Asian-Aust. J. Anim. Sci.
Vol. 23, No. 6 : 757 - 761
June 2010

www.ajas.info

Effect of Dietary Copper Sources (Cupric Sulfate and Cupric Methionate) and Concentrations on Performance and Fecal Characteristics in Growing Pigs

Y. Huang, T. X. Zhou, J. H. Lee, H. D. Jang, J. C. Park[1] and I. H. Kim*
Department of Animal Resource and Science, Dankook University, Cheonan, Choongnam, 330-714, Korea

ABSTRACT : This study was conducted to assess the effects of organic and inorganic copper on performance in growing pigs. A total of 100 pigs, average age 63 d and initial body weight 21.46±1.13 kg, were assigned to five treatment groups. Dietary treatments included i) CON (basal diet, 0 ppm Cu), ii) T1 (basal diet with 67 ppm Cu as cupric sulfate, $CuSO_4$), iii) T2 (basal diet with 134 ppm Cu as $CuSO_4$), iv) T3 (basal diet with 67 ppm Cu as cupric methionate, CuMet) and v) T4 (basal diet with 134 ppm Cu as CuMet). Throughout the entire experimental period, ADG (average daily gain), ADFI (average daily feed intake) and G/F (gain: feed) ratios showed no significant differences. The dry matter digestibility was improved in the T1, T2, T3, and T4 treatments ($p<0.05$), as compared with CON. Nitrogen digestibility was improved in the T3 treatment group as compared with CON ($p<0.05$). As compared with the T1 treatment group, fecal pH values were improved in the CON, T3, and T4 treatment groups ($p<0.05$). Fecal Cu concentrations were significantly lower in the CON, T3, and T4 treatment groups than in T1 and T2 ($p<0.05$). The incidence of diarrhea was reduced when the pigs were fed on the T2, T3, and T4 diets as compared with CON. In conclusion, diets supplemented with 67 or 134 ppm Cu as CuMet may prove effective in improving nutrient digestibility and fecal pH value in growing pigs, and fecal Cu concentrations may be reduced by CuMet supplementation. (**Key Words :** Growing Pigs, Copper, Nutrients Digestibility, pH, Diarrhea)

INTRODUCTION

Copper is added routinely to the diets of growing swine. The NRC (1998) requirement of copper for pigs is only 5 to 6 ppm. When fed with 100 to 250 ppm, this element was determined to be effective for growth promotion with antibacterial activity in pigs (Barber et al., 1955; Braude, 1967, 1975; Cromwell et al., 1981). Cromwell et al. (1989) and Bowland (1990), however, have suggested that copper stimulated growth was associated with the solubility of the copper source; hence, copper must be available for absorption. The most frequently utilized dietary copper supplement in the animal diet is inorganic Cu, usually in the form of copper sulfate ($CuSO_4{\cdot}5H_2O$). Copper, occurs in the organic forms of chelates, complexes, or proteinates, and like other organic trace minerals, is often considered as an alternative to inorganic sources in animal diets. This is probably attributable to improved absorption, which enhances the efficiency of use (Downs et al., 2000; Yu et al., 2000; Guo et al., 2001). Baker and Ammerman (1995) reported that the relative bioavailability estimates of organic Cu sources ranged between 88% and 147% of the response to cupric sulfate in poultry, swine, sheep, and cattle. It has also been observed that such high levels of Cu supplementation result in its high excretion in the feces (Paik et al., 1999). The presence of high Cu concentrations in the feces inhibits the normal fermentation process, and the accumulation of Cu in the soil causes environmental concerns. If organic Cu in the form of Cu-methionine (CuMet) chelate is more readily absorbed, this may save copper resources, decrease Cu excretion, and ameliorate the environmental concerns attendant to Cu dietary supplementation.

MATERIALS AND METHODS

Copper- methionate (10.0% Cu, 8.5% H_2O) or $CuSO_4$ (33.0% Cu) replaced corn in the diet to create five dietary treatments. The basal diet contained a trace mineral premix

* Corresponding Author: I. H. Kim. Tel: +82-41-550-3652, Fax: +82-41-553-1618, E-mail: inhokim@dankook.ac.kr
[1] Swine Science Division, National Institute of Animal Institute of Animal Science, Rural Development administration, Korea.
Received August 7, 2008; Accepted September 17, 2009

that provided 16.5 ppm Cu as $CuSO_4$. The treatments assessed in this study were as follows: CON; T1, 67 ppm Cu as $CuSO_4$; T2, 134 ppm Cu as $CuSO_4$; T3, 67 ppm Cu as CuMet; T4, 134 ppm Cu as CuMet.

Experimental design, animals and diets

Total 100 pigs (Landrace×Yorkshire)×(Duroc) with an average of 63 d of age and initial weight of 21.46±2.13 kg were utilized. There were five replicate pens per treatment group with four pigs per pen. The growing phase lasted for a total of 35 d. During the experimental period, all diets were provided in meal form. Table 1 shows the ingredient composition of the basal diets. The diets were formulated to meet or exceed the requirements for all nutrients (NRC, 1998) for growing pigs. The pigs were provided with *ad libitum* access to feed and water throughout the entirety of the growing phase.

Sampling and measurements

At the outset and the end of experiment, body weight and feed intake were measured in order to determine the ADG, ADFI, and G/F ratio. The feed and fecal grab samples were randomly obtained from at least two pigs in each pen on days 21 and 35 in an effort to determine the digestibilities of DM (dry matter) and N (nitrogen). Chromic oxide (0.2%) was utilized as an indigestible marker in the diet in order to calculate the digestibility coefficients. After collection, the fresh samples were frozen in a refrigerator at -20°C until they were analyzed. Prior to chemical analysis, the fecal samples were dried at 70°C for 72 h and subsequently ground to pass through a 1-mm screen. All of the fecal samples, along with the feed samples, were analyzed for DM and N in accordance with the AOAC procedures (AOAC, 1995). Chromium levels were assessed via UV (Ultraviolet) absorption spectrophotometry (Shimadzu, UV-1201, Japan). Nitrogen levels were assessed using a Kjectec 2300 Nitrogen Analyzer (Itecator, Tecator AB, Hoganas, Sweden).

Table 1. Formula and chemical compositions of diets (as-fed basis)

Ingredients (g/kg)	CON
Ground corn	599.3
Soybean meal	237.5
Rice bran	50.0
Molasses	40.0
Animal fat	26.1
Rapeseed meal	20.0
Defl. phosphate	11.6
Calcium carbonate	4.4
L-lysine (78%)	3.4
Salt	1.5
Vitamin premix[1]	1.0
Mineral premix[2, 3]	2.5
DL-methionine (980 g/kg)	1.0
Choline chloride (600 g/kg)	0.8
L-threonine (980 g/kg)	0.9
Chemical composition[4]	
Digestible energy (MJ/kg)	144.2
Crude protein (g/kg)	177.2
Lysine (g/kg)	10.2
Calcium (g/kg)	7.0
Phosphorus (g/kg)	5.9

[1] Provided per kg of complete diet: 4,000 IU of vitamin A; 800 IU of vitamin D_3; 17 IU of vitamin E; 2 mg of vitamin K; 4 mg of vitamin B_2; 1 mg of vitamin B_6; 16 μg of vitamin B_{12}; 11 mg of pantothenic acid; 20 mg of niacin and 0.02 mg of biotin.

[2] Provided per kg of complete diet: 175 mg of Fe; 89 mg of Mn; 0.3 mg of I; 0.5 mg of Co and 0.4 mg of Se.

[3] T1 and T2 provided 67 and 134 ppm of Cu as $CuSO_4$; T3 and T4 provided 67 and 134 ppm of Cu as CuMet.

[4] Calculated values.

The blood samples were collected from the cervical vein into both K_3EDTA vacuum tubes and clot activator vacuum tubes (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ, USA) from 2 pigs in each pen at the beginning, and the same pigs were sampled at the end of the experiment. The concentrations of red blood cell (RBC) count, white blood cell (WBC) count and lymphocyte counts in the whole blood and IgG, albumin, and total protein concentrations in the serum were determined. All of the blood cell parameters (RBC, WBC and lymphocytes) and serum parameters (IgG) were assessed using an automatic blood analyzer (ADVIA 120, Bayer, USA).

To evaluate the incidence of diarrhea, the numbers of diarrheaic pigs per pen were collected at daily intervals during the experiment. Diarrhea ratio = total number of pigs with diarrhea/(whole number of experimental pigs×trial days) (Wang et al., 2007).

Statistical analyses

In this experiment, all data were analyzed as a completely randomized design using the GLM procedure of SAS (1996). The model included the effects of the blocks (replication) and treatment groups. The pen was established as the experimental unit. Variability in the data was expressed as the standard error (SE) of the mean, and the level of significance was set at 5%.

RESULTS AND DISCUSSION

Table 2 shows the effects of dietary cupric sulfate and cupric methionate on growth performance in growing pigs. Over the entirety of the 35-d nursery period, ADG, ADFI, and G/F ratios were not affected significantly by the experimental diet ($p>0.05$).

Table 2. Effects of organic and inorganic copper on growth performance over the entire 5-week period in grower pigs

Item	CON[1]	T1[1]	T2[1]	T3[1]	T4[1]	SE[2]
ADG (kg)	0.725	0.747	0.762	0.740	0.727	0.019
ADFI (kg)	1.818	1.896	1.910	1.906	1.794	0.054
G/F	0.399	0.394	0.399	0.388	0.405	0.008

[1] Abbreviated CON, basal diet; T1, basal diet+67 ppm inorganic copper; T2, basal diet+134 ppm inorganic copper; T3, basal diet+67 ppm organic copper; T4, basal diet+134 ppm organic copper.
[2] Pooled standard error.

The effects of Cu supplements on DM and N digestibilities are shown in Table 3. The DM and N digestibilities were not influenced by the addition of Cu at the end of day 21. However, at the end of day 35, DM digestibility was improved in the T1, T2, T3 and T4 treatment groups ($p<0.05$) as compared with the CON treatment groups, which indicates that both $CuSO_4$ and CuMet exert significant effects on DM digestibility. N digestibility was increased only in the T3 treatment groups as compared with the CON treatment group ($p<0.05$).

Hawbaker et al. (1961) and Bunch et al. (1965) suggested that copper functions in an antibiotic-like manner, by influencing microbial growth in the intestine. This indicates that high Cu levels exert an "antibiotic-like effect" on the microflora within the intestinal tract (Braude, 1967; Cromwell, 2001). However, the mechanism inherent to the limited antibacterial properties of the pharmacological concentrations of Cu in swine diets remains to be elucidated (Apgar et al., 1995; Hill et al., 2000). This may explain why the growth response to high Cu as $CuSO_4$, which is similar to the feeding of antibiotics for growth promotion, does not occur in 100% of cases. As far as we know, antibiotics work only in cases in which the pigs are in generally poor physical condition. However, in our study, the pigs evidenced excellent growth characteristics, making this "antibiotic-like effect" less obvious. Similar results have been chronicled in the published research of Stansbury et al. (1990), who conducted three experiments in order to evaluate the growth promotional levels of $CuSO_4$ for weanling swine, and detected no increases in ADFI or ADG in pigs fed with 125 or 250 ppm Cu as $CuSO_4$ in any of the experiments. The only response in feed efficiency was an increase in the gain:feed ratio in one experiment for pigs fed on 250 ppm Cu as compared with 62.5 or 125 ppm Cu in the form of $CuSO_4$. Dove et al. (1990) reported that no effects of Cu supplementation on ADG, ADFI, and G/F ratios in their experiments with growing pigs. In one experiment, Smith et al. (1997) also determined that the 28-d growth performance of weanling swine fed on 250 ppm Cu in the form of $CuSO_4$ on a commercial farm did not differ from that observed in the control group.

At the end of the experiment, the fecal pH value in pigs fed on 67 ppm Cu in the form of cupric sulfate significantly decreased, as compared with what was observed in the CON and CuMet treatments ($p<0.05$), which meant that the fecal acidity level had been attenuated (Table 4).

The blood cell counts of IgG, WBC, RBC, and lymphocytes were not affected ($p>0.05$) by $CuSO_4$ or CuMet (Table 5). Smith et al. (1996) reported that the inclusion of Cu in the diet did not influence lymphocyte proliferation. Some previous researches (Robscheit-Robbins and Whipple, 1942; Bunch et al., 1965) demonstrated that high dietary level of copper resulted depression of hemoglobin which leaded to the anemia.

The effects of organic and inorganic copper on diarrhea in growing pigs was studied and presented in Figure 1. Supplemental 134 ppm $CuSO_4$ and 67 and 134 ppm CuMet significantly reduced the diarrhea ratio, by 57.1% ($p<0.05$), 56.0% ($p<0.05$) and 56.0% ($p<0.05$), respectively, as compared with the control treatment. No significant

Table 3. Effects of organic and inorganic copper on nutrients digestibility in grower pigs

Item	CON[1]	T1[1]	T2[1]	T3[1]	T4[1]	SE[2]
Dry matter						
3 weeks	0.790	0.792	0.806	0.802	0.804	0.651
5 weeks	0.789 [b]	0.808 [a]	0.814 [a]	0.816 [a]	0.817 [a]	0.793
Nitrogen						
3 weeks	0.804	0.807	0.803	0.804	0.802	0.492
5 weeks	0.806 [b]	0.816 [ab]	0.812 [ab]	0.828 [a]	0.816 [ab]	0.564

[1] Abbreviated CON, basal diet; T1, basal diet+67 ppm inorganic copper; T2, basal diet+134 ppm inorganic copper; T3, basal diet+67 ppm organic copper; T4, basal diet+134 ppm organic copper.
[2] Pooled standard error.
[a, b] Means in the same row with different superscripts differ ($p<0.05$).

Table 4. Effects of organic and inorganic copper on fecal pH value and fecal Cu concentrations (DM basis) over the entire 5-week period in grower pigs

Item	CON1	T11	T21	T31	T41	SE2
pH						
0 weeks	6.13	6.09	6.13	6.02	6.05	0.09
3 weeks	5.97	5.97	5.97	5.95	5.99	0.05
5 weeks	5.89[a]	5.79[b]	5.85[ab]	5.91[a]	5.89[a]	0.10
Fecal Cu (mg/kg)	48.5[b]	285.5[a]	327.4[a]	96.7[b]	150.2[b]	28.21

[1] Abbreviated CON, basal diet; T1, basal diet+67 ppm inorganic copper; T2, basal diet+134 ppm inorganic copper; T3, basal diet+67 ppm organic copper; T4, basal diet+134 ppm organic copper.
[2] Pooled standard error.
[a, b] Means in the same row with different superscripts differ ($p<0.05$).

Table 5. Effects of organic and inorganic copper on blood characteristic in grower pigs

Item	CON[1]	T1[1]	T2[1]	T3[1]	T4[1]	SE[2]
IgG						
0 weeks	616.00	520.00	513.80	498.80	472.60	44.17
5 weeks	598.00	611.60	679.00	623.60	711.00	64.31
WBC						
0 weeks	16.22	16.15	16.73	12.50	14.92	1.37
5 weeks	17.50	16.39	16.57	15.49	16.93	0.97
RBC						
0 weeks	6.70	6.71	6.48	6.44	6.58	0.24
5 weeks	7.38	7.05	7.49	7.21	7.62	0.23
Lymphocyte						
0 weeks	50.20	51.80	52.00	53.80	49.40	4.79
5 weeks	70.60	62.40	70.20	72.00	60.80	4.57

[1] Abbreviated CON, basal diet; T1, basal diet+67 ppm inorganic copper; T2, basal diet+134 ppm inorganic copper; T3, basal diet+67 ppm organic copper; T4, basal diet+134 ppm organic copper.
[2] Pooled standard error.

differences were observed between the Cu- sulfate and Cu-methionine treatment groups.

Fecal Cu concentration was significantly lower ($p<0.05$) in the CON, T3, and T4 treatment groups than in the T1 and T2 treatment groups (Table 4). This indicates that the CuMet treatments reduced the fecal Cu concentration as compared with the $CuSO_4$ treatments. Some studies in which organically bound Cu has been supplemented into swine diets showed that more Cu can be absorbed and retained, and less can be excreted, than the pigs fed on Cu in the form of $CuSO_4$ (Fouad, 1976; Apgar et al., 1995; Veum et al., 2004). Fouad (1976) demonstrated that metals in a complexed form are more efficiently absorbed, and their retention in the body is higher than that of similar minerals in their inorganic forms. On the basis of these results, fecal Cu concentration for pigs receiving the complexed Cu should have been higher than that of pigs receiving $CuSO_4$.

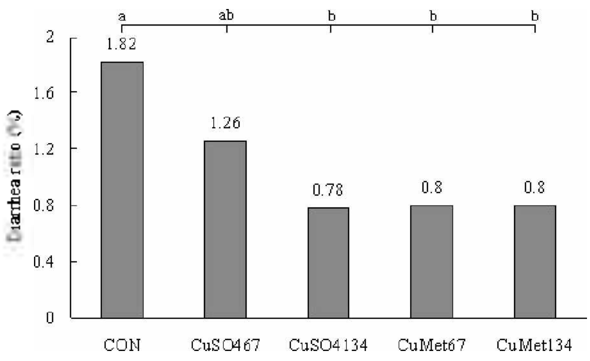

Figure 1. Effect of organic and inorganic copper on diarrhea appearance (diarrhea ratio) in grower pigs. Different letters mean differ significantly ($p<0.05$).

CONCLUSIONS

Both cupric sulfate and cupric methionate both improve DM digestibility in growing pigs. Therefore, considering that CuMet supplementation can attenuate fecal acidity, diarrhea incidence, and fecal Cu concentration, this form of copper additive may be more environmentally friendly than $CuSO_4$ supplements.

REFERENCES

AOAC. 1995. Official Methods of Analysis (16th Ed.). Association of Official Analytical Chemists, Washington, DC.

Apgar, G. A., E. T. Kornegay, M. D. Lindeman and D. R. Notter. 1995. Evaluation of copper sulfate and a copper lysine complex as growth promoters for weanling swine. J. Anim. Sci. 73:2640-2646.

Baker, D. H. and C. B. Ammerman. 1995. Copper bioavailability.

In: Bioavailability of Nutrients for Animals (Ed. C. B. Ammerman, D. H. Baker and A. J. Lewis). Amino Acids, Minerals, and Vitamins, Academic Press, San Diego, CA, pp. 127-156.

Barber, R. S., R. Braude, K. G. Mitchell and J. Cassidy. 1955. High copper mineral mixture for fattening pigs. Chem. Ind. (Lond.) 21:601.

Bowland, J. P. 1990. Copper as a performance promoter for pigs. Pig News Infor. 11:163.

Braude, R. 1967. Copper as a stimulant in pig feed (*cuprum pro pecunia*). World Rev. Anim. Prod. 3:69-81.

Braude, R. 1975. Copper as a performance promoter in pigs. In: Farming Symp. Development Assoc., London.

Bunch, R. J., J. T. McCall, V. C. Speer and V. W. Hays. 1965. Copper supplementation for weanling pigs. J. Anim. Sci. 24:995.

Cromwell, G. L., S. T. Stahly and H. J. Monegue. 1989. Effects of source and level of copper on performance and liver copper stores in weanling pigs. J. Anim. Sci. 67:2996-3002.

Cromwell, G. L., T. S. Stahly and W. D. Williams. 1981. Efficacy of copper as a growth promotant and its interaction with sulfur and antibiotics for swine. Proc. Distill. Feed Conf. 36:15.

Cromwell, G. L. 2001. Antimicrobial and promicrobial agents. Pages 401-426 in Swine Nutrition. 2nd ed. (Ed. A. J. Lewis and L. L. Southern). CRC Press LLC, Boca Raton, FL.

Dove, C. R. and R. C. Ewan. 1990. Effect of excess dietary copper, iron or zinc on the tocopherol and selenium status of growing pigs. J. Anim. Sci. 68:2407-2413.

Downs, K. M., J. B. Hess, K. S. Macklin and R. A. Norton 2000. Dietary zinc complex and vitamin E for reducing cellulities incidence in broilers. J. Appl. Poult. Res. 9:319-323.

Fouad, M. T. 1976. The physiochemical role of chelated minerals in maintaining optimal body biological functions. J. Appl. Nutr. 28:5.

Guo, R., P. R. Henry, R. A. Holwerda, J. Cao, R. C. Littell, R. D. Miles and C. B. Ammerman. 2001. Chemical characteristics and relative bioavailability of supplemental organic copper sources for poultry. J. Anim. Sci. 79:1132-1141.

Hawbaker, J. A., V. C. Speer, V. W. Hays and D. V. Catron. 1961. Effects of copper sulfate and other chemotherapeutics in growing swine rations. J. Anim. Sci. 20:163-167.

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, L. L. Southern and T. L. Veum. 2000. Growth promotion effects and plasma changes from feeding high dietary concentrations of zinc and copper to weanling pigs (regional study). J. Anim. Sci. 78:1010-1018.

National Research Council. 1998. Nutrient requirements of swine. 10th revised ed. National Academic Science, Washington, DC, USA.

Paik, I. K., S. H. Seo, J. S. Um, M. B. Chang and B. H. Lee. 1999. Effects of supplementary copper-chelate on the performance and cholesterol level in plasma and breast muscle of broiler chickens. Asian-Aust. J. Anim. Sci. 12:794-798.

SAS Inc., 1996. SAS® User's Guide. Version 6.12 Edition SAS Inc., Cary, NC, USA.

Smith II, J. W., J. D. Arthington, M. D. Tokach, F. Blecha, R. D. Goodband, J. L. Nelssen, B. T. Richert, K. Q. Owen, J. R. Bergstrom and W. B. Nessmith, Jr. 1996. The effect of supplemental mineral regimen on weanling pig blood and immune parameters, and liver and plasma mineral concentrations. J. Anim. Sci. 74 (Suppl. 1):57(Abstr.).

Smith II, J. W., M. D. Tokach, R. D. Goodband, J. L. Nelssen and B. T. Richert. 1997. Effects of interrelationships between zinc oxide and copper sulfate on growth performance of early-weaned pigs. J. Anim. Sci. 75:1861-1866.

Stansbury, W. F., L. F. Tribble and D. E. Orr, Jr. 1990. Effect of chelated copper sources on performance of nursery and growing pigs. Anim. Sci. 68:1318-1322.

Veum, T. L., M. S. Carlson, C. W. Wu, D. W. Bollinger and M. R. Ellersieck. 2004. Copper (L)proteinate in weanling pig diets for enhancing growth performance and reducing fecal copper excretion compared with copper sulfate. J. Anim. Sci. 82:1062-1070.

Wang, Y. Z., T. Z. Shan, Z. R. Xu, J. Feng and Z. Q. Wang. 2007. Effect of the lactoferrin (LF) on the growth performance, intestinal microflora and morphology of weanling pigs. Anim. Feed Sci. Technol. 135:263-127.

Yu, B., W. J. Huang and P. W. Chiou. 2000. Bioavailability of iron from amino acid complex in weanling pigs. Anim. Feed Sci. Technol. 86:39-52.