



Effects of water quality on growth performance and health of nursery pigs

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ABSTRACT

An experiment was conducted to determine the effects of providing drinking water of differing qualities on growth performance and health of nursery pigs. Weaning pigs ($n = 450$; 150 pigs/group; 10 pigs/pen) were assigned randomly to one of three experimental groups consisting of three water sources of varying qualities: 1) Water source A containing 1,410 ppm hardness (CaCO_3 equivalent), 1,120 ppm sulfates, and 1,500 ppm total dissolved solids (TDS); 2) Water source B containing 909 ppm hardness (CaCO_3 equivalent), 617 ppm sulfates, and 1,050 ppm TDS; and 3) Water source C containing 235 ppm hardness (CaCO_3 equivalent), 2 ppm sulfates, and 348 ppm TDS. Pigs were provided ad libitum access to their respective water sources for the duration of the study which began at weaning (21 d of age) and ended 40 d later (61 d of age). Individual pig weights were recorded weekly along with feed intake on a pen basis. Occurrences of morbidity and mortality were recorded daily. Subjective fecal scores were assigned on a pen basis and blood samples were used to evaluate blood chemistry, cytokine concentrations, and phagocytic activity. A differential sugar absorption test was used to assess intestinal permeability. Fecal grab samples were used to establish diet digestibility, and drinking behavior was video-recorded to assess pigs' acceptance of water sources provided. The statistical model considered fixed effects of water source, room, and their interaction with the random effect of pen. A repeated measures analysis was conducted to determine the effects of water quality over time. There were no differences ($P > 0.440$) among water sources in average daily gain (A, 0.46 kg/d; B, 0.46 kg/d; C, 0.47 kg/d) or average daily feed intake (A, 0.68 kg/d; B, 0.69 kg/d; C, 0.71 kg/d). Overall mortality of pigs was 0.44% and did not differ across the three water sources. There were no differences in apparent total tract digestibility of the diet, intestinal permeability, immune parameters, or blood chemistry attributable to quality of water consumed by pigs. Pigs did not show an aversion to the water sources provided, because total time pigs spent at the drinker did not differ ($P > 0.750$) among water sources on days 1 through 3 of the experiment. These data indicate that the water sources of differing quality studied did not affect growth performance or health of nursery pigs.

Key words: drinking behavior, growth performance, health, nursery pigs, water quality

INTRODUCTION

The role of water quality in pig health and growth performance is poorly understood. Researchers have expended more effort to determine optimal water availability than they have on determining optimal quality of water for pigs. In recent years, some pork producers have questioned if the quality of water in their barns contributes to observed increases in the percentage of pigs with suboptimal performance relative to their contemporaries. Unfortunately, little research has been published recently that evaluated the effects of water quality on nursery pig growth performance and health. The research studies that have been published have focused on determining the effects of one water characteristic in isolation from other potentially important characteristics. [McLeese et al. \(1992\)](#) studied effects of total dissolved solids (TDS) content in water provided to pigs post-weaning and found that as levels of TDS increased, nursery pig growth rate declined marginally, and frequency and severity of diarrhea increased. Similarly, other researchers ([Paterson et al., 1979](#); [Anderson et al., 1994](#); [Patience et al., 2004](#)) reported that increasing concentrations of sulfates in water increased the presence

and severity of diarrhea with no differences in pig growth performance. Although these studies are informative, they ignore potential interactions among different water characteristics that might influence pig performance and health.

Current recommendations for acceptable quality water to be fed to livestock were established in the 1970s and 1980s ([NRC, 1974](#); [CCME, 1987](#)) and have remained unchanged since then. In response to pork producers' concerns about the role of water quality in nursery pig performance, we wondered if a range of water qualities would affect nursery pig performance and health. Therefore, the objectives of this study were to evaluate the effects of varying drinking water quality on growth performance, diet nutrient digestibility, health, immune function, blood chemistry, and drinking behavior of pigs post-weaning. We hypothesized that varying water qualities would influence growth performance and health status of pigs.

MATERIALS AND METHODS

This experiment was conducted in the research nursery barn at the University of Minnesota's West Central Research and

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Outreach Center in Morris, MN. The experimental protocol was reviewed and approved by the University of Minnesota Institutional Animal Care and Use Committee (IACUC # 1907-37251A). The study was conducted from September 11 to October 21, 2019.

Animals, Housing, and Experimental Groups

Weanling pigs [$n = 450$; 19 ± 2 d of age; initial BW = 6.24 ± 0.15 kg; (Large White \times Landrace) \times Duroc] were sourced from a single, commercial sow farm (Christensen Farms, Sleepy Eye, MN) that was negative for Porcine Reproductive and Respiratory Syndrome Virus (PRRSV) and *Mycoplasma hyopneumoniae*. Pigs were assigned at random to one of three experimental groups upon arrival at the research barn. Experimental groups were assigned randomly to pairs of adjacent pens, and pairs of pens were assigned randomly to one of three water sources.

Water sources were as follows: 1) Water source A containing 1,410 ppm hardness (CaCO₃ equivalent), 1,120 ppm sulfates, and 1,500 ppm TDS; 2) Water source B containing 909 ppm hardness (CaCO₃ equivalent), 617 ppm sulfates, and 1,050 ppm TDS; and 3) Water source C containing 235 ppm hardness (CaCO₃ equivalent), 2 ppm sulfates, and 348 ppm TDS (Table 1). Characteristics of all three water sources were within acceptable ranges as described by CCME (1987) except for sulfate concentration in Water source A which was 120 ppm higher than the maximum limit of 1,000 ppm. Nonetheless, Water sources A and B were presumed to be of lesser quality because they contained greater concentrations of hardness, sulfates, TDS, calcium, and iron than Water source C, which was selected to represent good quality water. All three water sources were collected near the head of wells located on commercial swine farms in Minnesota. Commercial farms were selected after analyzing water characteristics of 15 farms in Minnesota and were selected to represent the range of water quality being fed to nursery pigs in 2019. Pigs remained on their assigned water source for 40 d with ad libitum access. All pigs had ad libitum access to the same high quality, commercial, four-phase nursery feeding program across all water sources (Table 2). The phase one diet was a proprietary, pelleted diet provided by VitaPlus Corp. (Madison, WI) and included a combination of tiamulin (Denagard) and CTC (chlortetracycline) at 40 and 440 ppm in the final diet, respectively. Duration of dietary feed phases 1, 2, 3, and 4 was 4, 10, 14, and 12 d, respectively. All diets met or exceeded nutrient concentrations recommended by NRC (2012) for weaned pigs.

Pigs were housed in pens of 10 pigs with floor space allowance of 0.28 m² per pig for the duration of the experiment. Two mirror-image rooms were used, each room contained 32 pens. Each pen was equipped with one stainless steel feeder with five feeding spaces and one water cup (Drink-o-Mat, Vittetoe Inc; Keota, IA) with fully slotted, plastic flooring over a deep manure pit. For this experiment, 45 pens (15 pens per experimental group) were utilized (22 pens in Room 1 and 23 pens in Room 2). Each room used an independent ventilation controller for heaters and ventilation fans.

Water Storage and Quality Management

Water sources were transported to the research barn in an insulated milk tanker truck. Water was stored in separate

Table 1. Initial analysis of water provided to nursery pigs

Analyte	Water source ¹		
	A	B	C
Arsenic, ppm	< 0.10	< 0.10	< 0.10
Bicarbonate (as CaCO ₃), ppm	397	375	270
Boron, ppm	0.25	0.24	0.13
Cadmium, ppm	< 0.01	< 0.01	< 0.01
Calcium, ppm	284	214	58.7
Carbonate (as CaCO ₃), ppm	< 1.0	< 1.0	< 1.0
Chloride, ppm	2	0	2
Chromium, ppm	< 0.01	< 0.01	< 0.01
Conductivity, mmhos/cm	2.31	1.62	0.536
Copper, ppm	n.d. ²	0.02	0.02
Fecal coliforms, CFU/100 mL	< 2	< 2	< 2
Fluoride, ppm	0.2	0.2	0.4
Hardness, ppm hardness (CaCO ₃ equivalent)	1,410	909	235
Iron, ppm	5.43	5.22	1.33
Lead, ppm	< 0.05	< 0.05	< 0.05
Magnesium, ppm	171	90.9	21.4
Manganese, ppm	0.048	0.117	0.045
Mercury, ppm	< 0.01	< 0.01	< 0.01
Nickel, ppm	< 0.01	< 0.01	< 0.01
Nitrate, ppm	n.d. ²	n.d.	n.d.
Nitrite (NO ₂), ppm	< 0.02	< 0.02	< 0.02
pH	8	8	7.5
Phosphorus, ppm	0.12	0.15	0.1
Potassium, ppm	5.34	6.33	2.67
SAR ³	0.7	0.5	0.8
Sodium, ppm	64	37.4	29.4
Sulfate, ppm	1,120	617	2
TDS ⁴ , ppm	1,500	1,050	348
Zinc, ppm	0.03	< 0.01	0.05

¹A (1,410 ppm hardness (CaCO₃ equivalent), 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness (CaCO₃ equivalent), 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness (CaCO₃ equivalent), 2 ppm sulfate, 348 ppm TDS).

²n.d., not detected.

³SAR, sodium absorption ratio.

⁴TDS, total dissolved solids, determined directly.

water bladders (9,464 L capacity; Potable Pillow Bladder Tank, Aire Industrial; Meridian, ID) outside of the barn on a shaded, level platform. Each water bladder was connected to a new water distribution system that included new piping, pump, and pressure tank. Pressure tanks for every bladder were set to maintain a flow of about 0.5 liter/min to drinker cups within pens. All water sources were delivered to pairs of adjacent pens. Previously existing standpipes (stainless steel pipe connecting new pipes to water cups) and water nipples placed in drinker cups were used during the study. New water drinking nipples were installed and calibrated to deliver a uniform flow rate (0.5 liter/min).

Water flow rates at the drinker were measured at the pen level weekly by collecting water from each water drinker for 30 s. Water flow rates were measured in every pen through week 4. During weeks 5 and 6, water flow rates in every other pen were measured. To monitor water quality, water samples were collected from pressure tanks 1 d after initial arrival of water, 1 d before the second delivery of water, and on the last day of the experiment. Water samples were analyzed at

Table 2. Ingredient and nutrient composition of nursery diets (as-fed basis)

Ingredient, %	Phase 2 ¹	Phase 3 ²	Phase 4 ³
Corn	47.14	54.00	64.82
Soybean meal	12.50	25.00	30.50
Specialty proteins ⁴	30.37	16.09	–
Titanium dioxide pre-blend ⁵	4.00	–	–
Soy oil	1.25	1.00	1.00
Aureomycin 50® ⁶	0.10	–	–
Dicalcium phosphate 21% P	0.51	0.69	0.91
Calcium carbonate	0.47	0.49	0.90
Salt, White	0.46	0.40	0.58
L-Lysine 98.5%	0.39	0.47	0.48
Zinc oxide	0.32	0.32	–
Vitamin trace mineral premix	0.25	0.27	0.17
Other ⁷	2.24	1.28	0.64
Total	100	100	100
Calculated nutrient composition			
ME, kcal/kg	3,372	3,367	3,364
Crude protein, %	22.0	21.7	21.3
Crude fat, %	4.26	4.05	3.92
Calcium, %	0.68	0.71	0.63
STTD Phosphorus, % ⁸	0.57	0.53	0.46
SID Lys, % ⁹	1.42	1.39	1.27
SID Trp, % ⁹	0.26	0.26	0.23
SID Met + Cys, % ⁹	0.80	0.81	0.72
SID Thr, % ⁹	0.91	0.86	0.77

¹All ingredients minus corn, soybean meal, soy oil, and pre-blend were provided by Nursery Base 700 (Team Nutrition, Inc., Cyrus, MN). Phase 2 diet was fed for 10 d.

²All ingredients minus corn, soybean meal, and soy oil provided by TNI 400 Nursery Base (Team Nutrition, Inc., Cyrus, MN). Phase 3 diet was fed for 14 d.

³All ingredients minus corn, soybean meal, and soy oil provided by TNI 25-80 NG Premix (Team Nutrition, Inc., Cyrus, MN). Phase 4 diet was fed for 12 d.

⁴Specialty proteins (mix of specialty animal and plant proteins).

⁵Composed of 46.5% soybean meal (87.5%) and titanium dioxide (12.5%).

⁶Aureomycin 50 (Zoetis, Parsippany-Troy Hills, NJ) added to Phase 2 diet to control *Streptococcus suis*.

⁷Other (mixture of carbohydrate sources, synthetic amino acids, flavors, preservatives, and yeast products).

⁸Standard total tract digestible phosphorus.

⁹SID, standard ileal digestible.

Midwest Laboratories (Omaha, NE) for 29 different analytes as presented in Table 1. Ambient temperature near each water bladder was recorded every 10 min with HOBO temperature recorders (HOBO MX2203 Underwater Temp Recorder, Onset Products; Cape Cod, Massachusetts) for the duration of the experiment.

Pig Growth Performance and Health Measurements

Pigs were identified by individual ear tags. Initial body weight and sex of each pig were recorded. All feed additions to feeders were weighed and recorded. Individual pig weights and pen feed disappearance were measured each week to determine average daily gain (ADG), average daily feed intake (ADFI), and the gain:feed (G:F). Pigs were observed multiple times daily to identify any adverse health conditions.

Records of morbidity included pig identification number, sex, pen number, date, clinical signs, if any treatment was administered, drug name, withdrawal period, and the treatment outcome. Records of mortality included pig identification number, sex, bodyweight, pen number, date, and suspected cause of death. To assess occurrence of diarrhea, fecal grab samples were collected from two randomly selected pigs in each pen daily during the first 7 d of the experiment. Fecal samples were pooled within pen for determination of fecal moisture. Fecal grab samples were collected once daily in the morning, placed in Ziploc bags, and stored at –20 °C. To determine moisture content, samples were dried in a forced draft oven at 60 °C and weighed twice daily until samples maintained a constant weight. Fecal scores were assigned to each pen during the first 7 d of the experiment and were recorded by the same researcher each day to ensure consistency of scoring. Fecal scores were assigned on a scale that ranged from 1 (firm feces) to 4 (liquid consistency; Wellock et al., 2006).

Apparent Total Tract Digestibility of Nutrients

The phase 2 diet contained 0.5% titanium dioxide to act as an indigestible marker for nutrient digestibility determination. This diet was introduced to all pigs on day 4 of the experiment and pigs were allowed 5 d to adapt to the diet. On days 10, 11, and 12 of the experiment, fecal grab samples were collected from two randomly selected pigs per pen in the morning, pooled in a Ziploc bag (pen-basis), and stored at –20 °C. Moisture content of these fecal samples was determined as described above. After drying, feces were ground until they passed through a 1-mm screen and samples were pooled on a pen-basis across all 3 collection days. Pooled samples were stored in Whirl-pak bags until analysis.

Approximately 2 kg of the phase 2 diet was collected at mixing and on each fecal collection day and was stored at –20 °C. A subsample of feed from each collection day was pooled and sent to the University of Missouri Agriculture Experiment Station Chemical Laboratories (Columbia, MO) for proximate analysis and determination of titanium concentration. Similarly, dried and ground fecal samples (12–15 g) were submitted for proximate analysis and determination of titanium concentrations. Standard procedures (AOAC International, 2006) were followed for the analysis of moisture (method 934.01), ash (method 942.05), crude protein (LECO; method 990.03), crude fat (method 920.39), and crude fiber (method 978.19). Diet and fecal samples were analyzed for titanium concentration according to procedures of Myers et al. (2004). Gross energy content of diet and fecal samples was determined using an isoperibol bomb calorimeter (Model 6400, Parr Instruments, Moline, IL) using benzoic acid as the internal standard. Apparent total tract digestibility (ATTD) was calculated using the index method (Adeola, 2001) with the following equation:

$$\text{ATTD (\%)} = 100 - [100 \times ([\text{titanium in feed}] \times [\text{nutrient in feces}] / [\text{titanium in feces}] \times [\text{nutrient in feed}])]$$

Gut Permeability Measurement

A differential sugar absorption test was conducted to assess gut barrier function. On day 12 of the experiment, feed was removed at about 0800 h from eight randomly selected pens per water source for a 3-h fasting period and time of feed removal was recorded. Pigs had ad libitum access to water during the fast. Following the 3 h fast, 1 pig was selected at

random from each fasted pen (8 pigs/water source) for blood collection via blind venipuncture. Blood was collected into a heparinized tube (5 mL; Vacutainer; Becton, Dickinson and Company, Franklin Lakes, NJ) and a tube with no additive (5 mL; Vacutainer; Becton, Dickinson and Company, Franklin Lakes, NJ). The time of blood collection for each pig was recorded. Blood samples were stored on ice until further processing. Immediately following initial blood collection, each pig was dosed orally with a water-based solution that contained 15 g of a sugar mixture (90% lactulose, 6% L-rhamnose, 1.2% 3-O-methylglucose, and 2.8% D-xylose) in 200 mL of water. Each pig was dosed with 5 mL of the sugar solution. The sugar solution was administered carefully to maximize the chances that all the mixture was ingested by the pig. Two hours after dosing, blood was collected as described previously.

All blood samples were centrifuged at $2,000 \times g$ for 10 min at 4°C . Once centrifuged, plasma (1.0 mL) and serum (0.5 mL) were aliquoted from the heparinized and additive-free blood tubes, respectively, into new tubes, sealed, and stored at -80°C . Plasma samples were analyzed for concentrations of lactulose, L-rhamnose, xylose, and 3-O-methylglucose. Plasma samples were derivatized with 3-amino-9-ethylcarbazole through reductive amination according to the procedures of Han et al. (2013). Derivatized samples were injected into an Acquity ultra-performance liquid chromatography system (Water, Milford, MA) and separated in a BEH-C18 column. The liquid chromatography eluate was introduced directly into a SYNAPT G2-Si-QTOF mass spectrometer for detection according to the procedures of Ma et al. (2019). Mass chromatograms and mass spectral data were acquired and processed by MassLynx TM software. Individual sugar levels were determined by calculating the ratio between the peak area of sugar detected and the peak area of internal standard, and fitting with a standard curve using QuanLynx TM software.

Blood Chemistry and Cytokine Analysis

For blood chemistry and cytokine analyses, blood (5 mL) was collected in a heparinized vacuum tube from 1 pig per pen ($n = 45$) via blind venipuncture on day 8 of the experiment. Immediately following blood collection, each sample was placed on ice. After all samples had been collected (about 2 h), blood samples were centrifuged at $2,000 \times g$ for 10 min at 4°C , and plasma (0.5–1.0 mL) was aliquoted into two separate sample tubes and stored at -80°C until analysis. One of the two aliquots from each pen was analyzed at Marshfield Labs (Marshfield, WI) for blood chemistry (ANP2 Large Animal Profile). The other sample from each pen was analyzed for cytokine concentrations using a multiplex ELISA kit (MILLIPLIX Porcine Cytokine Magnetic Bead Panel; Merck Millipore; Darmstadt, Germany) following manufacturer's instructions at the University of Minnesota Cytokine Reference Laboratory.

Phagocytic Activity

On day 11, blood (1 mL) was collected in a heparinized tube from eight randomly selected pigs per experimental group (1 pig per pen) via blind venipuncture. Blood was kept at room temperature (20 – 25°C) during transport to the laboratory for processing within 24 h of collection. Using a PHAGOTEST kit (ORPEGEN Pharma, Heidelberg, Germany), whole blood was incubated with opsonized *Escherichia coli*-FITC (fluorescein

isothiocyanate) to evaluate phagocytic activity. Samples were processed following manufacturer's instructions and flow cytometry analysis was performed at the University of Minnesota Flow Cytometry Resource. Monocytes and granulocytes were gated using forward-scatter vs. side-scatter dot plots. Further, side-scatter vs. *E. coli*-FITC plots were used to determine the amount of phagocytic monocytes and granulocytes (Hodkinson et al., 2006).

Drinking Behavior

To evaluate pigs' acceptance of their assigned water source, drinking behavior was video recorded in randomly selected pens (5 pens/water source). Digital cameras (TruVision High Definition TVI Bullet Camera TVB-4403, Interlogix, Costa Mesa, CA) that were connected to a computer equipped with time-lapse video-recording software (Geo Vision Multicam Digital Surveillance System V8.2; USA Vision Systems Inc., Irvine, CA) were used to capture video footage of pigs at the drinker. Each pen was video recorded for 6 h per day (0900–1600 h) over the first 3 d of the experiment. Videos were viewed by the same researcher who was blinded to the experimental groups to avoid inter-observer discrepancy and subjective errors. The method of behavioral sampling (Martin and Bateson, 1993) was used to analyze drinking behavior from the videos. Drinking behavior was defined as a pig touching the drinker with its mouth (Li et al., 2005). The researcher recorded the number of drinking bouts (number of times pigs visited the drinker) and duration of drinking bouts (number of seconds a pig spent at the drinker per visit) of pigs in each pen for 6 h per day. Data were summarized as drinking frequency (number of drinks/pig/h), mean duration of drinking bouts (sec/drinking bout), and total amount of drinking time (or total amount of time spent at the drinker) over the observation period (sec/pig/6 h).

Statistical Analysis

Data were evaluated for the presence of outliers and normal distribution among experimental groups. Experimental data were analyzed using the PROC GLIMMIX procedure of SAS (Version 9.4, SAS Institute Inc., Cary, NC). Pen was considered the experimental unit. The statistical model included fixed effects of experimental group, room, and their interaction with random effects of pen. Means were separated using the PDIF option with the Tukey–Kramer adjustment for multiple comparisons. A similar model was used for a repeated measures analysis to evaluate changes over time. Count data (fecal score, days on medical treatment, number of pigs treated, and number of dead pigs) were analyzed using the PROC FREQ procedure with chi-square. The significance level was declared at $P < 0.05$ and trends are described at $0.10 > P > 0.05$.

RESULTS AND DISCUSSION

Water Storage and Quality Management

Within each experimental group, minimal changes occurred in analyte concentrations of water from the initial sample collected at the source on commercial farms in July 2019 to completion of the experiment in Morris, MN in October 2019 (data not shown). Differences in water quality that were observed initially for each water source when sampled at the source well (Table 1) were the same differences observed 3 mo

later at the end of the experiment. This observation implies that quality of well water does not change rapidly. This observation agrees with work of Vinten and Dunn (2001) who reported that concentrations of analytes in well water did not change over the course of 10 yr. Average water flow at the drinker cup in pens over the entire experiment did not differ among groups (Fig. 1). However, during week 2, Water source B's drinkers had a lower flow rate than Water source C's but was not different than Water source A. This aberration resulted in additional monitoring of water flow rates and an adjustment of pressure for Water source B to improve consistency. The aberrant low flow rate returned to the desired level during week 3 and was the same as Water sources A and C for the remainder of the experiment.

Temperatures around each water storage bladder were summarized as 12 h averages for both daytime (0700–1900 h) and nighttime (1900–0700 h). The range of daytime temperatures was 21 to 5 °C and 20.5 to 4.9 °C for nighttime (data not shown). Storage temperatures across all three water sources were not different at any time during the experiment.

Pig Growth Performance

We observed no differences in body weight of pigs among experimental groups at conclusion of the study (Table 3). Similarly, ADG, ADFI, and G:F (Fig. 2) of pigs over the entire experiment were not different among pens of pigs consuming the three sources of water. Furthermore, there were no differences among water sources in ADG (Fig. 3), ADFI (Fig. 4), or G:F (Fig. 5) of pigs at any week throughout the experiment. We theorized that pigs would be most sensitive to differences in water quality during the early portion of the nursery period when pigs experience stress associated with the weaning event. We expected these differences to diminish as pigs acclimated to their new environment and their assigned water source. Clearly, quality of water did not influence growth performance of pigs at any time during this experiment.

Apparent Total Tract Digestibility of Diet

The ATTD of diet dry matter, crude protein, crude fiber, ether extract, and gross energy in pigs consuming the phase 2 diet was not affected by the water source that pigs consumed

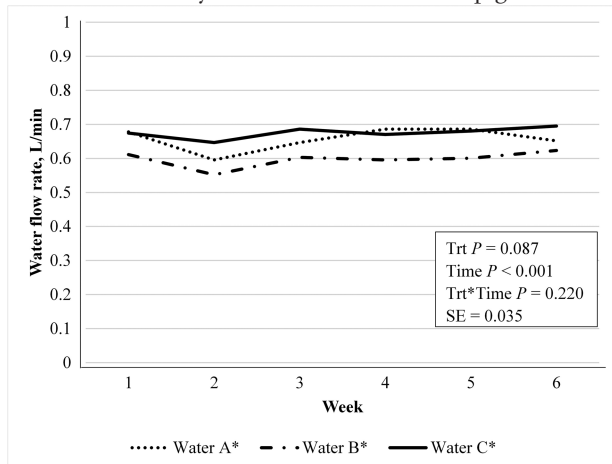


Fig. 1. Average water flow rate at each drinker over the 6-wk period in pens with different qualities of water. * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

Table 3. Effect of differing water qualities on initial and final BW of nursery pigs

Item	Water source ¹			SE	P-value
	A	B	C		
No. of pens	15	15	15	–	–
No. of pigs	150	150	150	–	–
Initial BW, kg	6.18	6.20	6.34	0.150	0.691
Final BW, kg	24.60	24.55	25.37	0.440	0.357

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

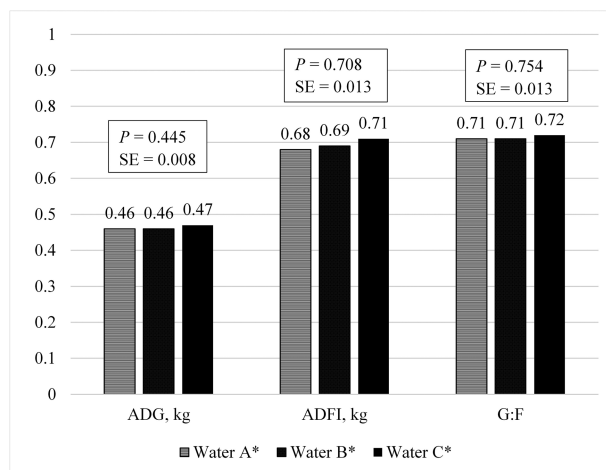


Fig. 2. Overall growth performance of nursery pigs fed different water sources * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

(Table 4). The average ATTD of crude fat and crude fiber that we present is a positive value. However, there were some observations that had negative ATTD of crude fat and crude fiber (17 and 6 negative values, respectively). Negative values for ATTD of a nutrient indicate that excretion of the nutrient was greater than intake of the nutrient. The greater excretion than intake of a nutrient may be the result of endogenous losses that are greater than intake of the nutrient. Although endogenous losses of amino acids and lipids are commonly described events, endogenous losses of fiber have been described only recently (Montoya et al., 2016). The ATTD of ash among pigs fed the phase 2 diet was affected by the water source fed to pigs ($P = 0.016$). Pigs consuming Water source C had greater ($P < 0.05$) ATTD of ash compared with pigs consuming Water sources A and B. Both ash and TDS are similar in that they are composed mainly of inorganic minerals. Interestingly, ash digestibility was greater for pigs fed water with the lowest concentration of TDS. Total dissolved solids concentration of Water source C was 25% and 33% of that present in Water sources A and B, respectively (Table 1). The low TDS concentration of Water source C may have elicited improved apparent ash digestibility. However, the observed improvement in ash digestibility may have been an

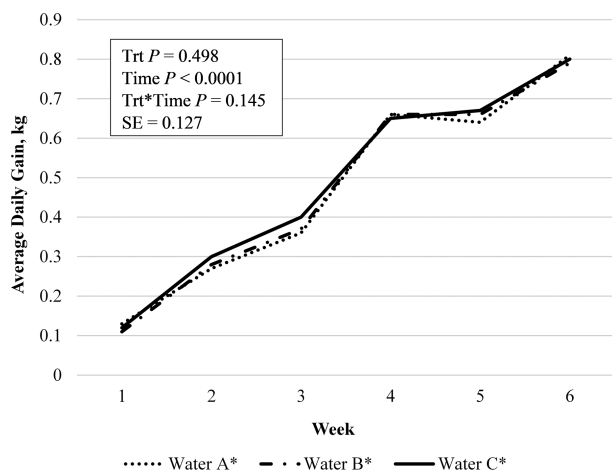


Fig. 3. Effect of water quality on average daily gain of nursery pigs over time * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

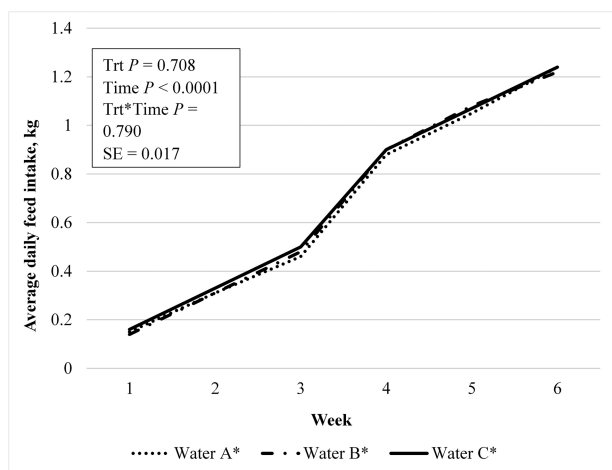


Fig. 4. Effect of water quality on average daily feed intake of nursery pigs over time * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

artifact of our digestibility measurements. Ash consumed by pigs in water was not considered in our measurement of ash intake because we were not able to measure water intake of pigs in this experiment. However, ash present in water was measured as part of fecal ash. Consequently, the relatively higher TDS content of Water sources A and B may have resulted in a lower estimate of ash digestibility compared with Water source C.

Intestinal Permeability

To determine permeability and sugar absorptive capacity of the intestine, an orally administered, sugar mixture was used to measure ratios of specific sugars in blood, a similar approach to methods used in humans (Zuckerman et al., 2004; Wijtten et al., 2011). A low ratio of d-xylose to l-rhamnose indicates a low permeability and a healthy intestine (Zuckerman et al., 2004). An increased ratio of l-rhamnose

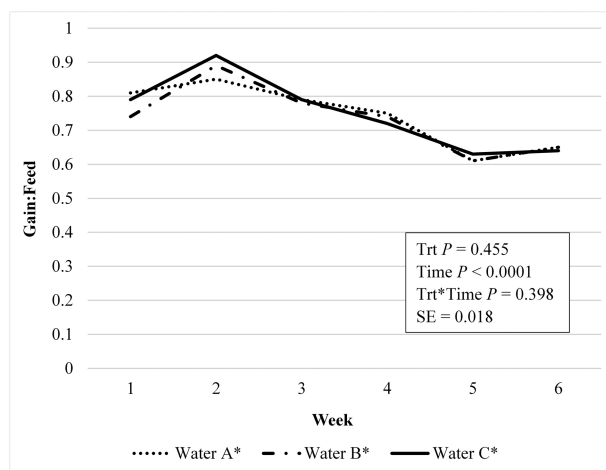


Fig. 5. Effect of water quality on gain efficiency of nursery pigs over time * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

Table 4. Effects of water quality on apparent total tract digestibility of nutrients in diets fed to nursery pigs (days 10, 11, and 12 of experiment)

Item, %	Water source ¹			SE	P-value
	A	B	C		
No. of observations	15	15	15	–	–
Dry matter	79.05	78.00	78.30	0.41	0.195
Crude protein	71.72	69.84	70.05	1.07	0.170
Crude fiber	26.04	16.34	16.17	3.34	0.995
Ash	56.57 ^a	56.55 ^a	59.02 ^b	0.66	0.016
Ether extract	5.62	2.40	6.60	4.69	0.804
Gross energy	76.45	75.12	75.24	0.50	0.125

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

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

to 3-O-methyl-glucose indicates increased absorptive capacity of the intestine (Zuckerman et al., 2004). In humans, an increased ratio of d-xylose to 3-O-methyl-glucose is associated with presence of illness or disease due to a decrease in d-xylose metabolism after absorption (Zuckerman et al., 2004). Changes in intestinal permeability result from compromised integrity of the gut epithelial wall, which allows unwanted material from the lumen to enter the blood stream (McLeod et al., 2019). Conversely, intestinal absorption is the movement of desirable nutrients from the small intestinal lumen into the blood supply (Kiela and Ghishan, 2017). We observed no differences in the ratios of d-xylose to l-rhamnose, l-rhamnose to 3-O-methylglucose, or xylose to 3-O-methylglucose resulting from exposure to the different water sources (Table 5). These findings suggest that water fed to pigs did not influence barrier function or absorptive capacity of the gastrointestinal tract. However, we cannot disregard the possibility that time between initial and final blood samples was too long allowing sugars to be metabolized or

discarded in urine. We were not able to collect urine in this study to measure sugars that may have appeared in urine.

Pig Morbidity and Mortality

Number of pigs treated for sickness and total number of injections administered were not different among water sources evaluated in this experiment (Table 6). Mortality for the entire experiment was 0.44% among all water sources with only 2 mortalities which were unrelated to quality of water consumed. Mortality rate in about 7,600 groups of nursery pigs annually averaged from 2.97% to 3.24% in 2017 through 2019 (MetaFarms and National Pork Board, 2020). The extremely low mortality rate of pigs in the current study relative to mortality rates commonly observed in the U.S. pork industry indicates the high health status of pigs in this experiment. Percentages of fecal moisture (Fig. 6) and subjective fecal scores (data not shown) were not different among water sources from days 3 to 7 of the study. Fecal moisture determinations were not possible on days 1 and 2 due to a lack of fecal matter produced by pigs.

Immune System Characteristics and Blood Chemistry

Cytokines

Cytokines are important in mediating and regulating immune and inflammatory responses. The water sources provided to pigs had no influence on proinflammatory, intermediary, or anti-inflammatory cytokine concentrations in blood (Table 7).

Phagocytic activity

We evaluated the ability of blood immune cells to phagocytize opsonized, labeled *E. coli*. Opsonization is the process

by which bacteria can easily be engulfed by phagocytes. Therefore, when less opsonin-coated bacterial cells are present, there are more monocytes and granulocytes available to pursue phagocytosis (Magnusson and Greko, 1998; Hodkinson et al., 2006). Higher percentages of active monocytes and granulocytes are desired to participate in phagocytosis, increasing the pig's ability to fight infections. We observed no differences in the percentage of total monocytes

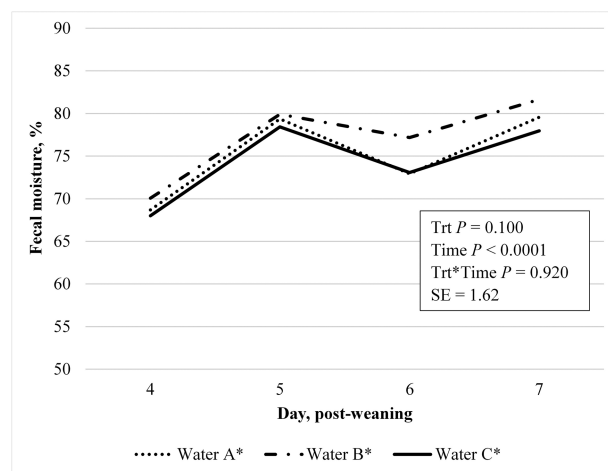


Fig. 6. Effect of water quality on average fecal moisture (%) of nursery pigs over time (days 4 through 7 post-weaning) * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

Table 5. Effect of water quality on intestinal integrity of nursery pigs

Ratio	Water source ¹			SE	P-value
	A	B	C		
No. of observations	8	8	8	–	–
Xylose/rhamnose	2.97	1.99	2.39	0.457	0.337
Rhamnose/3-O-methyl-glucose	1.39	2.03	1.36	0.237	0.143
Xylose/3-O-methyl-glucose	3.67	3.64	3.03	0.557	0.655

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

Table 6. Effect of water quality on morbidity and mortality of nursery pigs

Item	Water source ¹			P-value
	A	B	C	
Total pigs, No.	150	150	150	–
Pigs treated, No.	9	5	8	0.472
Injections administered, No. ²	20	19	18	0.606
Mortality, No.	0	1	1	–

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

²Injections administered for pigs that exhibited compromised health.

Table 7. Effect of differing water qualities on plasma cytokine concentrations (pg/mL) of nursery pigs (day 8 of experiment)

Item	Water source ¹			SE	P-value
	A	B	C		
No. of observations	24	24	24	–	–
GM-CSF ²	1,605	1,748	1,566	–	1.000
IFN γ ³	89,783	88,097	120,401	4,867	0.960
IL-1 α ⁴	700	682	693	24	0.860
IL-1 β ⁵	3,758	3,544	3,587	145	0.538
IL-1ra ⁶	7,201	7,059	6,383	329	0.223
IL-2	11,234	10,497	10,648	419	0.431
IL-4	118,032	115,685	120,401	8,464	0.931
IL-6	4,049	3,684	3,884	215	0.480
IL-8	523	535	445	31	0.101
IL-10	20,102	19,415	19,776	3,049	0.899
IL-12	3,246	3,353	3,442	117	0.845
IL-18	32,194	31,394	32,215	1,318	0.891
TNF ⁷	1,248	1,261	1,050	89	0.195

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

²Granulocyte-macrophage colony-stimulating factor.

³Interferon-gamma.

⁴Interleukin (IL)-1 alpha.

⁵IL-1 beta.

⁶IL-1 receptor antagonist.

⁷Tumor necrosis factor.

Table 8. Effect of water quality on percentage of total monocytes and granulocytes displaying phagocytosis in nursery pigs (day 11 of the experiment)

Item	Water source ¹			SE	P-value
	A	B	C		
No. of observations	8	8	8	–	–
Phagocytic Monocytes, %	74.28	73.20	74.49	2.296	0.913
Phagocytic Granulocytes, %	95.27	93.56	93.79	0.967	0.451

¹A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); C (235 mg Eq CaCO₃/L hardness, 2 ppm sulfate, 348 ppm TDS).

or granulocytes that displayed phagocytosis from the blood of pigs exposed to the different water sources (Table 8).

Blood chemistry

Blood chemistry was performed for 22 different parameters including glucose, aspartate aminotransferase (AST), sorbitol dehydrogenase (SDH), bilirubin, cholesterol, total protein, albumin, urea nitrogen, creatinine, phosphorus, calcium, potassium, sodium, chloride, creatine kinase (CK), gamma-glutamyl transferase (Gamma-GT), anion gap, globulin, albumin-globulin ratio (A/G ratio), urea-creatinine ratio, sodium-potassium ratio, and bicarbonate. Except for bilirubin concentration, water quality did not influence blood chemistry of pigs among water sources fed in this experiment (data not shown). Furthermore, blood parameters observed for pigs fed each water source were within reference ranges for swine provided by Marshfield Labs.

Bilirubin of pigs fed Water source A was higher ($P = 0.030$) than for pigs fed Water source C (0.35 vs. 0.16 mg/dL, respectively). The reason for elevated bilirubin concentration in pigs fed Water source A is not clear. Elevation of blood bilirubin levels in pigs can result from starvation or near starvation (Cornelius, 1980; Smith et al., 2013), which suggests that pigs had low feed consumption post-weaning. However, feed intake in the first 7 d of the experiment for pigs fed Water source A was 110 to 180 g/d, which was not different from the intake of pigs drinking Water source B (110–180 g/d) and Water source C (130–170 g/d). Feed intakes recorded in this study are consistent with industry published data indicating that 150 to 200 g/d during the first week post-weaning is common among commercial production conditions (Whittemore et al., 2001). Thus, reduced feed intake of pigs fed Water source A does not explain elevated bilirubin concentration of these pigs.

Drinking Behavior

Drinking frequency and mean duration of drinking bouts were not influenced by the source of water pigs consumed on day 1, 2, or 3 of the experiment (data not shown). Consequently, total amount of drinking time during the observation period did not differ among water sources over the 3 d of data collection (Fig. 7). These results suggest that water sources did not affect drinking behavior and pigs were willing to drink the water provided to them.

General Discussion

Results from McLeese et al. (1992) found that growth performance was decreased in pigs consuming water containing 4,390 ppm TDS and fed an unmedicated diet compared to

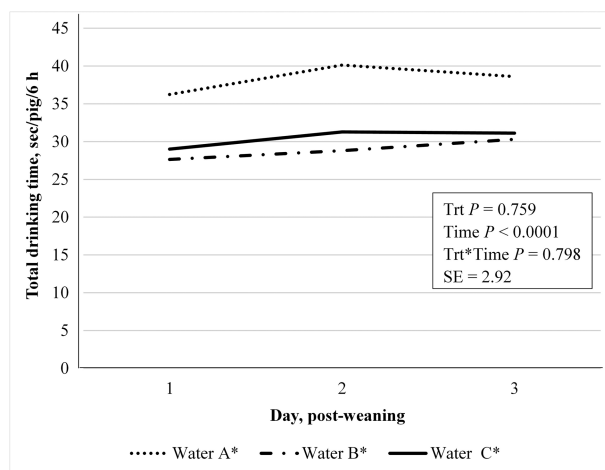


Fig. 7. Effect of water quality on total time spent at the drinker during 6 h (0900–1600 h) of each observation day per pig (days 1 through 3 of the experiment) * Water A (1,410 ppm hardness [CaCO₃ equivalent], 1,120 ppm sulfate, 1,500 ppm TDS); Water B (909 ppm hardness [CaCO₃ equivalent], 617 ppm sulfate, 1,050 ppm TDS); Water C (235 ppm hardness [CaCO₃ equivalent], 2 ppm sulfate, 348 ppm TDS).

pigs fed a medicated diet. In the same experiment, there was a tendency for pigs to grow faster when consuming water with a low TDS level and fed a medicated diet (McLeese et al., 1992). The three sources of water compared in our study did not yield differences in growth performance or health of nursery pigs. We expected to see differences in some response variables, but we also recognize that pigs may tolerate water sources of varying quality.

Specific water sources used in this study were selected from a survey of Minnesota pig farmers designed to understand the range of water quality being fed to nursery pigs. Water was initially sampled from 15 barns based on survey responses. All water samples were analyzed for 29 different characteristics. From these analyses, three water sources representing the most extreme range in characteristics were selected for feeding to nursery pigs (Lozinski, 2020). Our intent was to select two water sources that might be perceived to be of poor quality and one source perceived as good quality. Except for the sulfate concentration in Water source A, none of the selected water sources exceeded maximum recommendations for concentrations of analytes outlined by the CCME (1987) or NRC (1974). The two water sources with the highest concentrations of TDS (A and B), and presumably the poorest quality, were expected to support poorer growth performance of pigs compared with Water source C which contained a much lower TDS concentration. However, no differences in nursery pig performance or health were observed across water sources. In retrospect, this response is not surprising when one considers that the TDS concentration of water sources used in our experiment was lower than that of water used in the work reported by McLeese et al. (1992). Although we studied water sources with quite divergent characteristics that are reflective of those available in commercial pig production, they may not have been of poor enough quality to affect pig performance.

At weaning, pigs are expected to adapt quickly to a new environment (Patience, 2013) which includes the water they are offered. Pigs adapt to the quality of water presented to them within a few weeks (NRC, 2012; Patience, 2013) of initial exposure. Pigs adapt to differing water quality more quickly

if concentration of sulfates and TDS are low (< 1,000 ppm; Paterson et al., 1979; McLeese et al., 1992; Patience, 2013). Because the concentrations of minerals in water sources used in this experiment were mostly less than the recommended maximum values, we suspect pigs adapted very quickly to the water they were offered.

In conclusion, the three water sources fed to nursery pigs had no effects on growth performance or health of pigs, diet digestibility, gut permeability, immune response characteristics, or drinking preference. Based on these observations, we conclude that the quality of water studied did not affect nursery pig performance or health. Water that exceeds recommended guidelines for commonly evaluated constituents might influence performance and/or health of nursery pigs negatively, especially if pigs experience health challenges that compromise their immune systems.

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Conflict of Interest Statement

None declared.

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