European Journal of Science, Innovation and Technology

ISSN 2786-4936

Vol. 1 No. 6 (2021)

Effects of Animal Drinking Water Mineral Content on the Estimation of Magnesium, Copper, Iron and Manganese Digestibility and their Endogenous Losses in Growing Pigs by the Difference Method

Ntinya C. Johnson[1], Victor M. Ogbamgba[1] and James T. Mbachiantim[2]
[1]Rivers State University, Port Harcourt, Department of Animal Science, Nigeria
[2]Federal University of Agriculture, Makurdi, Department of Nutrition and Dietetics, Nigeria

Abstract. 12 Yorkshire barrows with initial BW of 23.9 ± 1.1 kg were assigned to two dietary treatments with six replications per treatment. The two diets were formulated in accordance with the principles of the difference method. The minerals' apparent and true digestibility (AD and TD) values as well as their endogenous fecal losses (EFL) were to be investigated. The pigs were randomly allotted to their individual feeder pens which enabled individual pig fresh fecal sample collections. The experiment was designed as a completely randomized design (CRD) and lasted for 15d consisting of 10d adaptation to diets and 5d of spot fecal collections. Results showed negative AD values for all the minerals studied, namely Mg, Cu, Fe and Mn. Therefore, their TD values and their EFL could not be estimated because of their negative AD values. Further investigations revealed that the negative AD values of the minerals were due to high levels of calcium (Ca) in the animals' drinking water during the study period. It was concluded that determination of apparent and true digestibility values of Mg/trace minerals and their endogenous losses also require measurement of drinking water mineral sources and intakes as to avoid excess mineral intakes in order to avoid mineral antagonisms and digestibility.

Key words: Digestibility, Mg/Trace minerals, Endogenous Losses, Water Mineral Content and the Pig

Introduction

Magnesium and trace mineral losses from animals into the environment are inevitable. However, the levels of these minerals released via animal manures into the environment causing eutrophication (Mallin, 2000) can be significantly curtailed or reduced via diet manipulation. According to Tamminga (2003), this can be achieved by matching animal needs of these minerals by avoiding provision of the nutrients beyond animal requirements (NRC, 2012). One of the means by which this is realizable is by the determination of the true digestible values of these minerals for use in diet formulation in place of the current apparent digestible values in the pig's diet.

As stated previously, this would result in supplying these nutrients based on the true animal requirement that subsequently leads to their solubility and absorption and thus reduce their contents in the animal's manure. Accumulation of trace minerals in the soil particularly Cu and Fe leads to medium and long term toxicity effects in plants as well as soil micro-organisms (Dourmad & Jondreville, 2007). To this point, it has been shown that more than 90% of ingested nutrients, particularly the micro-minerals by pigs are excreted in manure (Aarnink & Verstegen, 2007). These observations are revelations that in the near future legislation on trace mineral contents in swine diets may be enacted to mitigate their effects on environmental pollution and eutrophication (Mallin, 2000).

Therefore, the quantification of true Mg and trace minerals' digestibilities and their endogenous losses would help match animal needs with requirements thereby avoiding their excessive inclusions in diets leading to their high levels of excretions in the manure into the environment. The usefulness of this finding therefore may not be limited to swine diets alone as it can also be useful in human nutrition, particularly for vegetarians and lacto-vegetarians whose diets composed mainly of plant-based ingredients; as the pig model has been demonstrated to be a useful model to elucidate mechanisms governing dietary influences on mineral metabolism and absorption in humans (Paterson *et al.*, 2008). It is also imperative to note that when the dietary supplies match animal requirements, they would be solubilized in the stomach thereby preventing the chelating effects of phytate on the minerals which triggers and subsequently exacerbates the formation of insoluble-phytate-mineral-complexes in the small intestine resulting in excessive endogenous losses of the minerals in addition to the dietary sources in the manure (Lei & Pores, 2003; Montminy *et al*, 2007). To our knowledge there is no information to date on the true digestibility value of Mg, Cu, Fe and Mn in a corn/SBM-based diet for growing pigs. Therefore, the objectives of this study were to estimate the true digestibility and the endogenous losses of these minerals associated with a corn/SBM-based diet for growing pigs by the difference method.

Materials and Methods

Animals, Housing and Experimental Design

12 Yorkshire growing barrows with an average initial BW of 23.9 ± 1.1 (mean ± SD) kg were acquired from Arkell Swine Research Station, University of Guelph, Ontario and used for the study, designed as a CRD. The animals were housed in tender-foot^{TM} plastic floor pens (1.5 x 2.1 m) with smooth transparent plastic sides in a room that was mechanically ventilated to provide an ambient temperature of 20 – 22^{0}C. All procedures used in the management of the pigs were reviewed and approved by the University of Guelph Animal Care Committee and animals were cared for with strict compliance to the guidelines of the Canadian Council on Animal Care (CCAC, 1993).

Experimental Diets

Two dietary treatments were formulated at 100% referred to as the high-nutrient (HN) diet and 60% referred to as the low-nutrient (LN) diet, respectively of the NRC, (1998) requirements for Mg, Cu, Fe and Mn for the growing pig according to the principles of the difference method, involving 40% (100 – 60) difference. Accordingly, therefore, the LN diet was formulated by partially replacing corn, SBM, limestone, dicalcium phosphate and vitamin-mineral premix with increased cornstarch and solka-flock^{TM} to balance for DE and NDF contents (Table 1). Titanium oxide was added at 0.3% of diet DM as an indigestible marker (Table 1).

Table 1. Diet formulation for measuring true Mg, Cu, Fe, and Mn digestibility and their endogenous losses in grower pigs (20 – 50 kg) by the substitution method

	Diets	
Ingredients (Kg)	High nutrient (HN)	Low nutrient (LN)
Soybean meal	27.07	16.24
Corn	66.00	39.60
Cornstarch	2.71	36.93
Solka-flock (100% cellulose)	0.00	3.98
L-Lys-HCL (79% L-Lys)	0.17	0.102
L-threonine (100%)	0.05	0.03
Animal fat	0.40	0.40
Limestone (38.5% Ca)	0.84	0.50
Dicalcium phosphate	0.86	0.52
Vit-mineral premix[1]	0.50	0.30
Antibiotic mixture	0.00	0.00

Titanium oxide	0.30	0.30
Total (100 kg)	100.00	100.00
Calculated nutritive values (as-fed basis)		
DE (MJ/Kg)	14.57	14.81
CP (%)	17.33	10.40
Total Ca (%)	0.62	0.37
Total P (%)	0.52	0.31
Total Ca/total P ratio	1.19	1.19
NDF (%)	9.94	9.94
ADF (%)	4.39	2.64

[1] Vit-mineral premix contained vit. A, 2,000,000IU; vit. D_3, 200,000IU; vit. E, 8,000IU; vit. K, 500mg; pantothenic acid 3,000mg; riboflavin 1,000mg; folic acid, 400mg; niacin 5,000mg; thiamine 300mg; pyridoxine 300mg; vitamin B_{12}5,000mcg; biotin, 40,000mcg; Se 60mg; choline 100,000mg; I, 100mg; Cu, 3,000mg; Fe, 20,000mg; Mn, 4,000mg; Zn, 21,000mg.

Diet and Fecal Samples Collections and Processing

Feed samples were collected immediately after each diet mixing and stored in sealed sample bags at 4^0C. They were later ground in a Wiley mill through a 1-mm screen and stored again until analysis. From the 11th to the 15th d in the study, fecal samples were spot collected from each pen at 2-h intervals for all animals on the two dietary experimental treatments. Fecal samples collected were sealed in the fecal sample containers and were immediately frozen at -23^0C. They were later freeze-dried and ground in a Wiley mill through a 1-mm screen and also stored at 4^0C until analysis.

Chemical Analyses

Dry matter of diet and feces were determined according to the method of AOAC (2000). Titanium oxide in diet and feces were measured according to the method of Short *et al.* (1996). Dietary mineral contents of the two diets and feces namely: Mg, Cu, Fe and Mn were also determined according to the method of AOAC (2000).

Calculations and Statistical Analyses

The apparent fecal Mg, Cu, Fe and Mn digestibilities (AFD; %) were first computed using the marker technique according to equation 1:

$$AFD = \{(N_{diet}/M_{diet} - N_{feces}/M_{feces})/N_{diet}/M_{diet})\} \tag{1}$$

Where N_{diet} = the concentration of nutrient in the diet (%), M_{diet} = the concentration of titanium marker in the diet (% titanium), N_{feces} = the concentration of nutrient in the feces (%) and M_{feces} = the concentration of marker in feces (%). All percentages were on a DM basis.

Based on average AFD of Mg, Cu, Fe and Mn true fecal digestibilities (TFD; %) were to be estimated according to equation 2 as:

$$TFD = \{(AFD_{diet1}\ N_{diet1} - AFD_{diet2}N_{diet2})/(N_{diet1} - N_{diet2})\} \tag{2}$$

Where diet 1 and diet 2 are the HN and LN diets, respectively.

Endogenous fecal losses (EFL; g/kg DMI of Mg, Cu, Fe and Mn were to be estimated as in equation 3:

$$EFL = (TFD_{diet} - AFD_{diet})N_{diet}10 \tag{3}$$

Data were subjected to ANOVA using PROC GLM of SAS according to:

$Y_{ij} = \mu + Di + E_{ij}$: where Y_{ij} is the observation, μ is the overall mean common to all treatments, *Di* is the effect of the ith diet and E_{ij} is the error term. Furthermore, homogeneity of variances across the two diets was tested for and confirmed ($P > 0.05$) by Levene's test using SAS. The pig was the experimental unit and an *α-level* of *0.05* was used for all statistical comparisons to represent significance.

Results and Discussion

Table 2. Analyzed dietary mineral contents of diets

Item	Diet 1 (amount/kg diet)	Diet 2 (amount/kg diet)
Mg	1.6 g	0.88 g
Cu	30 mg	6.6 mg
Fe	260 mg	130 mg
Mn	51 mg	36 mg

In the vitamin-mineral premix Cu was provided by copper sulphate (25%), Fe was provided by ferrous sulphate (30%), Mn was provided by manganese sulphate (31.5%).

The final actual analyzed dietary contents of Mg, Cu, Fe and Mn for the HN and LN diets are shown in Table 2. Results of the apparent fecal digestibility values of Mg, Cu, Fe and Mn in the experimental diets are presented in Table 3. Apparent digestibility values of the minerals studied were negative (Table 3). These negative apparent mineral digestibilities observed in this study have also been demonstrated by Jongbloed *et al.* (2004) and Woyengo *et al.* (2009).

Table 3. Apparent fecal digestibility values of Mg, Cu, Fe, and Mn in the experimental diet measured for the growing pig fed corn/SBM-based diets by the substitution method

Item	Diet 1 (n = 6)	Diet 2 (n = 6)	*P - Value*
	%		
Mg	-29.70 ± 3.68	-10.17 ± 3.1	*0.0023*
Cu	-10.68 ± 1.85	-139.2 ± 4.42	*P < .0001*
Fe	-20.14 ± 9.15	-21.17 ± 2.52	*0.9164*
Mn	-22.29 ± 3.39	18.70 ± 2.08	*P < .0001*

The true digestibility values for Mg, Cu, Fe and Mn and their endogenous losses could not be estimated because of their negative apparent digestibility values. It had been shown that more than 90% of ingested trace-minerals are excreted in the pig manure (Aarnink and Verstegen, 2007). This had also been attributed to the presence of inhibiting or promoting dietary components (Liu *et al.*, 2000). For instance, Woyengo *et al.* (2009) showed that increasing dietary phytate level significantly reduced Mg digestibility from 29% to a negative value by increasing Mg endogenous losses.

However, the negative apparent digestibility values estimated for these minerals in this current study were somewhat surprising initially, since their dietary contents were within requirements and the calculated Ca to P ratio was close at 1.19: 1 (Table 1). To this extent, the dietary ratio of Ca to P has been shown to be very important in Mg digestibility (Liu *et al.*, 2000) and trace-minerals digestibilities (Hu *et al.*, 2010). In fact, in the study of Hu *et al.* (2010), it was shown that as the ratio of dietary Ca to P gets wider, Cu, Fe and Mn showed negative apparent digestibility values to a greater extent than values observed in this current study.

Traditionally, mineral contents of the animal drinking water are not considered in diet formation. One of the striking and profound findings in our study strongly disagrees with this tradition in swine production practices on most commercial hog farms. To this point therefore, the mineral contents of our pigs’ drinking water, especially for Ca, Mg and P were very revealing (Table 4).

Table 4. Ca, P, Mg, Fe, Cu and Mn content of pig drinking water

Mineral	**Water content (ppm)**
Ca	51
P	< 0.10
Mg	15
Fe	< 0.01
Cu	0.08
Mn	0.01

According to NRC, (2012) growing pigs will consume 4 kg of water per kg of feed on an as-fed basis. A study (Li *et al.*, 2005) estimated average water intake of 5.38 kg water/kg DMI. Additionally, growing pigs normally have a high tendency to drink water, especially in the afternoon when feed is restricted as in this study, due to a desire for abdominal fill (NRC, 2012). Therefore, although the exact volume of water our pigs consumed was not measured there is no doubt that the quantity consumed by the pigs for the entire experimental duration would have been high enough to provide excess Ca (7.3g; 4.5g) and Mg (2.4g; 1.6g)/kg DMI for the HN and LN diets, respectively, and to a lesser extent P, Zn, Cu, Mn and Fe for the animals, particularly P. This therefore would have resulted in widening the ratio of Ca to P for the animals, coupled with Mg a well-known antagonist of Ca and P (O'Dell, 1997).

This necessitated the need to investigate the effect of mineral intakes from both ingested feed and drinking water sources on mineral digestibility. When total estimated mineral intakes from diet and water sources were considered in the estimation of mineral digestibilities, there was little effect compared to digestibilities when water source minerals were ignored (Table 5).

Table 5. Effect of water mineral intake on apparent mineral digestibility

Mineral	**Diet 1 (n = 6)**	**Diet 2 (n = 6)**
	%	
Mg	-25.35	-3.63
Cu	-9.60	-128.91
Fe	-20.13	-21.13
Mn	-22.20	18.78

In this study, P content of drinking water did not appear to be the problem since it was less than 0.1ppm. However, the amount of Ca consumed via water significantly altered the dietary ratio of Ca to P from 1.19: 1 to approximately 1.26: 1 and 1.25: 1 for the HN and LN diets, respectively and possibly causing more Ca to precipitate and instigate a series of antagonistic interactions, especially in the presence of P and Mg with the dietary minerals rendering them unavailable for solubility and absorption (Liu *et al.*, 2000; Hu *et al.*, 2010). Solubility of these minerals is required for absorption. Their insolubility would therefore decrease the animal's ability to digest the minerals (Knowlton *et al.*, 2004) coupled with some possible negative hormonal feedback mechanisms because of their effects on increasing the pH of the stomach medium (Montminy *et al.*, 2007; Shoback & Sellmeyer, 2010).

Our findings of a negative effect of drinking water minerals on mineral digestibility have been substantiated to by previous researchers (Flipot & Queller, 1988; Castillo *et al.*, 2007). Therefore, estimates from our current study alert us to the importance of mineral content of drinking water to be known and if possible included in the provision of mineral supplies to the animal or if toxic should be treated before accessibility by the animals for quality animal productivity. Thus, in the overall water source mineral should always be considered where appropriate in feed formulation to better guide the feed formulator in meeting animal mineral requirements as to avoid excess minerals intake to enable the reduction of minerals export from the pig barn into the environment.

Conclusions

It is thus concluded that determination of true digestibility values of trace minerals and their endogenous losses require measurement of mineral intakes via the animal drinking water.

References

Aarnink, A. J. A. & Verstegen, M. W. A. (2007). Nutrition, key factor to reduce environmental load from pig production. *Livest. Sci., 109*, 194-203.

AOAC. (2000). *Official Methods of Analysis* (17th ed.). Assoc. Off. Anal. Chem. Arlington, VA.

Castillo, A. R. Santos, J. E. & Tabone, T. J. (2007). Estimation of mineral balances in dairy herds including minerals in the drinking water. *Calif. Agric., 61*(2), 90-95.

CCAC. (1993). *Canadian Council on Animal Care Guide. Guide to the care and use of experimental animals* (Vol. 1, 2nd ed.). CCAC Ottawa, ON.

Dourmad, J. Y. & Jondreville, C. (2007). Impact of nutrition on nitrogen, phosphorus, Cu and Zn in pig manure and on emissions of ammonia and odours. *Livest. Sci., 112*, 192-198.

Flipot, P. M. & Quellet, G. (1988). Mineral and nitrate content of swine drinking water in four Quebec regions. *Can. J. Anim. Sci., 68*, 997-1000.

Hu, L. Yang, J., Fang, S. X., Wang, X. G., Shu, Z. Y., et al. (2010). Estimating an optimal ratio of true digestible Ca to P ratio in corn-rough-soybean meal-based diets for 20-50 kg growing pigs. *J. Food Agric. & Environ., 8*, 556-562.

Jongbloed, A. W., van Diepen, J. M., Kemme, P. A., & Broz, J. (2004). Efficacy of microbial phytase on mineral digestibility in diets for gestating and lactating sows. *Livest. Prod. Sci., 91*, 143-155.

Knowlton, K. F., Radcliffe, J. S., Novak, C. L., & Emmerson, D. A. (2004). Animal management to reduce phosphorus losses to the environment. *J. Anim. Sci., 82*(E.Suppl.), E173-E195.

Lei, X. G. & Pores, J. M. (2003). Phytase enzymology, applications and biotechnology. *Biotechnol. Letters., 25*, 1787-1794.

Li, Y. Z., Chenard, L., Lemay, S. P., & Gonyou, H. W. (2005). Water intake and wastage at nipple drinkers by growing-finishing pigs. *J. Anim. Sci., 83*, 1413-1422.

Liu, J., Bollinger, D. W., Ledoux, D. R., & Veum, T. L. (2000). Effects of dietary calcium: phosphorus ratios on apparent absorption of calcium and phosphorus in the small intestine, cecum and colon of pigs. *J. Anim. Sci., 78*, 106-109.

Mallin, M. A. (2000). Impacts of Industrial Animal Production on Rivers and Estuaries. *Am. Sci., 88*(1), 26-37.

Montminy, M. P., Jondreville, C., Lescoat, P., Meschy, F., Pomar, C., et al. (2007). First step of a model of calcium and phosphorus metabolism in growing pigs: Fate of ingested phosphorus in the stomach. *Livest. Sci., 109*, 63-65.

NRC. (1998). *Nutrient Requirements of Swine* (10th ed.). Natl. Acad. Press, Washington, D. C.

NRC. (2012). *Nutrient Requirements of Swine* (11th ed.). Natl. Acad. Press, Washington, D. C.

O'Dell, B. L. (1997). Mineral-Ion Interaction as Assessed by Bioavailability and Ion Channel Function. In B. L. O'Dell & R. A. Sunde (Eds.), *Handbook of nutritionally essential mineral elements* (pp. 641-659). Marcel Dekker, Inc., New York.

Patterson, J. K., Lei, X. G., & Miller, D. D. (2008). The pig as an experimental model for elucidating the mechanisms governing dietary influence on mineral absorption. *Exp. Biol. Med., 233*, 651-664.

Shoback, D. M. & Sellmeyer, D. E. (2010). Disorders of the Parathyroids and calcium and phosphorus metabolism. In S. J. McPhee & G. D. Hammer (Eds.), *Pathophysiology of Disease* (6th ed., pp. 465-496). McGraw-Hill Companies Inc.

Short, F. J., Gorton, P., Wiseman, J., & Boorman, K. N. (1996). Determination of titanium dioxide added as an inert marker in chicken digestibility studies. *Anim. Feed Sci. Technol.*, *59*, 215-221.

Tamminga, S. (2003). Pollution due to nutrient losses and its control in European and production. *Livest. Prod. Sci.*, *84*, 101-111.

Woyengo, T. A., Cowieson, A. J., Adeola, O., & Nyachoti, C. M. (2009). Ileal digestibility and endogenous flow of minerals and amino acids: responses to dietary phytic acid in piglets. *Brit. J. Nutr.*, *102*, 428-433.