Effects of increasing copper from tri-basic copper chloride or a copper-methionine chelate on growth performance of nursery pigs^{1,2}

Corey B. Carpenter,* Jason C. Woodworth,* Joel M. DeRouchey,* Mike D. Tokach,* Robert D. Goodband,*^{,3} Steve S. Dritz,[†]^o Fangzhou Wu,* and James L. Usry[‡]

*Department of Animal Sciences and Industry, College of Agriculture, Kansas State University, Manhattan, KS; †Department of Diagnostic Medicine/Pathobiology, College of Veterinary Medicine, Kansas State University, Manhattan, KS 66506; and [‡]Micronutrients, Indianapolis, IN 46231

ABSTRACT: A total of 2,117 pigs were used in two 35-d growth experiments to determine the effects of increasing added Cu from tri-basic copper chloride (TBCC) or a Cu-methionine chelate (Cu-chelate) on nursery pig growth performance. In experiment 1, 1,452 pigs (350 barrows [DNA 200 × 400; initially 5.9 ± 0.17 kg] in group 1 and 1,102 pigs [PIC 1050×280 ; initially 6.0 ± 0.26 kg] in group 2) were weaned at approximately 21 d of age. In experiment 2, 665 pigs (350 barrows, DNA 200 × 400; initially 6.4 ± 0.19 kg, in group 3 and 315 pigs, DNA 241×600 ; initially 5.2 \pm 0.49 kg, in group 4) were weaned at approximately 21 d of age. Pigs in groups 1, 2, and 3 were fed a common starter diet for 7 d and pigs in group 4 were fed a common diet for 5 d after weaning before starting experiments. On d 0 of each experiment, pens of pigs were blocked by body weight (BW) and assigned to 1 of 7 dietary treatments. Treatments were arranged as a 2×3 factorial plus one control diet, with main effects of Cu source (TBCC vs. Cu-chelate) and level. Copper levels were 50, 100, or 150 mg/kg in experiment 1 and 75, 150, or

225 mg/kg in experiment 2. Diets were corn-soybean meal-based and fed in meal form in two phases (d 0 to 14 and 14 to 35). In experiment 1 from d 0 to 35, there was a Cu source \times level interaction (linear. P < 0.05) for average daily gain (ADG) and d 35 BW where the magnitude of improvement with increasing Cu was greater in pigs fed Cu-chelate compared to those fed TBCC. Increasing added Cu increased (linear, P < 0.01) average daily feed intake (ADFI) and gain:feed (G:F). Although Cu source did not influence G:F, pigs fed Cu from Cu-chelate had greater $(P \le 0.01)$ ADG and ADFI than those fed Cu from TBCC. In experiment 2, from d 0 to 35, there were no evidence for Cu source × level interactions. Increasing Cu increased (linear, P < 0.05) ADG and final BW. The increase in ADG combined with unaffected ADFI resulted in marginally increased G:F (linear, P = 0.052). In summary, these results suggest that increasing dietary Cu from TBCC or a Cu-chelate improved overall ADG, and d 35 BW in nursery pigs and Cu source has potential to influence nursery pig performance.

Key words: copper-chelate, growth, nursery pig, tri-basic copper chloride

Published by Oxford University Press on behalf of the American Society of Animal Science 2018. This work is written by (a) US Government employee(s) and is in the public domain in the US. This Open Access article contains public sector information licensed under the Open Government Licence v2.0 (http://www.nationalarchives.gov.uk/doc/open-government-licence/version/2/).

Transl. Anim. Sci. 2019.3:369–376 doi: 10.1093/tas/txy091

¹Contribution no. 17-344-J. from the Kansas Agric. Exp. Stn., Manhattan, 66506-0210.

³Corresponding author: goodband@ksu.edu Received June 2, 2018. Accepted August 21, 2018.

INTRODUCTION

The NRC (2012) reports a nutrient requirement estimate of 6 mg/kg of Cu for pigs weighing less than 11 kg body weight (BW) and suggests that the Cu requirement decreases to 5 mg/kg for pigs between 11 and 135 kg BW. However, according

²Appreciation is expressed to New Horizon Farms for use of research facilities and feed mill. The authors would also like to express appreciation to Micronutrients, Indianapolis, IN, for partial financial support.

to Flohr et al. (2016), many U.S. swine nutritionists formulate nursery pig diets to contain between 11 and 250 mg/kg Cu. This is likely because previous data have demonstrated increased growth performance of pigs to be associated with feeding high concentrations of Cu through 250 mg/kg (Dove, 1995; Cromwell et al., 1998; Hill et al., 2000).

Research has shown that different inorganic Cu sources ranging from 11 to 327 mg/kg in the diet influence nursery pig growth performance similarly (Cromwell et al., 1998; Shelton et al., 2011; Huang et al., 2015). Tri-basic copper chloride (TBCC; IntelliBond C; Micronutrients, USA, Indianapolis, IN) is an inorganic mineral source, which is nonhygroscopic and has low solubility in water, but is highly soluble in acidic conditions (Miles et al., 1998). Organic Cu sources are argued to be more bioavailable to young pigs due to their chemical structure compared with inorganic sources. Copper-methionine chelate (Cu-chelate; Mintrex Cu; Novus International, St Charles, MO) is an organic form of Cu [Cu(HMTBa),] that has been shown to be more bioavailable to the pig because of decreased binding activity with other dietary constituents, thus suggesting that lower concentrations are required compared with inorganic Cu (Zhao et al., 2009; Ma et al., 2015).

To our knowledge, there are no published data that directly compare the effects of increasing added Cu from TBCC or a Cu-chelate on growth performance of nursery pigs. Therefore, our studies were designed to investigate the effects of increasing Cu from either TBCC or a Cu-chelate on growth performance of nursery pigs.

MATERIALS AND METHODS

General

The Kansas State University Institutional Animal Care and Use Committee approved the protocols used for these studies. Four groups of pigs were used in two experiments with groups 1 and 2 combined to represent experiment 1, and groups 3 and 4 combined to represent experiment 2. Groups 1 and 3 pigs were housed at the Kansas State University Segregated Early Weaning Facility in Manhattan, KS. Group 2 pigs were housed in a commercial research facility in southwestern Minnesota. Group 4 pigs were housed at the Kansas State University Swine Teaching and Research Center in Manhattan, KS. The research facilities were environmentally controlled. For groups 1 and 3, each pen $(1.2 \times 1.2 \text{ m})$ had tri-bar flooring and contained one 4-hole dry self-feeder and one cup waterer to provide ad libitum access to feed and water. For group 2, each pen $(3.7 \times 2.3 \text{ m})$ had plastic slatted flooring and contained one 6-hole dry self-feeder and one pan waterer to provide ad libitum access to feed and water. For group 4, each pen $(1.2 \times 1.5 \text{ m})$ had tri-bar flooring and contained one 4-hole dry self-feeder and one nipple waterer to provide ad libitum access to feed and water. Dietary treatments for groups 1, 3, and 4 were manufactured at the O.H. Kruse Feed Mill in Manhattan, KS. Group 2 dietary treatments were manufactured in a commercial feed mill located in Pipestone, MN.

Live Animal Management, Experiment 1

In group 1, 350 barrows (DNA 200×400 ; initially 5.9 kg) were weaned at approximately 21 d of age and allotted to pens based on initial BW with five pigs per pen and 10 replicate pens per treatment. In group 2, 1,102 pigs (PIC 1,050 \times 280; initially 6.0 kg) were weaned and randomly placed over 2 consecutive days with 24 to 27 pigs per pen and three replicate pens per treatment for each day. A constant barrow to gilt ratio was maintained across pens. Groups 1 and 2 pigs were fed a common starter diet for 7 d after weaning before beginning the experiment.

Live Animal Management, Experiment 2

In group 3, 350 barrows (DNA 200×400 ; initially 6.4 kg) were weaned at approximately 21 d of age and allotted to pens based on initial BW with five pigs per pen and 10 replicate pens per treatment. In group 4, 315 barrows and gilts (DNA 241×600 ; initially 5.2 kg) were weaned and allotted to pens based on initial BW and age. Age block 1 consisted of four replicate pens per treatment and pigs ranged in age from 16 to 20 d. Age block 2 consisted of five replicate pens per treatment and pigs ranged in age from 21 to 24 d. A constant barrow to gilt ratio was maintained across pens. Groups 3 and 4 pigs were fed a common starter diet for 7 and 5 d, respectively, after weaning.

Diets and Response Criteria

Prior to the start of both the experiments, all pigs received a common starter diet containing 17 ppm Cu from $CuSO_4$ in trace mineral premix. At the start of each experiment (d 0), pens were allotted

by weight to 1 of 7 dietary treatments. Treatments were arranged as a 2×3 factorial plus a control diet, with main effects of Cu source and level. The two Cu sources tested were TBCC (IntelliBond C; Micronutrients, USA) and a copper-methionine chelate (Mintrex Cu; Novus International). Copper levels were 50, 100, or 150 mg/kg in experiment 1 and 75, 150, and 225 mg/kg in experiment 2. Diets were corn-soybean meal-based and fed in meal form in two phases (d 0 to 14 and 14 to 35; Table 1). The trace mineral premix added to all diets provided complete diets with 17 mg/kg Cu from $CuSO_4$. The Met concentration contributed by the Cu-chelate was considered in all diet formulation. For each group, pigs and feeders were weighed on d 0, 7, 14, 21, 28, and 35 to calculate average daily gain (ADG), average daily feed intake (ADFI), and gain:feed ratio (G:F).

Chemical Analysis

Complete diet samples for each group were collected from a minimum of six feeders and combined to form one composite sample per treatment and phase. Samples were then split, ground, and sent to commercial analytical labs for chemical analysis.

For experiment 1, group 1 samples were sent to University of Missouri-Columbia Agriculture Chemical Laboratories (Columbia, MO) and analyzed for dry matter (method 934.01; AOAC, 2006), crude protein (method 984.13; AOAC, 2006), crude fiber (method 978.10; AOAC, 2006; AOCS Ba 6a-05, 2006), ether extract (method 920.39; AOAC, 2006), ash (method 942.05; AOAC, 2006), and Cu (method 980.02; AOAC, 2006) concentrations. All samples were also analyzed for Ca, P, and Cu concentrations (Ward Laboratories Inc., Kearney, NE) by iCAP 6000 series ICP Emission Spectrometer (Thermo Electron Corporation, Marietta, OH) using methods outlined by AOAC (2012). Group 1 Cu concentrations were determined by averaging the analyzed values from each lab. Group 2 samples were sent to Midwest Laboratories (Omaha, NE) for duplicate analysis of dry matter (method 930.15; AOAC, 2000), crude protein (method 990.03; AOAC, 2000), crude fiber (method 978.10; AOAC, 2006; AOCS Ba 6a-05, 2006), ether extract (method 945.16; AOAC, 2000), ash (method 942.05; AOAC, 2006), and Cu (method 985.01; AOAC, 2000) concentrations. For experiment 1, nutrient concentrations of complete diets represent the average of the chemical analyses from groups 1 and 2.

For experiment 2, groups 3 and 4 samples were sent to Cumberland Valley Analytical

Table 1. Diet composition (as-fed basis)^a

	Ph	ase
Item	1	2
Ingredient, %		
Corn	48.47	57.30
Soybean meal	27.68	33.73
Dried whey	10.00	
Fat ^b	5.00	5.00
Limestone	0.85	0.85
Monocalcium P (21.5% P)	1.60	1.70
Salt	0.30	0.35
L-Lys-HCl	0.33	0.33
L-Thr	0.15	0.16
Hydroxymethylthio-butanoic acid ^e	0.22	0.18
HP-300 ^d	5.00	
Vitamin premix ^e	0.15	0.15
Trace mineral premix ^f	0.25	0.25
Cu source ^{g,h}		
Total	100.00	100.00
Calculated analysis		
Standardized ileal digestible (SID) amino	acids, %	
Lys	1.30	1.25
Ile:Lys	63	62
Met:Lys	36	35
Met + Cys:Lys	58	58
Thr:Lys	65	65
Trp:Lys	18.4	18.4
Val:Lys	67	67
Net energy, kcal/kg	2,645	2,615
SID Lys:net energy, g/Mcal	4.91	4.78
Crude protein, %	21.7	21.3
Ca, %	0.85	0.80
P, %	0.78	0.75
Standard total tract digestible P, %	0.49	0.44

^a In experiment 1, phase 1 was fed from 6.0 to 9.8 kg and phase 2 was fed from 9.8 to 22.6 kg. In experiment 2, phase 1 was fed from 5.8 to 9.4 kg and phase 2 was fed from 9.4 to 21.6 kg. Dietary treatments were formed by adding 50, 100, or 150 mg/kg Cu (experiment 1) or 75, 150, or 225 mg/kg of Cu (experiment 2) from either tri-basic copper chloride or Cu-chelate at the expense of corn. The trace mineral premix was formulated to contribute 17 mg/kg of Cu to the complete diet for each experiment.

^b Choice white grease for groups 1, 3, and 4; beef tallow for group 2. ^c Alimet, Novus International.

 $^{\rm d}$ HP-300, Hamlet Protein, Findlay, OH, formulated with 3.25% SID Lys.

^e Provided per kilogram of diet: 8,818 IU vitamin A, 2,205 IU vitamin D₃, 44.1 IU vitamin E, 0.04 mg vitamin B₁₂, 8.3 mg riboflavin, 82.7 mg niacin, 27.6 mg pantothenic acid, 4.4 mg menidione.

^f Provided per kilogram of diet: 17 mg Cu from copper sulfate, 0.3 mg I from Ca iodate, 110 mg Fe from ferrous sulfate, 33 mg Mg from manganese sulfate, 0.3 mg Se from sodium selenite, 110 mg Zn from zinc sulfate.

^g Copper-methionine hydroxy analogue (Mintrex Cu, Novus International).

^h Tri-basic copper chloride (IntelliBond C, Micronutrients, USA).

Services (Hagerstown, MD) for analysis of dry matter (method 930.15; AOAC, 2000); crude protein (method 990.03; AOAC, 2000); ether extract

(method 2003.05; AOAC, 2006); ash (method 942.05; AOAC, 2000); and Ca, P, and Cu (method 985.01; AOAC, 2000) concentrations. Copper concentrations were also analyzed at Ward Laboratories Inc.. Copper concentrations were determined by averaging three analyzed values (two from Cumberland Valley Analytical Services and one from Ward Laboratories Inc.). In group 4, nutrient concentrations were determined by averaging two analyzed values. For experiment 2, nutrient concentrations of complete diets represent the average of the chemical analyses from groups 3 and 4.

Statistical Analysis

For each experiment, group × treatment interactions were tested and were not significant (P > P)0.05); therefore, data were combined across groups 1 and 2 for experiment 1 and groups 3 and 4 for experiment 2. Each experiment was analyzed as a randomized incomplete block design using PROC GLIMMIX (SAS Institute, Inc., Cary, NC), with pen as the experimental unit and dietary treatment as the fixed effect. The random effect of weight block within group was used in the model. The main effects of Cu source (TBCC or Cu-chelate) and linear and quadratic effects of Cu level (0, 50, 100, and 150 mg/kg in experiment 1 or 0, 75, 150, and 225 mg/kg in experiment 2) as well as their interactions were analyzed using polynomial contrast statements. Results were considered significant with P < 0.05 and marginally significant when P < 0.10 and $P \ge 0.05$.

RESULTS

Diet Analysis, Experiments 1 and 2

The chemical analyses of the complete diets reasonably agreed to the intended formulation (Tables 2 and 3). The analyzed Cu concentrations were generally higher than the formulated levels but followed similar patterns as the designed treatment structure.

Growth Performance, Experiment 1

During the feeding period, 58 pigs (9 pigs from the control group; 11, 6, and 11 pigs from the 50, 100, and 150 ppm TBCC treatment groups, respectively; 2, 10, and 9 pigs from the 50, 100, and 150 ppm Cu-chelate treatment groups, respectively) were removed from the study due to poor health or death.

From d 0 to 14 (6.0 to 9.8 kg), there were no evidence for Cu source × level interactions in any response criteria (P > 0.26). Increasing added Cu increased (linear, P < 0.01; Table 4) ADG and ADFI, and marginally improved (linear, P = 0.057) G:F. Although Cu source did not influence G:F, pigs fed added Cu from Cu-chelate had marginally greater (P < 0.10) ADG and ADFI than those fed added Cu from TBCC.

From d 14 to 35 (9.8 to 22.6 kg), there was a source × level interaction (linear, P = 0.011) for ADG. ADG increased with each increasing level of Cu from Cu-chelate; however, pigs fed Cu from TBCC only had increased ADG at 150 mg/kg. Added Cu increased (linear, P = 0.001) ADFI and G:F. Similar to the performance from d 0 to 14, Cu source did not influence G:F; however, pigs fed Cu from Cu-chelate had greater (P < 0.01) ADG and ADFI than those fed Cu from TBCC.

Overall from d 0 to 35 (6.0 to 22.6 kg), a Cu source × level interaction was observed (linear, P = 0.042) for ADG where the magnitude of improvement with increasing Cu was greater in pigs fed Cu-chelate diets compared with those fed TBCC diets. Increasing added Cu increased (linear, P = 0.001) ADFI and G:F. Although Cu source did not influence G:F, pigs fed Cu from Cu-chelate had greater (P < 0.010) ADG and ADFI than those fed Cu from TBCC.

Growth Performance, Experiment 2

During the feeding period, 22 pigs (1 pig from the control group; 1, 3, and 3 pigs from the 75, 150, and 225 ppm TBCC treatment groups, respectively; 5, 3, and 6 pigs from the 75, 150, and 225 ppm Cu-chelate treatment groups, respectively) were removed from the study due to poor health or death.

From d 0 to 14 (5.8 to 9.4 kg), a marginal source \times level interaction (quadratic, P = 0.081; Table 5) was observed for ADG with maximum ADG observed at 150 mg/kg Cu from the Cu-chelate, but at 225 mg/kg for pigs fed TBCC. Increasing Cu increased (linear, P = 0.001) ADFI with no changes in G:F.

From d 14 to 35 (9.4 to 21.6 kg), there were no evidence for Cu source × level interactions. Neither Cu source nor level influenced ADG or ADFI; however, increasing Cu improved (linear, P = 0.019) G:F.

Table 2.	Chemical	analysis of	diets	(experiment	1; as-fed	basis) ^a

			Р	hase 1			Phase 2								
		Added Cu, mg/kg							Added Cu, mg/kg						
		TBCC ^b			Cu-chelate ^c			TBCC ^b			Cu-chelate ^c				
Item	Control	50	100	150	50	100	150	Control	50	100	150	50	100	150	
Dry matter, %	87.5	87.2	86.8	86.7	86.6	86.6	86.8	86.7	86.4	85.8	85.6	86.2	86.1	85.7	
Crude protein, %	21.9	21.0	20.6	20.9	20.8	20.8	20.8	19.2	20.4	19.9	21.9	18.9	19.6	21.2	
Crude fiber, %	1.9	2.0	1.5	1.6	2.0	2.1	2.1	2.6	2.4	2.1	2.4	1.9	2.0	2.0	
Ether extract, %	7.1	6.8	7.2	6.9	6.6	7.6	6.9	6.7	6.5	6.3	5.3	6.3	5.6	5.6	
Ash, %	5.7	5.7	5.6	6.0	5.9	5.8	5.9	5.4	5.4	5.3	5.7	5.4	5.7	5.6	
Ca, %	0.85	0.78	0.77	0.82	0.75	0.86	0.76	0.75	0.97	1.00	0.76	1.01	0.87	0.91	
P, %	0.76	0.74	0.68	0.79	0.73	0.83	0.78	0.62	0.72	0.79	0.77	0.81	0.66	0.88	
Cu, mg/kg	40	88	140	145	104	155	204	40	75	119	174	100	135	230	

^a Multiple samples of each diet were collected, blended and subsampled, and analyzed (Missouri Agricultural Experimentation Lab, Colombia, MO; Ward Laboratories, Kearney, NE; and Midwest Labs, Omaha, NE). All values represent the combined average of the chemical analyses of diets for the experiment.

^b Tri-basic copper chloride, (IntelliBond C, Micronutrients USA).

^c Copper-methionine hydroxy analogue (Mintrex Cu, Novus International).

Table 3. Chemical analysis of diets (experiment 2; as-fed basis)^a

		Phase 1								Phase 2						
		Added Cu, mg/kg							Added Cu, mg/kg							
			TBCC ^b			Cu-chelate ^c			TBCC ^b			Cu-chelate ^c				
Item	Control	75	150	225	75	150	225	Control	75	150	225	75	150	225		
Dry matter, %	88.9	88.1	88.9	89.0	89.0	89.0	88.9	88.9	88.4	88.2	88.6	88.4	88.6	88.6		
Crude protein, %	24.1	24.3	24.3	24.5	24.7	24.8	24.0	24.3	23.8	24.3	32.9	24.4	24.0	23.4		
Crude fiber, %	2.5	2.5	2.4	2.5	2.8	2.7	2.6	2.6	3.0	2.7	2.6	2.5	2.5	2.3		
Ether extract, %	6.0	6.7	6.4	7.2	6.9	6.8	7.3	7.3	7.1	7.0	7.0	7.3	7.1	6.6		
Ash, %	6.6	6.6	6.6	6.8	6.5	6.4	6.4	5.9	6.1	6.0	7.2	5.7	5.7	5.3		
Ca, %	1.06	0.97	1.01	1.01	0.94	0.95	1.03	1.02	0.93	0.93	1.01	0.93	0.99	0.93		
P, %	0.86	0.85	0.84	0.89	0.84	0.88	0.88	0.82	0.81	0.82	0.84	0.79	0.84	0.76		
Cu, mg/kg	24	86	179	248	134	227	316	28	90	144	246	114	177	283		

^a For each group of pigs, multiple samples of each diet were collected, blended and subsampled, and analyzed (Cumberland Valley Analytical Services, Hagerstown, MD and Ward Laboratories, Kearney, NE). All values represent the combined average of the chemical analyses of diets for the experiment.

^b Tri-basic copper chloride (IntelliBond C, Micronutrients).

^c Copper-methionine hydroxy analogue (Mintrex Cu, Novus International).

Overall from d 0 to 35 (5.8 to 21.6), there were no evidence for Cu source × level interactions. Increasing Cu increased (linear, P < 0.05) ADG and final BW, and marginally improved G:F (linear, P = 0.052), but no Cu source effect was observed.

DISCUSSION

Studies have shown that adding levels of Cu well above the nutritional requirement improves nursery pig growth performance (Dove, 1995; Hill et al., 2000; Bikker et al., 2016). Dove (1995) fed diets to nursery pigs that contained either 15 or 250 mg/ kg added Cu from $CuSO_4$ and reported that pigs provided diets containing 250 mg/kg added Cu had greater growth performance than those fed diets containing 15 mg/kg Cu. These findings are similar to those from Cromwell et al. (1998) who reported that pigs fed 200 mg/kg Cu from CuSO₄ had greater ADG than those fed 13 mg/kg. A regional study completed by Hill et al. (2000) also observed that pigs fed high added Cu (250 mg/kg) had greater ADG and G:F than those fed diets without added Cu (other than in the trace mineral premix). More recently, Bikker et al. (2016) fed pigs (approximately 7 to 37 kg) increasing Cu (15, 80, 120, and 160 mg/ kg Cu) from CuSO₄ in barley-wheat-based diets with 20% corn. In their study, increasing added Cu from 15 to 160 mg/kg Cu increased ADG and ADFI for growing pigs.

Each of our experiment agrees that increasing added Cu increased overall ADG and BW. It

Table 4. Effects of increasing Cu and source on growth performance of pigs (experiment 1)^{a,b}

				Added C	u, mg/kg	;			Probability, <i>P</i> <					
			TBCC ^c			Cu-chelate ^d				Cu level		Source \times level		
Item	Control	50	100	150	50	100	150	SEM	Cu Source	Linear	Quadratic	Linear	Quadratic	
BW, kg														
d 0	6.0	6.0	6.0	6.0	6.0	6.0	6.0	0.05	0.112	0.998	0.613	0.372	0.402	
d 14	9.6	9.5	9.7	9.9	9.8	9.9	10.0	0.17	0.030	0.001	0.941	0.462	0.318	
d 35	21.5	22.3	22.4	22.8	22.6	23.0	23.4	0.25	0.001	0.001	0.031	0.019	0.384	
d 0 to 14														
ADG, g	251	251	266	274	268	271	281	10.2	0.081	0.001	0.953	0.780	0.377	
ADFI, g	356	353	373	374	373	373	388	14.5	0.053	0.003	0.917	0.511	0.718	
G:F	0.709	0.713	0.714	0.737	0.721	0.726	0.728	0.0122	0.678	0.057	0.794	0.650	0.265	
d 14 to 35														
ADG, g	567	602	602	614	612	621	635	6.8	0.001	0.001	0.004	0.011	0.568	
ADFI, g	841	870	861	882	888	894	901	13.6	0.009	0.001	0.159	0.167	0.222	
G:F	0.675	0.693	0.7	0.698	0.692	0.696	0.707	0.0084	0.800	0.001	0.216	0.471	0.417	
d 0 to 35														
ADG, g	440	461	467	477	474	480	493	5.9	0.001	0.001	0.027	0.042	0.334	
ADFI, g	646	662	665	678	682	684	694	9.7	0.007	0.001	0.279	0.211	0.277	
G:F	0.681	0.696	0.703	0.705	0.698	0.702	0.711	0.0062	0.632	0.001	0.179	0.540	0.669	

^a A total of 1,452 pigs (350 barrows [DNA 200 × 400; initially 5.9 kg; 5 pigs per pen; 10 pens per treatment] in group 1 and 1,102 pigs [PIC 1050 × 280; initially 6.0 kg; 24 to 27 pigs per pen; 6 pens per treatment] in group 2) were used in a 35-d growth studies. Data were combined across the two groups. The trace mineral premix was formulated to contribute 17 mg/kg of Cu to the complete diet.

^b Growth performance criteria include: body weight (BW), average daily gain (ADG), average daily feed intake (ADFI), and gain:feed (G:F).

^c Tri-basic copper chloride (Intellibond C, Micronutrients USA).

^d Copper-methionine hydroxy analogue (Mintrex Cu, Novus International).

appears that overall ADFI was also improved by increasing added Cu in experiment 1 but not in experiment 2. Results from experiment 1 also suggest that increasing added Cu increased growth performance in the early (6.0 to 9.8 kg), late (9.8 to 22.6 kg), and overall (6.0 to 22.6 kg) nursery periods. Others have found similar responses in previous studies with nursery pigs (Dove, 1995; Shelton et al., 2011; Bikker et al., 2016). However, in experiment 2, ADG and ADFI increased only in the first 14 d (5.8 to 9.4 kg) of the experiment for pigs fed increasing added Cu, with no dose response in the late (9.4 to 21.6 kg) or overall (9.4 to 21.6) nursery periods. However, the magnitude of growth improvement to increasing Cu was numerically similar during early and late periods. In the early and late nursery period, the ADG advantage was 22 and 18 g/d, respectively, compared to the pigs fed the control diet. It appears that the ADG response to increasing Cu was more difficult to detect in the late nursery period. Interestingly, G:F and d 35 BW were increased with increasing added Cu during the late nursery period in experiment 2. This may help support that while not significant, there may be potential for a growth advantage to increasing added Cu during the late nursery period.

To the best of our knowledge, there is no other published research that has directly compared the effects of increasing Cu from TBCC to a Cu-chelate on nursery pig performance. Much of the research in the current body of literature have compared increasing Cu from CuSO₄ alone or comparing $CuSO_4$ to TBCC. Huang et al. (2015) fed diets to nursery pigs that contained increasing Cu (14 to 273 mg/kg) and observed that no differences in growth among pigs fed Cu from either TBCC or $CuSO_4$. Huang et al. (2015) hypothesized that it may be due to the short duration (10 d) of the study that a response to Cu source was not observed. Cromwell et al. (1998) conducted three experiments and fed diets to nursery pigs (7.9 to 17.7 kg) that contained 13 (control), 100, 150, or 200 mg/kg added Cu from either TBCC or CuSO₄. They reported that pigs fed diets containing different Cu sources had similar growth performance and pigs fed 200 mg/kg Cu from CuSO₄ had improved performance compared with those fed 13 mg/kg. Alternatively to the aforementioned studies, Ma et al. (2015) completed a multi-trial meta-analysis to evaluate the dose-responses of Cu from either Cu-chelate (Mintrex Cu; Novus International) or $CuSO_4$ in nursery pig growth performance. The authors reported that only from d 14 to 21 postweaning (21 d weaning age) was the ADG response curve significantly different for pigs fed Cu-chelate compared with those fed CuSO₄. In addition, the

				Added C	Cu, mg/kg	5			Probability, P <					
			TBCC ^c			Cu-chelate ^d				Cu level		Source \times level		
Item	Control	75	150	225	75	150	225	SEM	Cu source	Linear	Quadratic	Linear	Quadratic	
BW, kg														
d 0	5.8	5.8	5.8	5.8	5.8	5.8	5.8	0.17	0.830	0.873	0.933	0.792	0.945	
d 14	9.1	9.1	9.4	9.6	9.3	9.6	9.5	0.31	0.365	0.001	0.884	0.469	0.074	
d 35	21.1	21.0	21.8	22.1	21.7	22.2	21.9	0.81	0.278	0.013	0.624	0.636	0.120	
d 0 to 14														
ADG, g	239	236	254	266	249	269	260	12.0	0.253	0.003	0.873	0.662	0.081	
ADFI, g	314	306	342	349	334	339	341	11.4	0.372	0.001	0.925	0.152	0.092	
G:F	0.758	0.767	0.745	0.760	0.743	0.788	0.757	0.0202	0.706	0.865	0.848	0.446	0.604	
d 14 to 35														
ADG, g	569	563	587	592	574	591	582	26.5	0.848	0.148	0.963	0.579	0.443	
ADFI, g	855	848	861	873	864	871	840	36.2	0.884	0.824	0.715	0.196	0.158	
G:F	0.663	0.664	0.682	0.679	0.661	0.679	0.687	0.0091	0.908	0.019	0.806	0.541	0.488	
d 0 to 35														
ADG, g	437	432	453	460	442	461	452	20.1	0.695	0.044	0.988	0.578	0.301	
ADFI, g	639	631	652	662	648	656	639	25.2	0.952	0.277	0.813	0.143	0.134	
G:F	0.681	0.684	0.693	0.695	0.677	0.701	0.702	0.0098	0.696	0.052	0.869	0.402	0.796	

Table 5. Effects of increasing Cu and source on growth performance of pigs (experiment 2)^{a,b}

^a A total of 665 pigs (350 barrows [DNA 200 \times 400; initially 6.4 kg; 5 pigs per pen; 10 pens per treatment] in group 3 and 315 pigs [DNA 241 \times 600; initially 5.2 kg; 5 pigs per pen; 9 pens per treatment] in group 4) were used in a 35-d growth studies. Data were combined across the two groups. The trace mineral premix was formulated to contribute 17 mg/kg of Cu to the complete diet.

^b Growth performance criteria include: body weight (BW), average daily gain (ADG), average daily feed intake (ADFI), and gain:feed (G:F).

^c Tri-basic copper chloride, (Intellibond C, Micronutrients USA).

^d Copper-methionine hydroxy analogue (Mintrex Cu, Novus International).

authors found similar ADFI responses among Cu sources. These studies generally conclude that increasing added Cu improves nursery growth performance: however, consistent with our findings in the study herein, it appears there is variation in the literature around whether or not Cu source affects nursery pig growth performance. Interestingly, in the present study, chemical analysis showed that Cu-chelate diets had consistently higher analyzed Cu by averagely 40 mg/kg compared with that of TBCC diets across treatment groups (Tables 2 and 3). It is unclear whether the discrepancy in analyzed Cu concentrations contributed to the Cu source effect observed in experiment 1. However, it is apparent that this discrepancy did not result in Cu source effect in experiment 2.

The present study is unique in that the added Cu sources used herein have not been directly compared with each other before. Due to a relatively small amount of research, little is known about the growth promotional mechanisms for TBCC and Cu-chelates. However, the responses observed in the study herein are consistent that Cu in general results in improved ADG in the nursery phase. Some studies report the feed efficiency responses may be related to disruption of bacterial cell membranes resulting in Cu ion penetration and bacterial cell toxicity (Pang and Applegate 2007). However, it is difficult to explain the source \times level interactions on ADG observed in both experiments 1 and 2.

In summary, these results suggest that increasing TBCC or Cu-chelate has the potential to affect both phase 1 and 2 nursery growth phases and subsequently improves d 35 BW. The present study provide conflicting results on whether or not nursery pig growth performance is dependent on Cu source; pigs provided Cu from Cu-chelate had greater ADG, ADFI, and d 35 BW than those provided Cu from TBCC in experiment 1 but not in experiment 2.

LITERATURE CITED

- AOAC. 2000. Official methods of analysis. 17th ed. Gaithersburg (MD): Association of Official Analytical Chemists.
- AOAC. 2006. Official methods of analysis. 18th ed. Washington (DC): AOAC Int.
- AOAC. 2012. Official methods of analysis. 19th ed. Gaithersburg (MD): AOAC Int.
- Bikker, P., A.W. Jongbloed, and J. van Baal. 2016. Dosedependent effects of copper supplementation of nursery diets on growth performance and fecal consistency in weaned pigs. J. Anim. Sci. 94:181–186. doi:10.2527/ jas2015-9874
- Cromwell, G.T., M.D. Lindemann, H.K. Monegue, D.D. Hall, and D.E. Orr. 1998. Tribasic copper chloride and copper sulfate as copper sources for weanling pigs. J. Anim. Sci. 76:118–123. doi:10.2527/1998.761118x

- Dove, C.R. 1995. The effect of Cu level on nutrient utilization of weanling pigs. J. Anim. Sci. 73:166–171. doi:10.2527/1995.731166x
- Flohr, J.R., J.M. DeRouchey, J.C. Woodworth, M.D. Tokach, R.D. Goodband, and S.S. Dritz. 2016. A Survey of current feeding regimens for vitamins and trace minerals in the US swine industry. J. Swine Health Prod. 24:290–303.
- Hill, G.M., G.L. Cromwell, T.D. Crenshaw, C.R. Dove, R.C. Ewan, D.A. Knabe, A.J. Lewis, G.W. Libal, D.C. Mahan, G.C. Shurson, et al. 2000. Growth promotion effects and plasma changes from feeding high dietary concentrations of zinc and copper to weanling pigs (regional study). J. Anim. Sci. 78:1010–1016. doi:10.2527/2000.7841010x
- Huang, Y.L., M.S. Ashwell, R.S. Fry, K.E. Lloyd, W.L. Flowers, and J.W. Spears. 2015. Effect of dietary copper amount and source on copper metabolism and oxidative stress of weanling pigs in short-term feeding. J. Anim. Sci. 93:2948–2955. doi:10.2527/jas.2014-8082
- Ma, Y.L., G.I. Zanton, J. Zhao, K. Wedekind, J. Escobar, and M. Vazquez-Añón. 2015. Multitrial analysis of the effects of copper level and source on performance in nursery pigs. J. Anim. Sci. 93:606–614. doi:10.2527/jas.2014-7796
- Miles, R.D., S.F. O'Keefe, P.R. Henry, C.B. Ammerman, and

X.G. Luo. 1998. The effect of dietary supplementation with copper sulfate or tribasic copper chloride on broiler performance, relative copper bioavailability, and dietary prooxidant activity. Poult. Sci. 77:416–425. doi:10.1093/ ps/77.3.416

- NRC. 2012. Nutrient requirements of swine. 11th rev. ed. Washington (DC): National Academies Press.
- Pang, Y., and T.J. Applegate. 2007. Effects of dietary copper supplementation and copper source on digesta ph, calcium, zinc, and copper complex size in the gastrointestinal tract of the broiler chicken. Poult. Sci. 86:531–537. doi:10.1093/ps/86.3.531
- Shelton, N.W., M.D. Tokach, J.L. Nelssen, R.D. Goodband, S.S. Dritz, J.M. DeRouchey, and G.M. Hill. 2011. Effects of copper sulfate, tri-basic copper chloride, and zinc oxide on weanling pig performance. J. Anim. Sci. 89:2440–2451. doi:10.2527/jas.2010-3432
- Zhao, J., R.J. Harrell, G. Allee, B. Hinson, P. Winkelbauer, C. Atwell, J.D. Richards, and M. Vazquez-Anon. 2009. Effect of an organic copper source on growth performance and tissue copper concentration in nursery pigs. J. Anim. Sci. 87(Suppl. 3):60 (Abstr.).