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Estimation of True Selenium and Zinc Digestibility and their Endogenous Outputs in Growing Pigs Fed Corn/Soybean Meal-Based Diets by the Substitution Method

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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 substitution method. Diets' dry matter (DM) digestibility, animals' performance, the minerals' apparent and true digestibility (AD and TD) values as well as their endogenous fecal losses (EFL) were 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. Results showed that the DM digestibility of diet 1 (78.7%) was significantly ($P < 0.05$) lower than that of diet 2 (85.6%). However, animals on diet 1 had better ($P < 0.05$) average daily gain (ADG) and feed efficiency (FE) compared with animals on diet 2. The AD values of Se (73.9%) and Zn (9.5%) were significantly ($P < 0.05$) lower than their TD values of 82.1% and 15%, respectively. Se and Zn EFL were 0.00004mg and 0.01mg/kg of DMI, respectively. It was concluded that the TD values of these minerals be employed in diet formulations instead of their AD values as to avoid their mutual antagonism during metabolism and thus reduce their levels in the animal manure.

INTRODUCTION

Although trace mineral losses from animals into the environment are inevitable (Tamminga, 2003), there is a need to decipher means of supplying them based on the true animal requirements as to reduce their contents in the animal's manure. Accumulation of trace minerals in the soil particularly Zn causes medium to long term toxicity effects to plants and soil micro-organisms (Dourmad and Jondreville, 2007). To this end, it has been shown that more than 90% of ingested trace minerals by pigs are excreted in the manure (Aarnink and Verstegen, 2007). These indices are pointers to the fact that in the near future legislation on trace mineral contents in swine diets may be enacted to mitigate their effects on environmental pollution.

The quantification of true trace minerals' digestibilities and their endogenous fecal 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 leading to excessive endogenous losses of the minerals in addition to the indigestible dietary sources in the manure (Lei and Pores, 2003; Montminy *et al.,* 2007). To our knowledge there is no information to date on the true digestibility of Se and Zn 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 substitution 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 and used in the study, designed as a CRD. The animals were housed in tender-foot™ 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 Se and Zn for the growing pig according to the principles of the substitution method, also known as the difference method involving 40% difference. Accordingly, the LN diet was formulated by partially replacing corn, SBM, limestone, dicalcium phosphate and vitamin-mineral premix with increased cornstarch and solka-flock™ 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 Se and Zn 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.

Computation of Performance Parameters

Feed intake was normalized for both HN- and LN-diets offered at 5% of BW. At the beginning of each day during the 15d duration of study, orts from the previous day were collected dried at 105^{0}C and weights recorded. The difference between dry feed delivered and the next day's orts represented DM consumed by the animal for the day. On the last day of study, all animals were re-weighed to obtain their final BW. Average daily feed intake (ADFI), ADG as well as gain to feed ratio, that is, FE for the study period were thus obtained.

Diet and Fecal Samples Collections and Processing

Feed samples were collected immediately after each diet mixing and stored in sealed sample bags at 4^{0}C. 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^{0}C. They were later freeze-dried and ground in a Wiley mill through a 1-mm screen and also stored at 4^{0}C 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: Se and Zn were also determined according to the method of AOAC (2000).

Calculations and Statistical Analyses

The apparent fecal DM, Se and Zn digestibilities (AFD; %) were first computed using the marker technique according to equation 1:

$$AFD = \{(N_{diet}/M_{diet} - N_{feces}/M_{feces})/N_{diet}/M_{diet})\} \quad \text{(equation 1)}$$

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 Se and Zn, true fecal digestibilities (TFD; %) were estimated according to equation 2 as:

$$TFD = \{(AFD_{diet1}\, N_{diet1} - AFD_{diet2} N_{diet2})/(N_{diet1} - N_{diet2})\} \qquad \text{(equation 2).}$$

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

Endogenous fecal losses (EFL; g/kg DMI of Se and Zn were estimated as in equation 3:

$$EFL = (TFD_{diet} - AFD_{diet}) N_{diet} 10 \qquad \text{(equation 3).}$$

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

All animals consumed their respective diets normally throughout the duration of study and thus also grew during the period. The performance of the animals on the two dietary treatments is presented in Table 2. The ADFI of animals on the two dietary treatments were similar ($P = 0.93$). As expected, the ADG of pigs on the HN diet was superior ($P < 0.0001$) compared with pigs that consumed the LN diet as the nutrients in the HN diet were formulated to meet the nutrient requirements of the animals compared to when they were formulated to be less at least by 40% difference for the LN diet (Table 3) based on the principles of the difference method (Fang *et al.,* 2007). This also resulted in a better ($P < 0.0001$) FE for the HN diet compared with the LN diet.

Table 2: Mean ± SE growth responses of pigs fed high-and low-nutrient corn/SBM-based diets (n = 6)

Item	High-nutrient diet (Diet 1)	Low-nutrient diet (Diet 2)	*P-Value*
ADFI (DM basis; kg/d)	1.3 ± 0.03	1.3 ± 0.03	0.93
Initial BW (kg)	23.7 ± 1.2	24.2 ± 1.1	0.48
Final BW (kg)	32.3 ± 1.3	30.2 ± 1.6	0.03
ADG (kg/d)	0.58 ± 0.01	0.40 ± 0.03	< 0.001
FE (ADG/ADFI)	0.51 ± 0.01	0.35 ± 0.02	< 0.001

Table 3: Analyzed dietary mineral contents of diets

Item	Diet 1 (amount/kg diet)	Diet 2 (amount/kg diet)
Se	0.47 mg	0.19 mg
Zn	150 mg	68 mg

In the vitamin-mineral premix Se was provided by sodium selenite (4.5%) and Zn was provided by zinc sulphate (35.5%).

The final actual analyzed dietary contents of Se and Zn for the HN and LN diets are shown in Table 3. Results of the apparent fecal digestibility values of DM, Se and Zn in the experimental diets are presented in Table 4. Apparent DM digestibility of the HN diet was 79% whereas that of the LN diet was 86% which is significantly ($P < .0001$) higher than that of the HN diet. This might be due to the higher cornstarch content of the LN diet compared to that of HN because the energy of cornstarch is almost 100% digestible compared to that of corn and SBM that are 96 and 92% digestible, respectively (NRC, 2012). NRC, (1998) had also shown apparent digestibility of 10% for Zn and 73.5% on average for Se. These values are in agreement with the findings of this current study (Table 4).

Table 4: Apparent fecal digestibility values of DM, Se and Zn 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*
		%	
DM	78.74 ± 0.66	85.63 ± 0.42	*P < .0001*
Se	73.86 ± 1.0	61.59 ± 1.81	*0.0001*
Zn	9.45 ± 3.31	2.66 ± 1.25	*0.0839*

True fecal digestibility values and fecal endogenous losses of Se and Zn are presented in Table 5. The true fecal digestibility of Se was 82.1% and that of Zn was 15%. Se fecal endogenous loss was 0.00004mg/kg DMI and that of Zn was 0.01mg/kg DMI. The animals therefore demonstrated low endogenous losses of Se and Zn. This might not be unconnected to the animal physiological adaptation in the metabolism of these micro-minerals in response to their micro-dietary inclusion levels in diets (NRC, 2012).

Table 5: The true fecal digestibility values and the fecal endogenous outputs of Se and Zn associated with corn/SBM-based diet for the growing pig measured by the substitution method (n = 12)

Item	**True digestibility (%)**	**Fecal endogenous loss (g/kg DM intake)**
Se	82.08	0.00004 mg
Zn	15.00	0.01 mg

CONCLUSIONS

The true Se and Zn digestibility values in corn/SBM-based diet for the growing pig (20 – 50 kg) BW are 82% and 15%, respectively. These values are significantly higher than their apparent digestibility values. Endogenous fecal losses are 0.00004 mg and 0.01 mg for Se and Zn, respectively. Therefore, the TD values of Se and Zn should be employed in diet formulations to reduce the levels of Se and Zn void in the pig manure.

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