

Evaluation of selenium source on nursery pig growth performance, serum and tissue selenium concentrations, and serum antioxidant status

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ABSTRACT

A total of 3.888 pias (337 × 1050, PIC, Hendersonville, TN; initially 6.0 ± 0.23 kg) were used in a 35-d study. At the time of placement, pens of pigs were weighed and allotted to one of three dietary treatments in a randomized complete block design with a blocking structure including sow farm origin, date of entry into the facility, and average pen body weight. A total of 144 pens were used with 72 double-sided 5-hole stainless steel fence line feeders, with one feeder serving as the experimental unit. For each feeder, 1 pen contained 27 gilts, and 1 pen contained 27 barrows. There were 24 replicates per dietary treatment. Diets were fed in three phases, and all contained 0.3 mg/kg added Se. A common phase 1 diet contained added Se from sodium selenite and was fed in pelleted form to all pigs from day 7 to approximately day 0. Three Se sources sodium selenite, Se yeast, and hydroxy-selenomethionine (OH-SeMet) were used to formulate three experimental diets in meal form for phase 2 (days 0 to 14) and phase 3 (days 14 to 35). During the pre-treatment period (days 7 to 0), there was a tendency (P = 0.097) of a difference in average daily feed intake between treatments, although no significant pairwise differences were observed (P > 0.05). There were no other differences in growth performance between treatments from days 7 to 0. Clinical disease attributed to Streptococcus suis was observed within the trial between days 0 and 14, and water-soluble antimicrobial therapy was administered to all treatment groups for 7 d. From days 0 to 35, pigs fed OH-SeMet tended to have decreased average daily gain (P < 0.10) and increased (P < 0.05) serum and tissue selenium concentration compared to other treatments. There was marginally significant evidence of a source \times day interaction (P = 0.027) for total antioxidant capacity where the numerical increase over time was less for the OH-SeMet than sodium selenite or selenium yeast treatments. There was no difference (P > 0.05) in antioxidant status as measured by serum glutathione peroxidase or thiobarbituric acid reactive substances assay between treatments. In summary, compared to sodium selenite and selenium yeast, OH-SeMet may have a greater bioavailability as indicated by increased serum and tissue selenium concentration; however, antioxidant status was similar between treatments and OH-SeMet tended to reduce growth performance compared with pigs fed sodium selenite.

LAY SUMMARY

Selenium (Se) is an essential trace mineral for selenoproteins that are crucial for antioxidant status and all stages of animal growth, and Se deficiency may result in health issues in pigs. To meet the Se requirement, several inorganic or organic Se sources can be added to animal feed. Different Se sources have shown bioavailability differences that affect absorption and storage of Se and may improve animal performance. In this 35-d experiment, a total of 3,888 nursery pigs (initially 6.0 kg) were used to test three different Se sources added at 0.3 mg/kg concentration. One inorganic Se source (sodium selenite) and two organic Se sources (Se yeast and hydroxy-selenomethionine [**OH-SeMet**]) were used to formulate three experimental diets. Overall, pigs fed OH-SeMet had decreased gain and increased serum and tissue Se concentration compared with other treatments. There were similar results in measures of antioxidant status tested between treatments. In summary, compared to sodium selenite and Se yeast, OH-SeMet may have greater bioavailability as indicated by increased serum and tissue Se concentration; however, antioxidant status was similar between treatments and OH-SeMet tended to reduce growth performance compared with pigs fed sodium selenite.

Key words: antioxidant status, growth, nursery pigs, selenium

INTRODUCTION

Selenium (Se) is an essential trace mineral for selenoproteins that are crucial for all stages of animal production because of their roles as antioxidant enzymes (e.g., glutathione peroxidase [GSH-Px] and thioredoxin reductase) that protect cells from oxidative damage (NRC, 2012; Hosnedlova et al., 2017). Selenium deficiency may result in sudden death, pale muscle, liver necrosis, mulberry heart disease, and damage to lungs and gastrointestinal tissues in pigs (NRC, 2012; Hosnedlova et al., 2017). Therefore, meeting the dietary Se requirement is important for animal production, especially in regions that use feed ingredients grown in low Se soils (Hosnedlova et al., 2017). For swine production, the dietary Se requirement ranges from 0.3 mg/kg for nursery pigs to 0.15 mg/kg for growing-finishing pigs and sows (NRC, 2012). Even though Se is an essential trace mineral for pigs, there is a

Received October 30, 2022 Accepted May 9, 2023.

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narrow range between requirement and toxicity (Shini et al., 2015). Excess Se excretion in animal waste can cause environmental pollution (NRC, 2012). Thus, the Food and Drug Administration (FDA) regulates the addition of Se in swine diets, and the maximum level of added dietary Se is 0.3 mg/kg in complete feed for all stages of production (FDA, 2021). To meet this requirement, several inorganic or organic Se sources can be added to animal feed. Sodium selenite is commonly used as the primary inorganic Se source. In nature, organic Se is predominantly found in seleno-amino acid forms where Se replaces the S in sulfur-containing AAs, such as cysteine and methionine, to form selenocysteine and selenomethionine (Shini et al., 2015). Commercial organic Se sources used in animal feed are in the form of selenomethionine, such as Se yeast or hydroxy-selenomethionine (OH-SeMet). Different Se sources have shown bioavailability differences in animals due to structural differences that affect absorption and storage of Se and may improve an animal performance (Shini et al., 2015). Nursery pigs fed 0.3 mg/kg OH-SeMet had increased tissue Se levels compared to pigs fed sodium selenite without a difference in growth performance (Chao et al., 2019). Growing pigs fed organic Se sources (Se yeast or OH-SeMet) had greater (P < 0.05) plasma and muscle Se concentrations compared to pigs fed inorganic Se source (sodium selenite) without differences (P > 0.05) in growth performance (Ilali et al., 2014). However, the differences between these products added to diets formulated with corn and soybean meal originating from low-soil Se regions and fed to nursery pigs are still not clear. Therefore, the objective of this study was to determine the effect of Se source (sodium selenite, Se yeast, and OH-SeMet) included at the legal limit in the United States of 0.3 mg/kg added Se on growth performance, serum and tissue Se concentrations, and serum antioxidant status of nursery pigs.

MATERIAL AND METHODS

General

The Kansas State University Institutional Animal Care and Use Committee approved the protocol used in this study. Experiments were conducted at a commercial research facility in north-central Ohio (Bucyrus, OH). Weaned pigs (approximately 21 d of age) from three sow farms entered the research facility over a 7-d period. Sows were fed diets containing 0.3 mg/kg of added Se with a minimum of 50% from yeastderived organic Se and the remainder from sodium selenite. At the time of weaning (day 7), a total of 3,888 pigs $(337 \times$ 1050, PIC, Hendersonville, TN; initially 6.0 ± 0.23 kg) were placed in the research barn with 27 pigs per pen $(2.2 \times 2.7 \text{ m})$. A total of 144 pens were used with 72 double-sided 5-hole stainless steel fence line feeders, each feeding 2 adjacent pens with a feeder serving as the experimental unit. For each feeder, 1 pen contained 27 gilts, and 1 pen contained 27 barrows. Each pen also contained a cup waterer to provide ad libitum access to feed and water. At the time of placement in the nursery facility, pens of pigs were weighed and allotted to one of three dietary treatments in a randomized complete block design with a blocking structure including sow farm origin, date of entry into the facility, and average pen bodyweight. There were 24 replicates (feeders) per dietary treatment.

There was an increase in clinical disease associated with *Streptococcus suis* above expected levels in the research barn during the phase 2 period (days 0 to 14) for approximately a week. Amoxicillin (250 mg/5 mL; NDC 0781-6041-55; Sandoz, Princeton, NJ) was administered through the water to all pens for 7 d.

Diets

Diets were fed in three phases, and all contained 0.3 mg/kg added Se (Table 1). The trace mineral premix used in the treatment diets did not contain Se (SEM Minerals, L.P., Quincy, IL). Phase 1 common diet was formulated with added Se from sodium selenite, manufactured and pelleted at Premier Feeds, LLC (Urbana, OH), and was fed to all pigs from weaning to approximately 6.0 kg body weight (BW) with a feed budget of 0.68 kg/pig. Three Se sources (sodium selenite [Phibro, Teaneck, NJ], Se yeast [Sel-Plex; Alltech, Lexington, KY], and OH-SeMet [Selisseo; Adisseo, Antony, France]) were used to formulate three experimental diets for phases 2 and 3, and were manufactured at the Hord Elevator (Bucyrus, OH). Phase 2 and 3 diets were fed in meal form with a feed budget of 11.6 and 14.5 kg/pig, respectively. The phase 1, 2, and 3 diets were fed approximately from days 7 to 0, days 0 to 14, and days 14 to 35, respectively. Prior to the initiation of the study, samples of corn, soybean meal, enzymatically treated soybean meal (HP300, Hamlet Protein Inc., Findlay, OH), and the three Se sources (sodium selenite, Se yeast, and OH-SeMet) were collected and analyzed for Se concentration at the Michigan State University Veterinary Diagnostic Laboratory (MSU VDL, East Lansing, MI). The analyzed Se concentrations were: corn (0.06 mg/kg), soybean meal (0.275 mg/kg), HP300 (0.321 mg/kg), sodium selenite (640.1 mg/ kg), Se yeast (648.8 mg/kg), and OH-SeMet (657.8 mg/kg). Selenium concentration of the basal diets without added Se was calculated based on the analyzed Se concentration (corn, soybean meal, HP300, and Se sources) and NRC (2012) Se concentration reference (dried whey and wheat). The basal Se concentration for phases 1, 2, and 3 were 0.141, 0.128, and 0.124 mg/kg respectively. All diets met the NRC (2012) vitamin and mineral requirement estimates.

The feed sampling procedures were adapted from Jones et al. (2018) for dietary minerals. The feed samples were collected from at least six feeders using a brass open-handle probe per treatment per phase, pooled, and subsampled for Se concentration (MSU VDL) and crude protein (Kansas State University Swine Laboratory, Manhattan, KS; Table 2). The feed Se assay was based on an Agilent Technologies Inc. (Santa Clara, CA) method using an inductively coupled plasma mass spectrometer (ICP/MS). The ICP/MS was tuned to yield a minimum of 7,500 cps sensitivity for 1 ppb yttrium (mass 89), less than 1.0% oxide level as determined by the 156/140 mass ratio, and less than 2.0% double charged ions as determined by the 70/140 mass ratio. Selenium concentration was calibrated using a 6-point linear curve of the analyteinternal standard response ratio. The standard curve was calibrated from 0.0002 to 2.5 mg/kg. Standards were from Inorganic Ventures (Christiansburg, VA). A National Institute of Standards and Technology (NIST; Gaithersburg, MD) typical diet standard was used as a control.

Data and Sample Collection

Feed additions to each feeder were made and recorded by an electronic feeding system (Dry Exact; Big Dutchman, Inc., Holland, MI). Pens of pigs were weighed by pen, and feed disappearance was calculated every 7 d until the study's

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Table 1. Diet composition, (as-fed basis)¹

Item	Phase 1	Phase 2	Phase 3
Ingredients, %			
Corn	35.00	60.64	62.90
Soybean meal	21.90	30.27	31.34
Dried whey	25.00	_	_
Soft red wheat	5.00	_	_
Enzymatically treated soybean meal ²	5.00	2.50	_
Corn oil	3.50	1.50	1.50
Limestone, ground	0.83	1.10	1.05
Monocalcium phosphate	0.90	1.10	0.95
Salt	0.35	0.50	0.35
l-Lys HCl	0.50	0.50	0.45
DL-Met	0.33	0.28	0.21
Thr source ³	0.26	0.33	0.28
l-Trp	0.08	0.06	0.05
L-Val	0.21	0.15	0.10
Phytase ⁴	0.05	0.10	0.10
Choline chloride	0.04	_	_
Vitamin premix ⁵	0.05	0.05	0.05
Sodium metabisulfite	0.50	0.50	0.50
Zinc oxide	0.38	0.26	_
Copper sulfate	_	0.03	0.03
Trace mineral premix ⁶	0.10	0.10	0.10
Se source ⁷	0.05	0.05	0.05
Total	100.00	100.00	100.00
Standardized ileal digestible amino acids, %			
Lys	1.42	1.38	1.30
Iso:Lys	55	56	57
Leu:Lys	105	113	117
Met:Lys	42	40	38
Met and Cys:Lys	62	61	59
Thr:Lys	66	65	65
Trp:Lys	21.8	20.7	20.2
Val:Lys	72	71	69
His:Lys	32	36	37
Net energy, kcal/kg	2,628	2,469	2,467
Crude protein, %	20.6	21.9	21.0
Ca, %	0.74	0.75	0.70
STTD P, ⁸ %	0.59	0.50	0.47

¹Corn, soybean meal, HP300, sodium selenite, Se yeast, and OH-SeMet were analyzed for Se concentration prior to the study at Michigan State University Veterinary Diagnostic Laboratory. Analyzed Se concentration: corn (0.06 mg/kg), soybean meal (0.275 mg/kg), HP300 (0.321 mg/kg), sodium selenite Veterinary Diagnostic Laboratory. Analyzed Se concentration: corn (0.06 mg/kg), soybean meal (0.275 mg/kg), HP300 (0.321 mg/kg), sodium selenite (640.1 mg/kg), Se yeast (648.8 mg/kg), and OH-SeMet (657.8 mg/kg). Basal Se concentration was calculated based on ingredient sample analysis prior to the study for corn, soybean meal, and HP300 at the MSU VDL, and Se concentration for dried whey and wheat was from NRC (2012). The basal Se concentration for phases 1, 2, and 3 were 0.141, 0.128, and 0.124 mg/kg respectively. All diets had 0.3 mg/kg added Se from either Se sources. ²HP300 (Hamlet Protein Inc.) is an enzyme-treated soybean meal. ³L-Threonine was used in phase 1 diet, and Thr pro biomass (CJ Bio America, Fort Dodge, IA) was used in phase 2 and 3 diets. ⁴Quantum Blue 5G (AB Vista, Plantation, FL) was used in phase 1 diet and provided 2,002 FTU per kg of diet with an expected STTD P release of 0.13%.

Quantum Blue 3G (AB vista, Frantation, FL) was used in phase 1 and and provided 2,002 FTU per kg of diet with an expected 3TTD P release of 0.15%. Quantum Blue 2G was used in phase 2 and 3 diets and provided 2,002 FTU per kg of diet with an expected STTD P release of 0.14%. ³Vitamin premix provided per kg of diet: 5,512 IU vitamin A; 1,653 IU vitamin D; 66 IU vitamin E; 3.3 mg vitamin K; 0.033 mg vitamin B12; 24.8 mg niacin; 27.6 mg pantothenic acid; 8.27 mg riboflavin; 0.22 mg biotin; 2.2 mg folic acid; and 0.99 mg pyridoxine. ⁶Trace mineral premix (SEM Minerals, L.P.) did not contain Se and provided per kg of diet: 161 mg Zn, 134 mg Fe, 42 mg Mn, 14 mg Cu, 0.66 mg Cl, and

9.0 µg Cr.

7Sodium selenite was used in phase 1 diet as the added Se source. Sodium selenite, Se yeast, and OH-SeMet were used as the added Se source in phase 2 and 3 diets as the three dietary treatments.

⁸STTD P, standardized total tract digestible phosphorus.

	Table 2. Analy	vzed dietarv	v Se and crude	protein concentration	s ¹
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	Sodium selenite	Se yeast	OH-SeMet
Phase 1 (common)			
Se, mg/kg	0.514	_	_
Crude protein, %	18.6	_	_
Phase 2			
Se, mg/kg	0.552	0.588	0.616
Crude protein, %	19.9	20.5	20.5
Phase 3			
Se, mg/kg	0.414	0.548	0.549
Crude protein, %	19.0	19.8	19.0

¹Sodium selenite was used in phase 1 diet as the added Se source. Sodium selenite, Se yeast, and OH-SeMet were used as the added Se source in phase 2 and 3 diets as the three dietary treatments. Feed samples were collected from at least six feeders per treatment per phase, pooled, and subsampled for Se concentration (MSU VDL) and crude protein (Kansas State University Swine Laboratory).

conclusion to calculate average daily gain (ADG), average daily feed intake (ADFI), average BW, and gain-to-feed ratio (G:F). Feed disappearance was measured using a volumetric regression equation, which estimates the quantity of feed remaining in the feeder subtracted from the quantity of feed added to the feeder.

One median-size barrow of each experimental unit was visually selected for serum collection on day 0. The same pig per experimental unit was marked with high visibility, numbered ear tag on day 0, and used in all subsequent serum collections (days 14 and 35 of the experiment). Serum samples from two pigs could not be collected due to the removal of the pig from the study; therefore, replacement barrows from the same pen were randomly selected and used for further sampling. Whole blood samples were allowed to clot for 30 min, centrifuged at $1,500 \times g$ for 15 min, and the resulting serum supernatants were divided into seven polypropylene tubes as aliquots and transferred and stored at -80 °C. Day 0 serum samples were collected when all pigs were still on phase 1 common diet formulated with sodium selenite. Serum Se concentration was analyzed on days 0, 14, and 35 samples at the MSU VDL. The serum Se assay used a similar method as the feed Se analysis but with in-house serum pools as the controls. Serum GSH-Px, total antioxidant capacity (T-AOC), and thiobarbituric acid reactive substances (TBARS) were evaluated on days 14 and 35 samples at the Kansas State University Swine Laboratory. For serum GSH-Px, assay kits were purchased from Cayman Chemical Company (Ann Arbor, MI; # 703102). For serum T-AOC, assay kits were purchased from Cell Biolabs, Inc. (OxiSelec, San Diego, CA; # STA-360). For TBARS, the assay was a modification of the methods of Yagi (1998) and Aguilar Diaz De Leon and Borges (2020). Serum GSH-Px, T-AOC, and TBAR samples were run in triplicate in 96-well microplates with an intraassay CV of \leq 5.0%. The malondialdehyde bis (MDA bis) standard curve was prepared freshly for each 96-well microtiter plate with a range of 1.56 to 100 µM MDA. A total of 100 µL of each standard or undiluted serum sample was added to each test tube, and then 200 µL of 10% TCA solution was added for MDA extraction. The solution was mixed with 1.0 mL of TBA/sodium acetate and incubated in a boiling water bath (95 °C)

for 60 min. After incubation, test tubes were placed in an ice bath for 15 min and then centrifuged at $1500 \times g$ for 10 min at 4 °C. Immediately after centrifugation, 150 µL of supernatant was aliquoted from each tube and placed into a separate well of a 96-well microtiter plate. The absorbance was read at 532 nm with a spectrophotometer. The average absorbance reading of the blank samples was subtracted from all other absorbance readings. A standard curve was created by plotting the blank-subtracted absorbance readings and the known concentrations of each standard. Sample data points were then fitted using the equation of the linear regression line obtained from the standard curve to calculate sample concentrations.

The same 72 barrows used for serum collection were euthanized with a penetrative captive bolt pistol on day 35 of the experiment for the collection of muscle and liver tissue by a licensed veterinarian for consistency of sample collection. Muscle samples were collected from the loin at the 10th rib. Liver samples were collected from the right median lobe adjacent to the gallbladder. Following collection, fresh tissue samples were stored on ice and transported to the MSU VDL for analysis of tissue Se concentration. The tissue Se analysis used a similar method as the feed Se analysis, but with NIST bovine liver and muscle standards as controls.

Statistical Analysis

Data were analyzed as a randomized complete block design for one-way ANOVA using the lmer function from the lme4 package for growth performance, percentage of injectable treatments, Se concentration, and antioxidant status, and the glmer function (binomial distribution) from the lme4 package for the percentage of removal and mortality in R program (R core team, 2022; Vienna, Austria). Feeder (two pens of pigs) was considered the experimental unit. Initial pen average BW, sow farm origin, and date of entry into the facility were the blocking factors. Treatment was used as the fixed effect. For days 0 to 35 growth performance, days 7 to 0 ADFI was used as a covariate because of the tendency of a difference between treatments. There was no evidence of treatment × sex interaction (P > 0.10) in BW throughout this trial. For serum selenium concentration, day 0 selenium concentration was used as a covariate for days 14 and 35 serum selenium concentrations, which were analyzed as repeated measures. For the serum GSH-Px, T-AOC, and TBARS assay, a microtiter plate was used as a random effect, and serum samples were balanced for the block when placed on microplates. For the serum GSH-Px assay, T-AOC, and TBARS were analyzed in triplicate, duplicate, and triplicate, respectively. Data were analyzed by accounting for subsampling and repeated measures over time. Tukey adjustment was used for multiple comparisons. All results were considered significant at $P \leq 0.05$ and marginally significant at $0.05 < P \le 0.10$.

RESULTS

Growth Performance

From days 7 to 0 (pre-treatment period), there was no evidence of difference (P > 0.05) in day 7 BW, day 0 BW, ADG, and G:F between treatment groups (Table 3). However, despite the common diet, there was marginally significant evidence of a difference (P = 0.097) in ADFI between treatments from days 7 to 0, with the OH-SeMet having a numerically

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Table 3. Evaluation of Se source on nursery pig growth performance ^{1,2}

	Sodium selenite	Se yeast	OH-SeMet	Р
Days 7 to 0 (pre-treatment) ³				
Day 7 BW, kg	6.0 ± 0.05	6.0 ± 0.05	6.0 ± 0.05	0.979
Day 0 BW, kg	6.6 ± 0.08	6.6 ± 0.08	6.6 ± 0.08	0.485
ADG, g	97 ± 6.2	93 ± 6.2	93 ± 6.2	0.367
ADFI, g	107 ± 3.6	103 ± 3.6	102 ± 3.6	0.097
G:F, g/kg	884 ± 33.8	897 ± 33.8	898 ± 33.8	0.815
Days 0 to 14				
Day 14 BW, kg	12.7 ± 0.07	12.7 ± 0.07	12.6 ± 0.07	0.121
ADG, g	435 ± 3.0^{a}	432 ± 3.0^{a}	422 ± 3.0^{b}	0.003
ADFI, g	490 ± 3.2^{a}	490 ± 3.1^{a}	479 ± 3.2^{b}	0.015
G:F, g/kg	888 ± 4.7	883 ± 4.6	884 ± 4.6	0.623
Days 14 to 35				
Day 35 BW, kg	26.9 ± 0.11	26.7 ± 0.11	26.7 ± 0.11	0.312
ADG, g	672 ± 4.4	664 ± 4.3	666 ± 4.4	0.328
ADFI, g	956 ± 7.8	940 ± 7.7	945 ± 7.8	0.176
G:F, g/kg	703 ± 3.0	707 ± 3.0	705 ± 3.0	0.352
Days 0 to 35				
ADG, g	577 ± 3.0^{x}	571 ± 2.9^{xy}	$567 \pm 2.9^{\text{y}}$	0.066
ADFI, g	769 ± 4.8	759 ± 4.8	756 ± 4.8	0.100
G:F, g/kg	750 ± 2.1	752 ± 2.1	750 ± 2.1	0.560
Days 7 to 35				
ADG, g	494 ± 2.5	490 ± 2.5	487 ± 2.5	0.110
ADFI, g	656 ± 3.9^{x}	647 ± 3.9^{xy}	$645 \pm 3.9^{\text{y}}$	0.090
G:F, g/kg	754 ± 2.2	757 ± 2.2	755 ± 2.2	0.450
Treatments, % ⁴	4.6 ± 0.78	3.9 ± 0.78	5.8 ± 0.78	0.196
Removal, % ⁵	2.65 ± 0.495	2.58 ± 0.487	2.94 ± 0.528	0.824
Mortality, % ⁶	0.39 ± 0.172	0.39 ± 0.172	0.46 ± 0.189	0.939
Removal with mortality, % ⁷	3.18 ± 0.509	3.11 ± 0.503	3.56 ± 0.540	0.781

¹A total of 3,888 pigs (initially 6.0 ± 0.23 kg) were used with 54 pigs per replicate and 24 replicates per treatment. All pigs were fed 0.68 kg per pig of phase 1 common starter pellet that contained 0.3 mg/kg of added Se from sodium selenite for approximately 7 d. Phase 2 and 3 treatment diets were fed after pigs finished the phase 1 feed budget. All treatment diets provided 0.3 mg/kg added Se from sodium selenite, Se yeast, or hydroxy-selenomethionine. ²BW, body weight; ADG, average daily gain; ADFI, average daily feed intake; G:F, feed efficiency.

³Days 7 to 0 ADFI was used as a covariate for growth performance in subsequent periods and overall growth performance.

⁴The number of injectable treatments given when pigs were on treatment diets divided by the number of pigs on day 0. ⁵The number of pigs removed based on body condition, lameness, and health status when they were on treatment diets divided by the number of pigs on day 0.

⁶The number of pigs found dead when they were on treatment diets divided by the number of pigs on day 0.

The number of pigs removed or dead when they were on treatment diets divided by the number of pigs on day 0.

^{a,b}Means within a row with different superscripts differ ($P \le 0.05$).

^{x,y}Means within a row with different superscripts differ (0.05 < $P \le 0.10$).

lower ADFI compared to the other treatment groups; therefore, days 7 to 0 ADFI was used as a covariate for growth performance in subsequent periods and overall growth performance. From days 0 to 14, pigs fed OH-SeMet had decreased (P < 0.05) ADFI and ADG compared to pigs fed the other two treatments. There was no evidence of difference (P > 0.05) in G:F between treatments. From days 14 to 35, there was no evidence of difference (P > 0.05) in any growth criteria between the three treatments. From days 0 to 35, pigs fed OH-SeMet tended to have decreased ADG (P < 0.10) compared to pigs fed sodium selenite. From days 7 to 35 (overall period), pigs fed OH-SeMet tended to have decreased ADFI (P < 0.10) compared to pigs fed sodium selenite. However, for the same period, there was no evidence of difference (P > 0.05) in ADG, G:F, day 35 BW, percentage of injections, removal, and mortality between treatments.

Serum and Tissue Se Concentration

There was a tendency (P = 0.072) of a source × day interaction for serum Se (Table 4). Pigs fed OH-SeMet had a greater increase in serum Se from days 14 to 35 than pigs fed sodium selenite or selenium yeast. On day 14, pigs fed OH-SeMet had increased (P < 0.05) serum selenium concentration compared to pigs fed selenium yeast. On day 35, despite a lower ADFI, pigs fed OH-SeMet had increased (P < 0.05) serum, muscle, and liver selenium concentration compared to all other treatments.

Antioxidant Status

There was a marginally significant increase in GSH-Px (P =0.074) over time; however, no source × day interaction or main effect of the source was observed (P > 0.10; Table 4). A source \times day interaction (P = 0.027) was found on serum

Table 4. Evaluation of selenium source on nursery pig serum and tissue selenium concentrations and serum antioxidant status¹

	Sodium selenite	Selenium yeast	OH-SeMet	Р		
				Source × day	Source	Day
Serum selenium, ng/mL ²				0.072	0.002	<0.0001
Day 14	131 ± 3.7^{cd}	122 ± 3.7^{d}	$139 \pm 3.7^{\circ}$			
Day 35	184 ± 3.7^{b}	184 ± 3.7^{b}	205 ± 3.7^{a}			
Liver selenium, µg/g				_	< 0.0001	_
Day 35	$1.97 \pm 0.049^{\text{b}}$	1.99 ± 0.049^{b}	2.45 ± 0.049^{a}			
Muscle selenium, µg/g				_	< 0.0001	_
Day 35	0.81 ± 0.024^{b}	0.87 ± 0.024^{b}	1.42 ± 0.024^{a}			
Serum GSH-Px, nmol/min/mL ³				0.710	0.971	0.074
Day 14	869 ± 129	888 ± 129	881 ± 129			
Day 35	$1,268 \pm 128$	1,210 ± 128	1,239 ± 128			
Serum T-AOC, CRE ⁴				0.027	0.306	0.089
Day 14	359 ± 22.9	389 ± 22.9	385 ± 22.9			
Day 35	440 ± 22.9	470 ± 22.9	417 ± 22.9			
TBARS, µM MDA ⁵				0.262	0.177	0.781
Day 14	5.92 ± 0.97	5.25 ± 0.97	6.34 ± 0.97			
Day 35	5.52 ± 0.97	4.28 ± 0.97	4.58 ± 0.97			

¹A total of 3,888 pigs (initially 6.0 ± 0.23 kg) were used with 54 pigs per replicate and 24 replicates per treatment. Serum and tissue samples were collected from the same one barrow per replication throughout this trial (24 pigs per treatment). All pigs were fed 0.68 kg per pig of phase 1 common starter pellet that contained 0.3 mg/kg of added selenium from sodium selenite for approximately 7 d. Phase 2 and 3 treatment diets were fed after pigs finished the phase 1 feed budget. All treatment diets provided 0.3 mg/kg added selenium. Selenium concentrations in serum, liver, and muscle were analyzed at MSU VDL.

²Day 0 serum samples were collected when all pigs were fed the phase 1 common diet and averaged 125 ng/mL across all treatments. Day 0 serum selenium concentration was used as a covariate for statistical analysis of days 14 and 35 serum selenium concentration as repeated measurement. ³GSH-Px, glutathione peroxidase. One unit is defined as the amount of enzyme that causes the oxidation of 1.0 nmol of NADPH to NADP⁺ per minute at

25 °C.

⁴T-AOC, total antioxidant capacity; CRE, µM copper reducing equivalents.

⁵Thiobarbituric acid reactive substances. µM of MDA (malondialdehyde) equivalent.

^{a-d}Means within a response with different superscripts differ ($P \le 0.05$).

T-AOC. Although no significant means separation was present, pigs fed OH-SeMet had a smaller numerical increase in serum T-AOC from days 14 to 35 compared to pigs fed sodium selenite or selenium yeast. Additionally, there was a marginally significant increase in T-AOC over time (P = 0.089). There was no evidence of a source × day, source, or day effect for TBARS (P > 0.10).

DISCUSSION

In the United States, the maximum legal limit for added Se is 0.3 mg/kg (FDA, 2021). This concentration is commonly used in nursery pig diets as reported by two industry surveys on vitamins and trace minerals (Flohr et al., 2016; Faccin et al., 2023). At this level, the added Se (0.3 mg/kg) provides the required Se which for nursery pigs is 0.3 mg/kg for 5- to 11-kg pigs and 0.25 mg/kg for 11- to 25-kg pigs, as stated by the NRC (2012). The requirement for selenium can be affected by dietary vitamin E as both nutrients are involved with antioxidant roles (NRC, 2012). Moreover, high levels of some nutrients, such as Cu, Zn, and Fe, in diets have the potential to destroy dietary vitamin E (Dove and Ewan, 1990), which indirectly affects the requirement of Se to maintain proper antioxidant function. Therefore, the dietary inclusion levels of Se used in the current experiment were reflective of the levels currently used in swine diets of 0.3 mg/kg of added Se with the focus being determination of response to different Se sources. The calculated vitamin E (66 IU/kg), Cu (14, 77, and 77 mg/kg for the three phases, respectively), Zn (2,861,

2,033, and 161 mg/kg for the three phases, respectively), and Fe (134 mg/kg) levels were all formulated within the ranges that were commonly used in the industry (Flohr et al., 2016; Faccin et al., 2023).

Effects of Se Sources on Growth Performance

Consistent with a limited number of previous trials with nursery and growing pigs, different Se sources did not affect growth performance. Chao et al. (2019) found no evidence of difference (P > 0.05) in growth performance between nursery pigs fed 0.3 mg/kg added Se from sodium selenite and OH-SeMet. Cao et al. (2014) reported no evidence of difference (P > 0.05) in growth performance between nursery pigs fed different levels of DL-SeMet (0.1 to 0.7 mg/kg) or between pigs fed 0.3 mg/kg Se from DL-SeMet and sodium selenite. Jlali et al. (2014) also reported no evidence of difference (P > 0.05) in growth performance between growing pigs fed either 0.1 or 0.3 mg/kg added Se from sodium selenite, Se yeast, or OH-SeMet. Moreover, Mahan et al. (1999) found no evidence of difference (P > 0.05) in growth performance between growing-finishing pigs fed 0.3 mg/kg of sodium selenite or Se yeast. However, the present study observed a reduction in early phase ADFI in nursery pigs fed OH-SeMet compared to pigs fed sodium selenite or Se yeast. The feed, serum, and tissue Se concentration analysis confirmed that OH-SeMet was added to the diets; therefore, we concluded that the reduction in ADFI was not caused by Se deficiency. In addition, factors that might reduce ADFI were closely examined, and no bias was found.

Effects of Se Sources on Serum and Tissue Se Concentration

For normal pigs, Ullrey (1987) and Mahan (1991) suggested that the serum Se concentration ranges from 80 to 150 ng/ mL, while Blood and Radostits (1989) considered above 120 ng/mL serum as normal levels. These suggest that all pigs from the three Se sources had serum Se concentration within normal range without deficiency. The inorganic and organic forms of Se are both used as feed mineral additives along with Se provided from other feed ingredients to meet the Se requirement of pigs; however, organic Se sources have been shown to have greater bioavailability than inorganic sources (Shini et al., 2015). Other studies comparing pigs fed OH-SeMet to sodium selenite observed similar results to our study. Chao et al. (2019) reported increased serum, liver, kidney, and muscle Se concentration in nursery pigs fed OH-SeMet compared to sodium selenite. Mahan et al. (1999) and found that growing-finishing pigs fed Se yeast had greater (Se source \times Se level, P < 0.01) increases in tissue Se as dietary Se level increased from 0.05 to 0.3 mg/kg compared to sodium selenite. Similarly, Mahan et al. (2014) also found that growing-finishing pigs fed Se yeast had greater serum Se and tissue Se (loin, liver, and heart) compared to sodium selenite. Growing pigs fed 0.3 mg/kg of added Se from OH-SeMet had the highest (P < 0.05) plasma, liver, and muscle Se concentration, followed by Se from Se yeast, and sodium selenite had the lowest concentration (Jlali et al., 2014). A similar result was reported in another study where growing pigs fed L-SeMet had the highest (P < 0.05) tissue Se concentration, followed by Se yeast and then sodium selenite (Falk et al., 2018). In a meta-analysis, Zhou et al. (2021) observed that sows fed organic Se sources had 29% greater serum Se concentration than inorganic Se sources. However, we observed no evidence of difference in pigs fed Se yeast compared to sodium selenite. Cao et al. (2014) also found no evidence of difference in plasma Se concentration between nursery pigs fed 0.3 mg/kg of Se from sodium selenite and DL-SeMet. By comparing the results herein with these previous studies, we can conclude that OH-SeMet may have greater bioavailability, followed by Se yeast, and sodium selenite. However, to determine the true bioavailability of OH-SeMet, slope ratio assays are needed. To understand the differences in bioavailability between Se sources, differences in Se absorption across the intestine wall should be considered. Inorganic Se, such as sodium selenite, is absorbed passively across the intestinal wall as ions, while selenomethionine is absorbed actively with amino acid or peptide transporters on the enterocytes (Mahima et al., 2012; Shini et al., 2015).

Moreover, the chemical properties of inorganic Se ions may form insoluble complexes with other feed components or interact with phytate in the digesta, which reduces the absorption of Se across the intestine wall (Mahima et al., 2012). On the other hand, organic Se, as a result of Se being bound to AAs, forms stable complexes that are less prone to interact with other feed components (Mahima et al., 2012). After absorption, both inorganic and organic Se are converted to selenocysteine or selenoproteins in the enterocytes and then transported via the portal vein to the liver, where selenocysteine is converted into selenoprotein P or extracellular GSH-Px for peripheral tissue, such as kidney and muscle (Shini et al., 2015). Consequentially, these factors result in the greater absorption and bioavailability of organic Se sources, as observed in the increased serum and tissue Se concentrations.

Effects of Se Sources on Antioxidant Status and Health

Because organic Se source elevates the Se status of animals. we were interested in whether this increase could translate to an elevated antioxidant status. The results of our study and the previous studies showed that the effects of Se sources on antioxidant status have some inconsistencies, which might be caused by the differences in the concentrations of Se in the basal diets, the oxidative stress caused by the environment, and/or the stage of production. We found no evidence of difference between sources of Se in the antioxidant status of nursery pigs. Mahan et al. (1999, 2014) observed no evidence of difference (P > 0.05) in serum GSH-Px between growing-finishing pigs fed 0.3 mg/kg of sodium selenite or Se yeast. Cao et al. (2014) reported no evidence of difference (P > 0.05) in serum MDA, liver GSH-Px, liver T-AOC, liver MDA, muscle T-AOC, and muscle MDA between nursery pigs fed 0.3 mg/kg added Se from sodium selenite and DL-SeMet: however, pigs fed DL-SeMet had increased (P < 0.05) serum GSH-Px, serum T-AOC, muscle GSH-Px compared to sodium selenite. Chao et al. (2019) found no evidence of difference (P > 0.05) in serum T-AOC, serum GSH-Px, liver GSH-Px, and liver MDA between nursery pigs fed 0.3 mg/kg added Se from sodium selenite and OH-SeMet; however, pigs fed OH-SeMet had reduced (P < 0.05) serum MDA and increased (P < 0.05) liver T-AOC compared to sodium selenite. These experiments suggest that there were some differences in antioxidant status between feeding 0.3 mg/kg of inorganic and organic Se sources to pigs in their growing stage, but the growth performance result showed no difference. However, these experiments were conducted in research facilities that may have lower oxidative stress from the environment and pathogens compared to commercial facilities. Therefore, the increased Se status did not consistently benefit the antioxidant system in these experiments. Moreover, pigs have lower feed intake in commercial facilities compared to research facilities; thus, the magnitude of the bioavailability of Se sources may play a more important role in commercial facilities. In a meta-analysis, the authors observed that sows fed organic Se sources had 6.4% higher GSH-Px activity than inorganic Se sources (Zhou et al., 2021). This consistent improvement may be because sows are under constant oxidative stress during gestation and lactation; therefore, the increase in tissue Se level from organic Se may benefit the antioxidant status and performance of the animals (Zhou et al., 2021). Selenium supplementation with OH-SeMet during the gestation period improved (P < 0.05) litter weight gain, antioxidant status, and intestinal antioxidant capacity, and reduced (P < 0.05) birth interval and inflammation level compared to sodium selenite (Mou et al., 2020a, 2020b, 2021). By implication, organic Se may also be beneficial in growing pigs under greater environmental or pathogenic oxidative stress.

During disease challenges, nursery pigs experience higher oxidative stress (Hao et al., 2021) and have a reduction in feed intake, growth rate, and serum and tissue Se status (Campbell et al., 2013; Sun et al., 2017). A Se source with higher bioavailability during this period may have the potential to benefit the pigs more by providing more absorbed Se or more mobilized Se from the higher Se reservoir in serum or tissue. Even though the Se was not directly fed to the piglets, Lipopolysaccharide (LPS)-challenged weaned pigs from sows fed OH-SeMet had improved antioxidant status and reduced inflammation levels compared to piglets from sows fed sodium selenite (Mou et al., 2020a, 2021). Doan et al. (2020) challenged nursery pigs with diquat and found no evidence of difference (P > 0.05) in post-challenge ADFI and BW between challenged pigs fed Se yeast and pigs without challenge. However, they observed improved (P < 0.05) antioxidant status in challenged pigs fed Se yeast compared to challenged nursery pigs fed sodium selenite. On the contrary, even though our pigs were reared under commercial conditions, we observed no difference in antioxidant status between Se sources when pigs experienced S. suis challenge. However, the challenge was not planned or controlled, and pigs may have recovered from the pathogen challenge before the blood samples were taken on day 14 of the trial. Therefore, more research is needed to confirm the effect of organic Se sources on pigs under oxidative stress or challenges.

In conclusion, organic Se sources, especially OH-SeMet, may be able to provide greater Se bioavailability as indicated by the increased serum and tissue Se concentration. However, the improved Se reservoir did not affect the pigs' growth performance, health status, or antioxidant status. This indicates that under the conditions of this experiment, both inorganic and organic Se sources added at 0.3 mg/kg can provide adequate Se to meet the pigs' requirement. A higher Se reservoir in tissue may benefit health and antioxidant status when pigs are under high oxidative stress. However, more studies are needed to confirm the effect of the organic Se source used in these scenarios.

ACKNOWLEDGMENTS

Contribution no. 23-019-J from the Kansas Agric. Exp. Stn., Manhattan, KS 66506-0210, USA. Appreciation is expressed to Hord Family Farms (Bucyrus, OH) for providing the animals and research facility and to Dwight Shawk, Phil Hord, and research staff for technical support.

Conflict of interest: The authors declare no conflict of interest.

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