	0.040	---	0.035	---
L-Thr	0.045	0.065	0.055	0.015
L-Trp	0.005	0.035	0.011	0.017
Phytase[3]	0.013	0.013	0.013	0.013
TBCC[4]	±	±	±	±
Total	100.0	100.0	100.0	100.0
Calculated analysis				
Standardized ileal digestible (SID) Lys:NE, g/Mcal	3.22	3.22	2.88	2.88
SID AA, %				
Lys	0.810	0.846	0.730	0.756
Ile:Lys	63	58	61	66
Met:Lys	30	31	31	34
Met + Cys:Lys	56	61	57	68
Thr:Lys	62	62	63	63
Trp:Lys	19.0	19.0	19.0	19.0
Val:Lys	69	73	69	82
Total Lys, %	0.92	1.01	0.83	0.93
ME, kcal/lb	1,499	1,527	1,502	1,528
NE, kcal/lb	1,139	1,192	1,149	1,192
CP, %	14.9	17.4	13.5	17.3
Ca, %	0.55	0.55	0.53	0.53
P, %	0.47	0.45	0.45	0.44
Available P, %	0.34	0.34	0.33	0.33
Base Diet Cost, [5]$/ton	207.12	196.66	200.54	191.49

[1] Phase 3 diets fed from d 45 to 68 and phase 4 diets fed from d 68 to 94; provided in meal form.
[2] Trace mineral premix provided 17 ppm Cu in the form of $CuSO_4$ to each diet.
[3] Optiphos 2000 (Huvepharma, Sofia, Bulgaria) provided 1,102 phytase units (FTU)/kg, with a release of 0.10% available P.
[4] Tribasic copper chloride (Intellibond C, Micronutrients, Inc., Indianapolis, IN) provided 150 mg/kg Cu and was added at the expense of corn.
[5] Cost of corn = $4.14/bushel; soybean meal-basedbean meal = $355/ton; DDGS = $201/ton; bakery = $120/ton; L-Lys = $0.64/lb; TBCC = $3.85/lb. The diet cost including TBCC in the base diet is the base diet cost, plus $2.02/ton.

Table 3. Composition of diets for phase 5 (as-fed basis)[1]

	Phase 5	
Item	Corn-soy	Byproduct
Ingredient, %		
Corn	86.10	47.45
Soybean meal-basedbean meal, 46.5% CP	11.36	5.41
Distillers dried grains with solubles	---	30.00
Bakery meal	---	15.00
Monocalcium P, 21% P	0.65	0.05
Limestone	1.00	1.25
Salt	0.35	0.35
Vitamin premix	0.08	0.08
Trace mineral premix[2]	0.10	0.10
L-Lys HCl	0.250	0.300
DL-Met	0.030	---
L-Thr	0.065	---
L-Trp	0.014	0.002
Phytase[3]	0.013	0.013
TBCC[4]	±	±
Total	100.0	100.0
Calculated analysis		
Standardized ileal digestible (SID) Lys:NE, g/Mcal	2.59	2.59
SID AA, %		
Lys	0.66	0.68
Ile:Lys	61	74
Met:Lys	31	38
Met + Cys:Lys	58	76
Thr:Lys	65	68
Trp:Lys	19.0	19.0
Val:Lys	69	91
Total Lys, %	0.75	0.85
ME, kcal/lb	1,503	1,528
NE, kcal/lb	1,157	1,192
CP, %	12.3	17.2
Ca, %	0.52	0.52
P, %	0.44	0.44
Available P, %	0.33	0.33
Base Diet Cost, [5]$/ton	195.53	187.79

[1] Phase 5 diets fed from d 94 to 117, provided in meal form.
[2] Trace mineral premix provided 17 ppm Cu in the form of $CuSO_4$ to each diet.
[3] Optiphos 2000 (Huvepharma, Sofia, Bulgaria) provided 1,102 phytase units (FTU)/kg, with a release of 0.10% available P.
[4] Tribasic copper chloride (Intellibond C, Micronutrients, Inc., Indianapolis, IN) provided 150 mg/kg Cu and was added at the expense of corn.
[5] Cost of corn = $4.14/bushel; soybean meal-basedbean meal = $355/ton; DDGS = $201/ton; bakery = $120/ton; L-Lys = $0.64/lb; TBCC = $3.85/lb. The diet cost including TBCC in the base diet is the base diet cost, plus $2.02/ton.

Table 4. Chemical analysis of diets (as-fed)[1]

	Added Cu,[2] ppm			
	0		150	
Item	Corn-soy	Byproduct	Corn-soy	Byproduct
Phase 2[3]				
DM, %	85.90	88.00	86.00	88.00
CP, %	15.98	18.30	15.74	18.48
NDF, %	6.61	14.87	8.00	15.05
Crude fiber, %	2.06	4.05	2.32	4.22
Ether extract, %	2.14	4.63	1.94	4.76
Ash, %	4.70	5.10	4.21	4.73
Ca, %	0.74	0.74	0.73	0.69
P, %	0.46	0.51	0.47	0.49
Cu, ppm	54	44	211	190
Bulk density, lb/ft^3	41.5	38.3	40.2	38.6
Phase 3				
DM, %	86.30	88.40	86.30	88.20
CP, %	13.81	16.53	13.72	15.88
NDF, %	6.82	15.20	7.25	14.02
Crude fiber, %	1.98	3.98	2.16	3.79
Ether extract, %	1.96	4.95	2.07	4.64
Ash, %	3.71	4.66	4.03	5.01
Ca, %	0.72	0.72	0.81	0.78
P, %	0.43	0.50	0.47	0.49
Cu, ppm	41	33	209	204
Bulk density, lb/ft^3	41.3	37.2	40.6	37.3
Phase 4				
DM, %	85.80	87.50	85.80	87.60
CP, %	11.50	15.93	12.70	15.94
NDF, %	8.24	14.70	9.87	14.37
Crude fiber, %	2.40	4.03	2.83	3.94
Ether extract, %	2.36	4.38	1.99	4.55
Ash, %	3.35	4.20	3.53	4.70
Ca, %	0.67	0.57	0.63	0.68
P, %	0.42	0.51	0.46	0.48
Cu, ppm	58	30	159	187
Bulk density, lb/ft^3	40.0	37.2	38.1	36.8
Phase 5				
DM, %	85.80	87.20	85.50	87.40
CP, %	11.93	15.52	11.54	15.47
NDF, %	7.89	14.04	7.35	14.95
Crude fiber, %	2.23	4.01	2.05	3.67
Ether extract, %	1.68	4.59	1.98	4.71
Ash, %	3.73	4.68	3.62	5.26
Ca, %	0.72	0.72	0.71	0.75
P, %	0.49	0.54	0.47	0.54
Cu, ppm	48	39	209	203
Bulk density, lb/ft^3	39.5	37.7	39.3	37.6

[1] Values represent means from one composite sample, analyzed in duplicate.
[2] Tribasic copper chloride (Intellibond C; Micronutrients, Inc., Indianapolis, IN).
[3] Phase 1 diets were not available for analysis.

Table 5. Sequences, annealing temperatures, amplicon length, and efficiency of primers used for real-time PCR quantification of gene expression

Item	Forward primer (5′ to 3′)	Reverse primer (5′ to 3′)	T_m,[1] °C	Amplicon length	Efficiency
Small intestine genes					
Copper transport protein -1	CCATGATGATGCCTATGACCTT	ATAGAACATGGCTAGTAAAAACACC	60.5	131	1.12
Glucan-like peptide-1	TACTTCTGGCTGCTGGTGGAG	ACCCCAGCCTATGCTCAGGTA	62.4	104	1.11
Intestinal fatty acid binding protein	CCTCGCAGACGGAACTGAAC	GTCTGGACCATTTCATCCCCG	64.5	135	1.03
Normalizing gene					
Ribosomal protein L4	AGGAGGCTGTTCTGCTTCTG	TCCAGGGATGTTTCTGAAGG	60.5	184	1.06

[1] T_m = melting temperature

Table 6. Effect of added Cu and diet type on growth performance of finishing pigs[1]

	Added Cu,[2] ppm					Probability, $P <$		
	0		150					
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
BW, lb								
d 0	60.8	60.8	60.7	60.8	1.40	1.000	0.954	0.988
d 45	144.6	141.0	146.1	143.1	2.25	0.776	0.082	0.004
d 117	276.3	277.0	281.5	278.0	3.13	0.419	0.237	0.567
d 0 to 45								
ADG, lb	1.86	1.78	1.90	1.82	0.027	0.874	0.076	0.001
ADFI, lb	3.75	3.68	3.87	3.70	0.063	0.221	0.114	0.009
F/G	2.02	2.07	2.04	2.03	0.025	0.147	0.758	0.303
d 45 to 117								
ADG, lb	1.88	1.93	1.93	1.91	0.033	0.207	0.504	0.551
ADFI, lb	5.91	6.00	5.95	6.07	0.086	0.881	0.519	0.224
F/G	3.15	3.11	3.09	3.18	0.035	0.058	1.000	0.390
d 0 to 117								
ADG, lb	1.87	1.87	1.92	1.87	0.023	0.311	0.191	0.269
ADFI, lb	5.06	5.08	5.12	5.12	0.064	0.814	0.376	0.824
F/G	2.71	2.72	2.67	2.74	0.025	0.342	0.737	0.110
Caloric efficiency[4]								
ME	4,056	4,153	4,009	4,174	37.9	0.345	0.720	0.001
NE	3,085	3,233	3,049	3,249	29.2	0.346	0.723	0.001

[1] A total of 757 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 26 to 28 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% DDGS and 15% bakery meal.
[4] Caloric efficiency is expressed as kcal per lb of live weight gain.

Table 7. Effect of added Cu and diet type on dry matter (DM) and gross energy (GE) digestibility of finishing pigs, %[1]

	Added Cu,[2] ppm							
	0		150			Probability, $P <$		
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Phase 2 digestibility[4]								
DM	94.31	92.09	95.02	91.45	0.306	0.029	0.906	0.001
GE	81.67	75.96	83.57	71.16	1.110	0.005	0.187	0.001
Phase 4 digestibility[5]								
DM	95.72	93.14	96.47	93.46	0.280	0.435	0.060	0.001
GE	85.97	77.88	88.11	80.32	0.676	0.832	0.003	0.001

[1] A total of 757 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 26 to 28 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% DDGS and 15% bakery meal.
[4] Phase 2 fecal samples collected over a 2 d period from d 25 to 26.
[5] Phase 4 fecal samples collected over a 2 d period from d 74 to 75.

Table 8. Effect of added Cu and diet type on carcass characteristics of finishing pigs[1]

	Added Cu,[2] ppm							
	0		150			Probability, $P <$		
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Carcass characteristics								
HCW, lb	206.7	203.0	210.4	205.4	2.67	0.776	0.195	0.067
Yield, %	74.28	73.12	74.37	73.26	0.370	0.953	0.752	0.007
BF,[4] in	0.73	0.71	0.73	0.71	0.017	0.135	0.719	0.349
LD,[4] in	2.62	2.50	2.57	2.59	0.045	0.910	0.951	0.192
Lean,[4] %	55.48	55.50	55.24	55.76	0.282	0.389	0.900	0.435
Carcass performance								
HCW ADG, lb	1.38	1.35	1.41	1.37	0.021	0.766	0.173	0.056
HCW F/G	3.68	3.78	3.64	3.76	0.043	0.863	0.110	0.013

[1] A total of 757 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 26 to 28 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% DDGS and 15% bakery meal.
[4] Hot carcass weight was used as a covariate.

Table 9. Effect of added Cu and diet type on economics of finishing pigs[1]

	Added Cu,[2] ppm					Probability, $P <$		
	0		150					
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Constant days, $/pig								
Feed cost	61.45	58.89	62.98	60.06	0.768	0.799	0.072	0.001
Cost/lb gain carcass wt.	0.370	0.362	0.370	0.363	0.005	0.326	0.330	0.001
Carcass gain value[4]	142.52	139.20	145.80	141.33	2.151	0.766	0.173	0.056
IOFC[5]	81.07	80.31	82.82	81.27	1.744	0.803	0.396	0.469
Constant carcass weight,[6] $/pig								
Adjusted Carcass F/G[7]	3.69	3.81	3.64	3.77	0.047	0.832	0.348	0.010
Feed cost	62.31	61.30	62.26	61.46	0.745	0.881	0.935	0.215
Cost/lb gain carcass wt.	0.380	0.374	0.379	0.374	0.005	0.802	0.887	0.224
Carcass gain value[8]	145.41	145.41	145.43	145.42	0.927	---	---	---
IOFC[5]	83.10	84.10	83.16	83.96	1.094	0.892	0.956	0.256
Facility cost[9]	11.92	12.20	11.70	12.05	0.194	0.842	0.284	0.069
IOFFC[10]	71.18	71.91	71.47	71.91	1.143	0.873	0.872	0.512

[1] A total of 757 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 26 to 28 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% DDGS and 15% bakery meal.
[4] Carcass gain value calculated using (HCW × $88.44/lcwt) – (initial wt. × $88.44/lcwt × assumed 75% yield).
[5] Income over feed cost = carcass gain value – feed cost.
[6] Adjusted to constant final carcass weight of 210 lb.
[7] Adjusted using a factor of 0.005 for 1 lb change in carcass weight × carcass yield.
[8] Adjusted gain value calculated using (Final carcass wt. × $88.44/lcwt) – (initial wt. × $88.44/lcwt) × assumed 75% yield.
[9] Facility cost at $0.10/hd/day.
[10] Income over feed and facility cost = IOFC – facility cost.

Table 10. Effect of added Cu and diet type on serum glucagon-like peptide 1 (GLP-1) and 2 (GLP-2) concentrations of finishing pigs[1]

	Added Cu,[2] ppm					Probability, $P <$		
	0		150					
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Serum Concentrations								
GLP-1, pM	12.97	14.07	14.54	12.24	2.659	0.530	0.960	0.825
GLP-2, ng/mL	1.92	1.67	1.78	1.68	0.354	0.831	0.854	0.616

[1] A total of 84 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 2 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% distillers dried grains with solubles (DDGS) and 15% bakery meal.

Table 11. Effect of added Cu and diet type on small intestine (SI) villus height and crypt depth of finishing pigs, um[1]

	Added Cu,[2] ppm					Probability, $P <$		
	0		150					
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Proximal SI								
Villus height	290	277	277	274	9.3	0.625	0.376	0.369
Crypt depth	244	244	214	221	16.7	0.843	0.102	0.874
Villus:crypt ratio	1.25	1.17	1.38	1.27	0.105	0.892	0.253	0.367
Distal SI								
Villus height	412	404	375	400	17.9	0.340	0.230	0.615
Crypt depth	227	223	202	212	7.6	0.330	0.017	0.683
Villus:crypt ratio	1.83	1.83	1.88	1.92	0.106	0.834	0.514	0.823

[1] A total of 84 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 2 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% distillers dried grains with solubles (DDGS) and 15% bakery meal.

Table 12. Effect of added Cu and diet type relative mRNA gene expression of intestinal fatty acid binding protein (iFABP), copper transporter-1 (CTR1), and glucagon-like peptide 1 (GLP-1R) in the proximal and distal small intestinal of finishing pigs[1]

	Added Cu,[2] ppm							
	0		150			Probability, *P* <		
	Corn-soy	Byproduct[3]	Corn-soy	Byproduct[3]	SEM	Cu × Diet Type	Cu	Diet Type
Proximal SI[4]								
iFABP	0.289	0.360	0.316	0.325	0.072	0.442	0.870	0.741
CTR1	0.591	0.601	0.661	0.528	0.130	0.363	0.882	0.457
GLP-1R	0.800	0.802	1.364	0.957	0.420	0.050	0.837	0.685
Distal SI								
iFABP	1.143	0.726	0.664	0.571	0.178	0.283	0.032	0.258
CTR1	1.189	0.995	1.028	1.151	0.198	0.713	0.813	0.634
GLP-1R	2.336	1.592	2.088	1.718	0.566	0.575	0.664	0.382

[1] A total of 84 pigs (PIC 337 × 1050; initially 60.8 lb) were used in a 117-d experiment with 2 pigs per pen and 7 replications per treatment.
[2] Tri-basic copper chloride (TBCC; Intellibond C; Micronutrients, Indianapolis, IN).
[3] Refers to a diet containing 30% distillers dried grains with solubles (DDGS) and 15% bakery meal.
[4] All values indicate relative expression of genes. Normalized expression (ΔCt) for each sample was determined using ribosomal protein L4 as an endogenous control gene. The average normalized expression of the pooled control sample was used as the calibrator to calculate relative gene expression. For each sample relative expression was calculated as 2 –ΔΔCt, in which ΔΔCt represents ΔCt sample – ΔCt calibrator (Livak and Schmittgen, 2001).