DIGESTIBILITY OF SUPPLEMENTAL ZINC SOURCES IN SOW DIETS AND

EFFECTS OF SUPPLEMENTAL ZINC ON PIGLET SURVIVABILITY

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DEDICATION

I would like to dedicate this thesis to my parents, Douglas and Kimberly Holen, and to my siblings (Kaitlyn, James, Mathias, Marcus, Grant, and Delaney). I would not have been able to complete this graduate program without your constant love, support, and encouragement.

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CHAPTER 1: INTRODUCTION

Geneticists have improved genetic potential for commercial sow production measures immensely within the last 10 to 15 years in the United States. Pork producers, veterinarians, and nutritionists have realized this improved genetic potential such that litter size at birth continues to trend upwards for today's commercial sows (Knauer and Hostetler, 2013; Stalder, 2018), with many farms consistently reporting 13 or more piglets born per litter (Knox et al., 2013; Metafarms Analytical Team, 2017). Yet, as the total number of piglets born has been increasing, piglet mortality has also continued to increase and thus, the number of pigs weaned per litter has remained relatively steady (Stalder, 2018). Some of the major contributors to piglet mortality include stillbirths, low viability, trauma from crushing, and starvation within the first few days of life (Vaillancourt et al., 1990; Edwards and Baxter, 2015). Numerous methods have been investigated to mitigate causes of high pre-weaning mortality. Factors influencing litter size such as genetics, environment, management, and nutritional interventions often become intertwined and directly influence production outcomes (Figure 1.1). As a result, one factor alone is unlikely to reduce pre-weaning mortality. Therefore, it is essential to consider a coordination of these factors to optimize survivability of piglets.

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Body condition ————————→ Ovulation rate ←————— Hormonal stimulation
                                         ←——————— Gilt selection
                                         ←——————— Boar stimulation

Vitamins and minerals                    ————— Semen quality
                          Fertilization rate ←——— Mating technique
                                         ←——— Stockmanship
Feed intake/appetite      Embryo survival ←————— Stress management

                                              ————— Housing
                          Piglets born ←——————— Farrowing management
Nutrition

                                              ————— Pre-weaning mortality
                          Piglets weaned ←——————— Milk yield
                                              ————— Health Status
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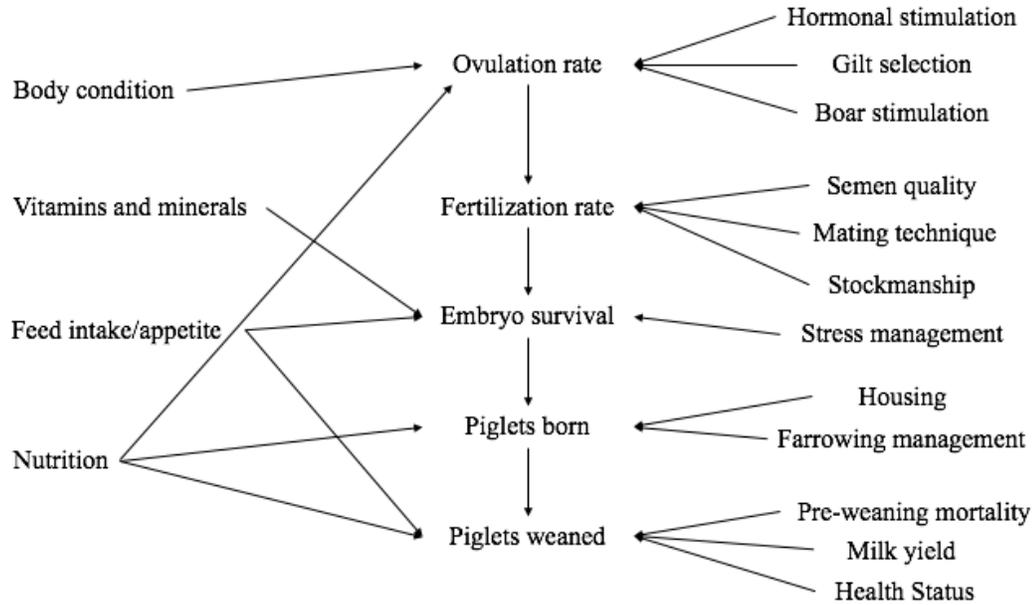
Figure 1.1. Factors contributing to litter size in sows (adapted from Close and Cole, 2004)

Even before birth, there may be potential to decrease pre-weaning mortality.

Clearly, the conceptus relies completely on the dam for proper growth and development

(Hidiroglou and Knipfel, 1981; Vonnahme, 2012; Dunlap et al., 2015). Some of the

nutrients essential for proper growth and development include trace elements, such as

zinc (Zn), which play a crucial role in reproduction (Hostetler et al., 2003). Zinc

deficiency in pregnant animals is widely known to cause catastrophic outcomes such as

abortion, intrauterine growth retardation, and teratogenic effects (Keinholz et al., 1961;

Swenerton and Hurley, 1968; King, 2000).

Even though Zn is an essential trace element for optimal reproductive

performance, requirements for dietary Zn in sow diets were established more than 40

years ago (NRC, 2012) and have not been re-evaluated since. Furthermore, Zn may be

supplemented in diets using either organic or inorganic sources. Among common

sources, digestibility and bioavailability of Zn to the pig varies greatly (Wedekind et al.,

1994; Schlegel et al., 2013). Other factors such as the presence of phytate, Ca, Fe, Cu, Cd, and Co in the diet may also influence utilization of Zn from dietary sources (Oberleas, 1983; Solomons, 1983; Solomons, 2001). Much of the literature available today provides answers to questions regarding digestibility and bioavailability of Zn for nursery and grower-finisher phases of swine production. However, very limited information is available specifically for sow diets in gestation and lactation. As a result, industry nutritionists may not be formulating diets that fully maximize sow and piglet performance while minimizing potentially negative environmental consequences.

As with any livestock production system, consumer demands and perceptions must be considered to achieve sustainable pork production. Environmental impacts of pork production, such as manure nutrient excretion and utilization as a soil amendment, are growing concerns to the public. Often, producers supplement diets with high concentrations of Zn to improve growth performance of pigs post-weaning (Carlson et al., 1999; Hill et al., 2001). However, this practice significantly increases total Zn, Fe, Cu, and Mn excretion in animal manure (Meyer et al., 2002), risking excessive levels of trace mineral incorporation on cropland. As a result, the European Commission has determined that the use of pharmacological levels of Zn oxide be prohibited by 2022 to minimize risk of excessive Zn excretion in manure (Beek, 2017). Environmental concerns will likely continue to be a point of emphasis from government and public audiences. Therefore, all pork producers must be prepared to implement practices that efficiently utilize nutrients and minimize mineral excretion for all phases of swine production.

CHAPTER 2: LITERATURE REVIEW

Commercial Swine Production

Sow productivity

Sow productivity has changed drastically in the last 30 to 40 years. In the last 10 years alone, the number of piglets born per litter has steadily increased from 12.5 in 2008 to 13.9 piglets born per litter in 2017 (Stock et al., 2014; Stalder, 2018). Furthermore, modern sows are much leaner, larger in mature body size, have greater milk yields, and support increased piglet growth rates compared to sows from 30 years ago (Close and Cole, 2004). If sows are producing greater number of piglets born and born alive per litter, one may speculate that a similar trend should occur regarding the total number of piglets weaned per litter. However, the number of piglets weaned per litter has remained relatively consistent, especially in the last five years (Stalder, 2018). After birth, the pig is subject to a variety of challenges, such as competition with littermates and environmental changes, that may create risk of low viability or death. Overall piglet viability is of great importance to ensure adequate growth and development later in life. Many factors cannot be easily controlled in the post-natal period of growth, but there may be more direct interventions within the prenatal environment to positively impact growth and survival of piglets.

Fetal growth and development is influenced heavily by the maternal environment, beginning as early as the initial stages of embryonic development (Robinson et al., 1995) and accelerates as pregnancy progresses (Ullrey et al., 1965; Knight et al., 1977; Richards, 1999; McPherson et al., 2004). Recent comparisons by Kim (2010) demonstrated that today's porcine fetus in the late portions of gestation is 40% heavier

than in the previous 40 years, sometimes leading to challenges regarding piglet survival

of smaller birthweight pigs (Figure 2.1). Heavier litters typically require more nutrients

from the sow to support fetal growth, but limited nutrient intake due to restricted feed

intake of pregnant sows, especially in late gestation, may negatively affect survival of

some piglets.
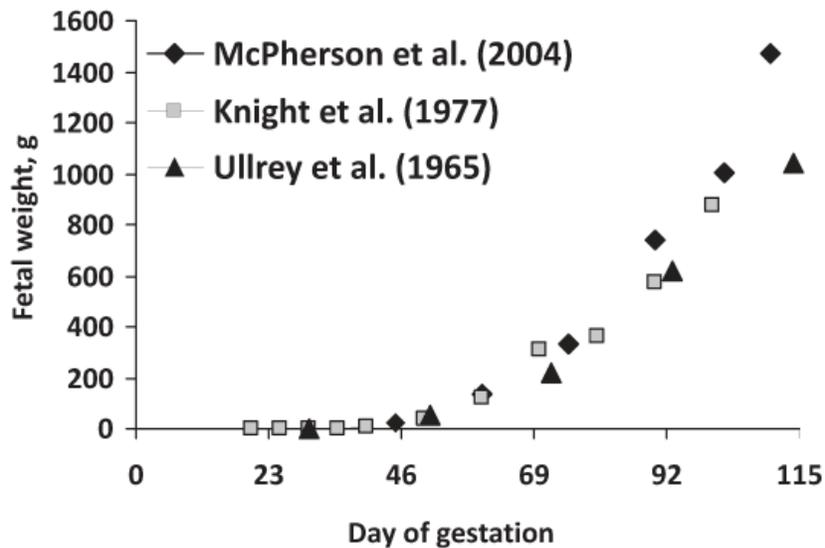
Figure 2.1. Fetal growth during gestation (Kim et al., 2010)

Ideally, sow productivity will improve year after year. However, some biological

factors specific to the sow may eventually limit dramatic advancements. Uterine capacity

is defined as the ability of the uterus to maintain normal development of conceptuses, and

is directly related to the number of piglets born and born alive (Vallet, 2000). Therefore,

increased uterine capacity will likely result in an increased litter size. Uterine, placental,

and fetal factors affect the overall uterine capacity of the sow (Vallet et al., 2014), but

this capacity becomes limited as the sow enters late gestation. The placenta, which is

responsible for nutrient transport, is smaller relative to piglet size as the sow enters this

reproductive stage compared with early gestation (Biensen et al., 1998). Several factors such as placental growth, utero-placental blood flow, nutrient availability, and placental metabolism and transport capacity contribute to overall nutrient delivery to fetuses (Dunlap et al., 2015). However, poor nutritional status of the sow is particularly detrimental to fetal growth due to the increased nutrient requirements for both the dam and fetuses during any stage of pregnancy. As genetic lines continue to be selected for increased litter size, intra-uterine crowding of fetuses may occur, often negatively affecting prenatal growth and survival (Foxcroft et al., 2009). In such situations, intrauterine growth retardation resulting from both limited uterine capacity and inadequate maternal nutrition, reduces the fetus' chance of survival (Wu et al., 2006). Should fetal growth become restricted, the risk of post-natal mortality and morbidity increases (Vonnahme, 2012).

Body weight of the piglet at birth has lifelong implications on growth rate, efficiency of growth, and carcass quality. Typically, as birth weight decreases, the likelihood of survival and optimal performance decreases. Researchers determined that low birth weight piglets at weaning continued to have lower bodyweights than their contemporaries at the end of the nursery period (Larriestra et al., 2006). Further, carcass quality and value of the pig at harvest decreases (Rehfeldt et al., 2008; Fix et al., 2010) as birth weight declines. Although lifetime performance of the pig includes many factors, low birth weight or weaning weight is a clear contributor to depressed post-natal performance (Douglas et al., 2013) and mortality (Calderon Diaz et al., 2017).

Not all low birthweight piglets are destined for poor growth performance. There are interventions to mitigate some of the negative impacts of low birthweight. Piglet

management practices such as maintaining smaller litter sizes through cross-fostering from birth to weaning, can improve post-natal performance of low birthweight pigs (Deen and Bilkei, 2004). In fact, some piglets are capable of compensatory growth at some point in the production cycle to match that of normal pigs at weaning or at market weight (Bee, 2004; Paredes et al., 2012; Douglas et al., 2013; Pardo et al., 2013). Nutritional interventions may serve a role in improving growth or survival of small piglets (Vallet et al., 2014), but this is not always effective (Douglas et al., 2014).

Still, as geneticists continue to focus on selection to increase litter size, many small pigs experience a greater risk of death shortly after birth. As depicted in Figure 2.2., many factors contribute to overall piglet pre-weaning mortality, but a few factors including stillbirth, disease, crushing, and starvation are especially detrimental (Vaillancourt et al., 1990; Lay et al., 2002). Pre-weaning mortality is one of the most vital concerns for pork producers because the pre-weaning mortality rates of piglets can reach 20% (Edwards and Baxter, 2015), and contribute to significant economic losses. Furthermore, scenarios for increasing mortality may create animal welfare concerns (Baxter and Edwards, 2017).

Figure 2.2. Factors contributing to piglet mortality (Edwards and Baxter, 2015)

A review conducted by Alonso-Spilsbury et al. (2007) suggested that a pre-weaning mortality rate of 8% is achievable through proper piglet management, specifically during the first three days after parturition. Herpin and Dividich (1995) determined that effective management practices such as: farrowing induction and supervision, assistance with respiration and colostrum intake for weak piglets, and provision of warm farrowing crate temperatures in a creep area through supplementary heat sources will likely reduce pre-weaning mortality rates. Beyond management practices, genetic selection for improved piglet survival rates, maternal behavior, and/or placental efficiency may also provide potential routes for reducing pre-weaning mortality of piglets (Van Rens et al., 2005; Roehe et al., 2009; Baxter et al., 2011). Although effective approaches to decrease mortality of piglets pre-weaning are available, it is still difficult to achieve very low mortality.

Trace minerals play an essential role in proper fetal growth and development of piglets. Zinc, copper, and manganese have a great impact on reproduction and are required for normal growth and development (Vallee and Falchuk, 1993; Hostetler et al., 2003). Without a doubt, fetal growth depends on maternal transfer of such nutrients and, in the case of poor or inadequate mineral supply, negative consequences are likely. There are numerous detrimental effects of Zn deficiency on reproductive performance including: difficulty farrowing, increased length of gestation, increased frequency of abortions, piglet malformations, growth retardation, and low birth weights (Favier, 1992; Bedwal and Bahuguna, 1994). A more recent study conducted by researchers at the U. S. Meat Animal Research Center concluded that high Zn supplementation in late gestation decreased stillbirth rate and increased subsequent survival of low birth weight pigs (Vallet et al., 2014).

Environmental concerns associated with swine production

Pressure from both government and the public regarding environmental impacts of animal agriculture continues to increase. The rise of intensive swine production in some regions of the world has exacerbated this pressure. Pork producers have been striving for processes and management practices to mitigate environmental impacts through reducing nutrient and odor emissions. A specific goal for producers and consumers alike is to minimize excretion of nitrogen, phosphorus, zinc and copper in manure which is ultimately applied to cropland. Excretion of these minerals have potential to accumulate in soil, run off into waterways, or contribute to air pollution (Jongbloed and Lenis, 1998). Excessive accumulation of these minerals may lead to phytotoxicity (Long et al., 2003), eutrophication of surface water (Carpenter et al., 1998),

and therefore, unsustainable crop production. Often, crops utilized for swine feeds are grown in concentrated areas that also contain conventional swine facilities, such as the Midwestern United States. Therefore, it is especially crucial for animal agriculture in these regions to minimize negative impacts on cropland. Researchers have identified numerous nutritional strategies (Table 2.1.) that provide practical and effective options to reduce nutrient excretion (Kornegay et al., 1997; van Heugten and van Kempen, 2000; Ferket et al., 2002).

Table 2.1. Potential reductions in nutrient excretions by nutritional strategies (adapted from Ferket et al., 2002 and van Heugten and van Kempen, 2000)

Strategy	Potential reduction in nutrient excretion
Accurate formulation to requirements	6-15% for all nutrients
Minimize feed waste	1.5% for all nutrients for every 1% reduction
Pelleting feed	5% for N, P, Zn, Cu
Grind feed to 700 - 1,000 µm	5% for N, P, Zn, Cu
Use highly digestible ingredients	5% for N and P
Reduce ingredient variability/quality control	10-25% for N and P
Reduce protein/amino acid supplementation	9% for N for every 1% reduction in dietary CP
Low-phytate corn	25-50% for P
Phytase/low dietary phosphorus (P)	2-5% for N, Zn, and 20-30% for P
Phytase/High available P (HAP) corn	2-5% for N, Zn, and 20-40% for P
Growth promoting feed additives	5% for all nutrients
Phase feeding	5-10% for N and P
Split-sex feeding	5-8% for N
Reducing microminerals/organic minerals	Up to 50% for Zn, Cu, Mn

Swine raised in commercial operations are offered diets that meet mineral requirements for their age, size, and physiological state. Usually, this requires inclusion of plant-based feedstuffs that are low cost and highly digestible to supply the most expensive nutritional components to the animal. For example, in the Midwest U. S., corn and soybean meal provide most of the energy and amino acids required by pigs. Any macrominerals, vitamins, and microminerals that are not adequately provided or available

in the primary dietary feedstuffs are supplemented in a premix so that all requirements can be met. Requirements for all phases of swine production are reported in NRC (2012). However, variation in and limited knowledge about bioavailability of trace minerals (e.g. Zn and Cu) in all ingredients, especially in corn and soybean meal, create challenges for nutritionists to formulate diets that meet requirements but also minimize nutrient excretion. Diets cannot be formulated accurately to minimize mineral excretion in manure without clearly understanding bioavailability of minerals in diet components.

Mineral requirements for swine are determined as the minimum daily intake required to avoid deficiency, and not the amount necessary for optimal animal performance. In the past, industry nutritionists have fed diets containing greater concentrations of trace minerals than the established dietary requirement (Table 2.2). This nutrient allowance, or safety margin, can lead to excess mineral excretion due to digestibility and utilization variation among mineral sources. There is obvious potential for improved efficiency of mineral nutrition for swine. The European Commission has already established a plan to control excessive Zn excretion in manure (Beek, 2017). Members of the swine industry in the United States must also be prepared to follow mineral excretion regulations if they are established in the future.

Table 2.2. Industry range of mineral concentrations of sow and finishing swine diets (adapted from NRC, 2012 and Flohr et al., 2015)

Mineral	NRC (2012) Requirement	Range[1]	Median[1]	Average[1]
Sow diets				
Copper, ppm	10 to 20	7 to 35	15	16
Iron, ppm	80	45 to 165	100	102
Manganese, ppm	25	21 to 70	38	32
Zinc, ppm	100	57 to 165	125	113
Early finishing swine diets[2]				
Copper, ppm	4	5 to 242	136	112
Iron, ppm	60	39 to 124	86	87
Manganese, ppm	2	7 to 40	29	25
Zinc, ppm	60	30 to 150	110	99
Mid-finishing swine diets[3]				
Copper, ppm	3	4 to 162	109	82
Iron, ppm	40 to 50	33 to 124	73	75
Manganese, ppm	2	6 to 40	22	21
Zinc, ppm	50	30 to 131	89	85
Late finishing swine diets[4]				
Copper, ppm	3	3 to 161	10	66
Iron, ppm	40	31 to 103	63	66
Manganese, ppm	2	3 to 40	19	19
Zinc, ppm	50	30 to 131	75	74

[1]From Flohr et al. (2015) and represents reported data from 39.4% of U.S. sow herd
[2]23-53 kg body weight
[3]54-99 kg body weight
[4]100-135 kg body weight

Role of Zinc in Swine Nutrition

Mineral nutrition of sows

Sow productivity is heavily impacted by a combination of minerals during the reproductive cycle (Figure 2.3). Whole body content of macro- and micro-minerals quantitatively increases from birth to maturity in pigs (Mahan and Shields, 1998). Additionally, supplying diets that exceed mineral requirements in developing gilts will likely enhance tissue storage of some minerals and thus, improve lifetime productivity of the sow. Storage of some macro- and micro-minerals in tissues can be mobilized and

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utilized during periods of high demand, such as during lactation. But, if the excretion or utilization of minerals exceeds body stores and daily mineral intake, body reserves of Ca, P, Mg, K, Na, Al, Zn, and Cu within the sow can eventually become depleted (Mahan and Newton, 1995) and sow longevity may be compromised (Mahan, 1990). Therefore, mineral status and nutrition of sows is essential to successful long-term reproductive performance.
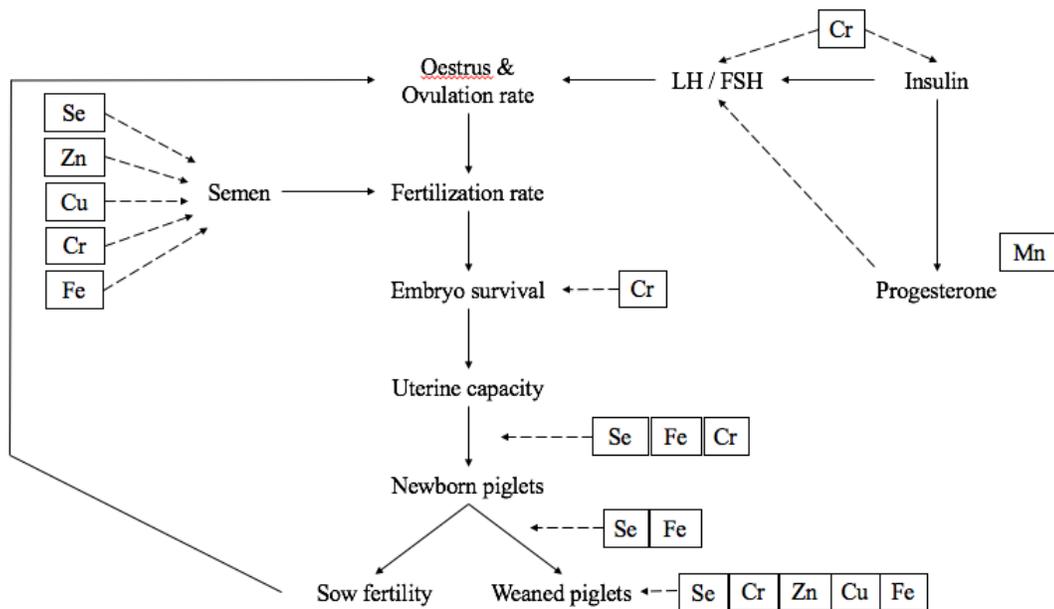
Figure 2.3. Role of minerals in sow reproduction (adapted from Close, 2010). Dashed lines indicate potential influences of trace minerals.

Importance of dietary zinc

Zinc is necessary for many metabolic functions to ensure animal health, reproduction, growth, and development because it is a fundamental component of more than 300 enzymes (McCall et al., 2000). More specifically, Zn is essential for gene expression, metabolic hormone activity, and immune responses (Suttle, 2010). Without an adequate supply of Zn, swine would begin to exhibit depressed feed intake,

parakeratosis (in growing pigs), loss of skeletal integrity, and reproductive complications. For reproduction, animals deficient in Zn will have lower concentrations of FSH and LH (Boland, 2003), abnormalities or malformations of organ systems (King, 2000), limited fetal growth, and compromised tissue healing after parturition (Lin et al., 2018). Furthermore, sows will produce fewer pigs born alive per litter (Hoekstra et al., 1967) and fewer pigs weaned per litter (Hill et al., 1983).

Without question, Zn is a vital trace element in swine nutrition, but Zn status and biomarkers used to assess Zn utilization in the body are not well defined for swine. Unlike other trace elements such as iron and copper, there is no single storage site for Zn in the body (Lowe et al., 2009), nor is there a single biomarker to determine Zn status (Wood, 2000). Potential biomarkers of Zn include blood plasma, cellular components of blood, hair, fecal and urine excretion, and activity of Zn-dependent enzymes such as alkaline phosphatase and superoxide dismutase (Hambidge, 2003). In humans, plasma Zn is often the most widely used and accepted biomarker.

Researchers have evaluated plasma, serum alkaline phosphatase, serum albumin, metallothionein, colostrum, and milk Zn concentrations as potential biomarkers of Zn status in sows. Van Riet et al. (2015) determined that all biomarkers fluctuated throughout gestation and lactation due to the role of Zn within such phases. Thus, numerous biomarkers should be considered when assessing Zn status of sows. Furthermore, fluctuations in Zn status depends on stage of the reproductive cycle which should be considered when evaluating Zn status of sows (Van Riet et al., 2018).

Uptake and regulation of steady state cellular Zn is controlled through several physiologic systems including absorption, excretion, exchange, and secretion (Wastney et

al., 1986; Roohani et al., 2013). Zinc is absorbed through an influx to the enterocyte and the basolateral membrane, followed by portal circulation transport (Krebs, 2000). Zinc absorption by the small intestine occurs through a non-mediated or mediated process dependent on zinc status and intake (Figure 2.4). Zinc transporter proteins are responsible for elevating or lowering intestinal cellular zinc concentrations (Cousins, 2010). Animals deficient in zinc will exhibit enhanced absorption of zinc, and high dietary intakes of zinc will cause an influx of zinc into the portal system. Newly absorbed zinc then binds to albumin for transportation (Cousins, 1989). Overall homeostasis of Zn status is maintained through a balance between absorption and excretion of Zn. The primary excretion route for zinc is through the feces (Poulsen and Larsen, 1995). Fecal zinc may include unabsorbed dietary zinc and endogenous pancreatic and intestinal secretions of zinc (King, 2000). Pigs typically excrete 70-95% of Zn present in common feedstuffs used in practical swine diets (Kornegay et al., 1997).

Lumen   Enterocyte   Plasma

Paracellular

Dietary Zinc

Transcellular

Zn-Proteins
Zn-Enzymes

Zn-Metallothionein

Enterocyte

Albumin
Albumin Zn

Albumin
Albumin Zn

Tight Junctions
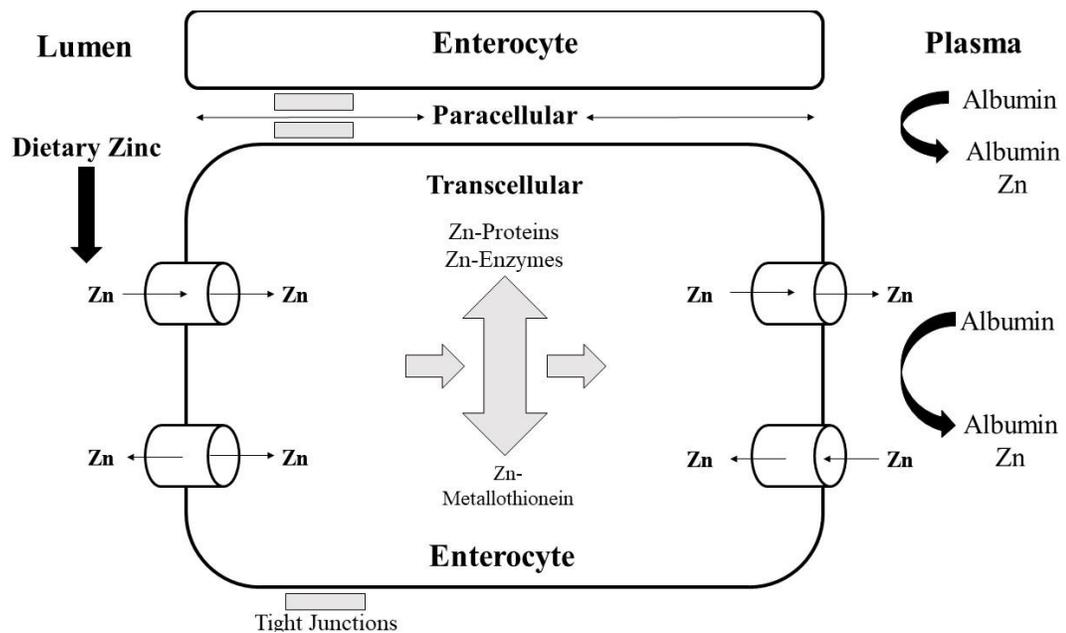
Figure 2.4. Mechanism for intestinal zinc absorption (adapted from Hempe and Cousins, 1992).

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Overall Zn status regulates efficiency of Zn absorption (Steel and Cousins, 1985;

Krebs, 2000). Furthermore, other dietary factors can either positively or negatively

influence absorption and overall bioavailability of Zn. Chelating agents such as phytic

acid (phytate), and interactions with metal ions (Figure 2.5), such as calcium and iron, are

strong antagonists of Zn absorption and bioavailability (Baker and Ammerman, 1995;

Lonnerdal, 2000). Phytate is a major component of plant-based feed ingredients such as

corn and soybeans, and has one of the strongest negative effects on Zn absorption

(O'Dell, 1969, 1989; Cheryan, 1980; Mills, 1985) due to the direct binding of phytic acid

to Zn. Although calcium does not directly bind to Zn, excess calcium may bind with

phytic acid-Zn complexes to form a highly insoluble complex that prevents absorption of

Zn. However, inclusion of phytase enzymes in swine feed can mitigate some of these

negative interactions (Lei et al., 1993; Jongbloed et al., 2004).
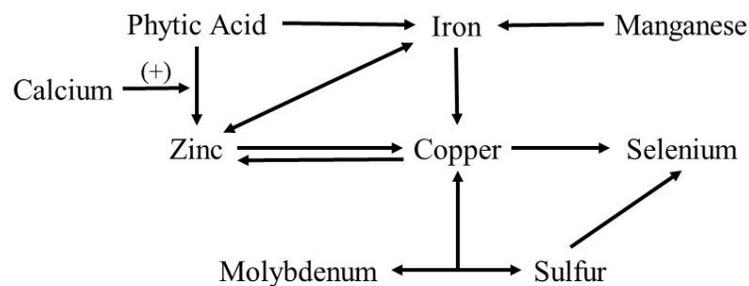
Figure 2.5. Interactions of phytic acid and trace minerals (adapted from O'Dell, 1989)

There are two classes of supplemental Zn sources for swine diets. Inorganic

sources, including sulfate, oxide, or carbonate, are commonly used and are relatively

inexpensive. Inorganic sources are broken down into free ions before absorption in the

gastrointestinal tract, yet, these free ions react to form complexes, sometimes inhibiting

their absorption (Close, 2010). In contrast, organic sources of Zn are typically chelated or

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complexed with amino acids, proteinates, or polysaccharides. Of these sources, amino acid complexes are the most thoroughly understood (Wedekind et al., 1992, Rupic et al., 1997). Organic Zn sources are believed to have greater digestibility, improved uptake, and overall less excretion when compared to inorganic Zn sources in growing-finishing pigs (Lee et al., 2001; Acda and Chae, 2002; Carlson et al., 2004; Liu et al., 2014). This greater digestibility allows nutritionists to provide less of the organic source in a diet while still meeting the animal's nutrient requirements. However, organic Zn sources are generally costly in comparison to inorganic sources. Degree of zinc supplementation among both inorganic and organic zinc sources need to be evaluated further for reproducing sows to optimize performance and control diet cost.

Zinc bioavailability and digestibility

Digestibility can be determined by evaluating absorption of a mineral, however, bioavailability of a mineral is defined as the degree to which an animal can absorb and utilize a mineral (Richards, 2010). There is not a single, direct indicator for a specific mineral's bioavailability. Therefore, numerous indicators should be considered to best determine the absorption and utilization of the mineral (Figure 2.6). True bioavailability of trace elements, such as Zn, must consider both absorption and utilization of the mineral. Assessing Zn at the site of absorption in an animal's small intestine may provide initial guidance to evaluating Zn bioavailability, but this method is usually not the most practical. As a result, other methods are needed to fully assess mineral bioavailability, and may include measuring tissue storage and deposition or considering biomarkers specific to the mineral of interest.
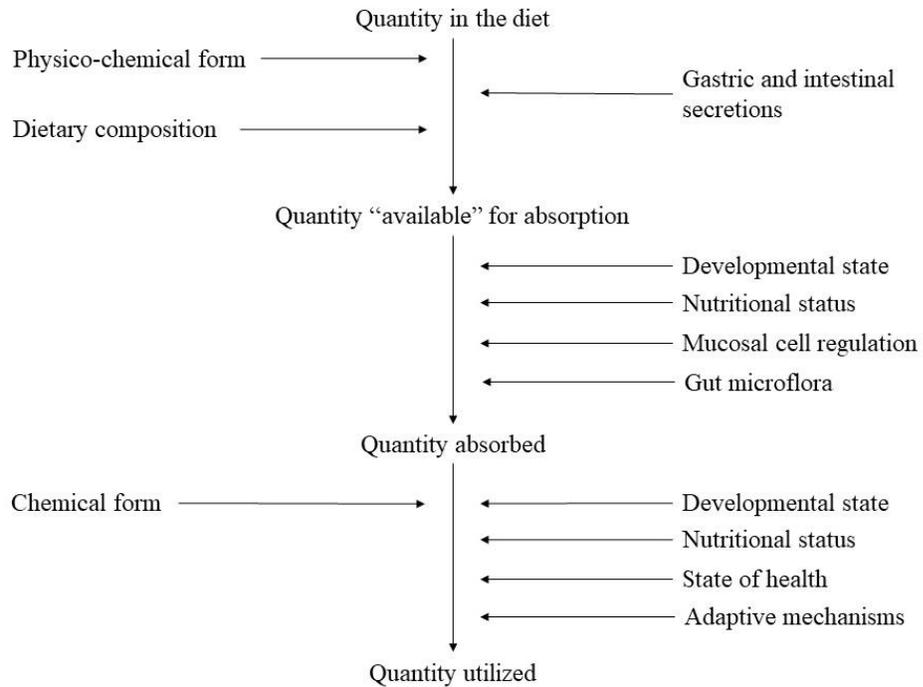
Figure 2.6. Factors to determine bioavailability (Fairweather-Tait, 1992)

Bioavailability among Zn sources has been investigated for nursery and growing-finishing swine and poultry (Nitrayova et al., 2012; Schlegel et al., 2013). Relative bioavailability of commonly used Zn sources is presented in Table 2.3, but, bioavailability estimates among organic and inorganic Zn sources vary (Hill et al., 1986; Wedekind et al., 1992; Wedekind et al., 1994; Close, 2003). Hill et al. (1986) investigated the effects of Zn-methionine, an organic Zn source, and Zn sulfate, an inorganic Zn source, on growth performance, and blood and bone characteristics of growing pigs. Ultimately, there were no differences in performance or bioavailability of Zn among pigs, regardless of the supplemented Zn source. A few years later, Wedekind et al. (1992) determined that organic Zn, fed as Zn-methionine, had greater bioavailability in growing chicks, when compared to the inorganic Zn sources of Zn sulfate and Zn oxide. Wedekind et al. (1994) further investigated effects of organic Zn

sources (Zn-methionine and Zn-lysine) and inorganic Zn sources (Zn sulfate and Zn oxide) on growth performance of and bioavailability for growing-finishing pigs. In agreement with results determined by Hill et al. (1986), growth performance was not affected by dietary Zn source. However, in contrast to the previous study, bioavailability was not equal among sources and was determined to be greatest for Zn sulfate, followed by Zn-methionine, Zn oxide, and Zn-lysine. Interestingly, the rankings of relative bioavailability seem to alternate between inorganic and organic Zn sources. Current dogma suggests the organic Zn sources are more bioavailable than inorganic sources. Star et al. (2012) revisited Zn bioavailability of organic and inorganic sources in broiler chickens. Although growth performance was not affected by Zn source, relative bioavailability, determined by analyzing tibia Zn, was greater for organic Zn supplemented as Zn-amino acid-complex, when compared to Zn sulfate. This observation seems to be consistent with conclusions determined by Wedekind et al. (1992). It appears that there may be some consistency regarding bioavailability of Zn sources in broiler chickens but not for pigs.

Current literature regarding estimates of bioavailability of Zn sources have focused on growing-finishing pigs, with few studies in sows. Some researchers have evaluated reproductive performance and body composition of sows fed inorganic and organic mineral sources at similar dietary concentrations, and have determined that there are minimal differences in farrowing performance, body composition, and piglet growth (Acda and Chae, 2002; Peters and Mahan, 2008; Peters et al., 2010).

Table 2.3. Relative bioavailability of supplemental Zn sources for livestock diets (adapted from Baker and Ammerman, 1995).[1]

Zn Source	Poultry	Swine	Cattle	Sheep
Inorganic				
Oxide	55 (2)	50 (1)	-	-
Sulfate	100	-	100	100
Chloride	100	100	-	-
Carbonate	105 (5)	-	60 (1)	-
Organic				
Lysine	-	100 (1)	-	-
Methionine	125 (3)	100 (2)	-	100 (2)
Proteinate	100 (1)	-	-	-
Chelated	-	-	-	110 (2)

[1]Values expressed as relative bioavailability in response to the standard sources of Zn. Values in parenthesis indicate number of studies or samples involved

Bioavailability of a mineral should not be confused with digestibility of the mineral. Digestibility of an ingredient only considers the effectiveness with which an element is absorbed without considering its post-absorptive utilization. Simple digestibility of a nutrient can be evaluated by measuring the difference between intake and excretion of the nutrient of interest. To better understand digestibility and total losses of a nutrient, a nutrient balance experiment may be conducted. Nutrient balance experiments require extensive documentation and organization. Accurate records of feed intake, fecal output, urinary output, and even milk output for lactating animals must be completed. Therefore, quantifying nutrient excretions or secretions often becomes more complex in reproducing animals due to the additional consideration of milk output.

One must measure intake of the nutrient and the amount of nutrient in the feces to determine the amount of nutrient digested. There are two available methods to do this. Nutrient digestibility can be estimated indirectly by using an index compound, or directly by total collection of feces. The total collection method requires extensive documentation

of individual animal feed intake and total fecal output, together with chemical analysis of the nutrient of interest in feed and feces. However, the index method eliminates the need for extensive record keeping, but relies heavily on accurate chemical analysis of the nutrient and index compound within feed and feces. An index compound, or tracer, is an indigestible, non-toxic, non-absorbable, non-essential, uniformly mixed, measurable substance that flows at the same rate as the rest of the digesta through the gastrointestinal tract and is completely voided in the feces (Adeola, 2001). Commonly used tracers include chromic oxide, acid insoluble ash, and titanium dioxide at 0.1 to 0.5% of the diet. By utilizing an indigestible marker, animals may not have to be placed in metabolism crates for total feces collection, but digestibility can still be evaluated. Digestibility can be calculated using the index method equation shown in Figure 2.7.

$$\text{Digestibility, } \% = 100 - \left[100 \times \left(\frac{\text{Concentration of marker in feed} \times \text{Concentration of nutrient in feces}}{\text{Concentration of marker in feces} \times \text{Concentration of nutrient in feed}} \right) \right]$$

Figure 2.7. Calculation of nutrient digestibility using the index method (Adeola, 2001).

Various measures of digestibility may be evaluated to attain accuracy regarding overall digestibility of a nutrient to an animal (Peterson and Stein, 2006; Stein et al., 2007). Apparent total tract digestibility (ATTD), true total tract digestibility (TTTD), and standardized total tract digestibility (STTD) are all acceptable references to digestibility of a nutrient, yet each reference differs in the level of precision. Apparent total tract digestibility represents the net disappearance of a nutrient from the digestive tract, and

therefore, includes endogenous losses that may be secreted or sloughed off in the gastrointestinal tract. True total tract digestibility is similar to ATTD, but corrects for the endogenous losses, therefore representing the total disappearance of a dietary nutrient from the gastrointestinal tract. True total tract digestibility is often more difficult to measure due to the extensive labor required to evaluate endogenous losses of the nutrient of interest, which may be even more difficult for trace elements. Standardized total tract digestibility is the most commonly used measure when formulating livestock diets and can be calculated by correcting for basal endogenous losses of a nutrient. Measuring basal losses of a nutrient by feeding a diet completely free from that nutrient of interest is often recommended. Digestibility of many macro nutrients have been extensively researched, allowing potential for feed formulation on all levels of digestibility (ATTD, TTTD, and STTD). However, this has not been extensively investigated for trace minerals.

Chemical zinc analysis

Indirect methods to assess nutrient digestibility rely heavily on accurate lab analysis of the nutrient of interest. With respect to Zn, a trace element, this is critical. Two analytical techniques, inductively coupled plasma atomic emission spectrometry (ICP-AES) or atomic absorption spectroscopy (AAS), are commonly used to determine Zn concentrations of biological samples (Dipietro et al., 1988). Atomic absorption spectroscopy is generally cheaper, but analysis of only a single element may be conducted at a time. On the other hand, ICP-AES is more expensive, but allows for analysis of multiple elements simultaneously. Although these methods are well established, variation of Zn analysis within complete feed samples can be challenging.

Furthermore, analytical variation among laboratories must be considered. Members within the swine nutrition industry commonly collect samples of finished feeds to ensure diet formulation and nutrient composition expectations are met (Goncalves et al., 2016). Therefore, it is crucial to collect a representative feed sample that minimizes analytical variation and provides confident nutrient composition results. Variation of Zn concentration in sampled feed was not affected by the method of sample collection when probe and hand grab methods were compared in either sample collection method, but, analyzed Zn concentration variation of the diet was very high, often ranging around 50 to 75 ppm from the expected Zn concentration. Because of this high analytical variation, Jones et al. (2017) estimated that at least 34 feed samples would need to be analyzed to have 95% confidence that the analyzed Zn concentrations of a diet are within 4 ppm of the actual value. However, this practice would be very costly and may not be realistic. Therefore, to achieve accurate sample analysis results within 15 ppm, 2 to 5 feed samples must be analyzed. Overall, variation of Zn analysis in complete feed samples should be expected, but when very high variation occurs, accurate dietary Zn concentrations cannot be concluded. However, with multiple feed sample analysis, accurate Zn concentrations may be determined.

Zinc Supplementation in Sow Diets

Dietary zinc requirements for gestating and lactating sows

Much of the research conducted to determine the sow's daily Zn requirement published in the NRC (2012) is based on experimental trials conducted in the 1980's and earlier (Table 2.4). As previously indicated, productivity of modern genetic lines has improved dramatically over the last 40 years. Since the 1980's, there have not been any

experiments to fully re-evaluate daily zinc requirements for sows. Relative to most ingredients in a typical swine diet, Zn tends to be low in cost. Therefore, Zn is typically supplemented at concentrations above the requirement to allow a "safety margin" that compensates for the uncertainty of requirements for modern genetic lines (Brugger and Windisch, 2017). The observation that signs of Zn deficiency are rare in commercial production suggests that common diets are above the minimum Zn requirement. Yet, over-supplementing Zn may lead to excess excretion, further exacerbating environmental concerns. Conducting an experiment with a large population of sows to fully evaluate the daily zinc requirement of current genetic lines would be costly, but could also lead to enhanced sow performance and/or reduced environmental impacts.

Table 2.4. Zinc requirements for gestating and lactating sows (adapted from NRC, 1988; 1998; 2012)

	Gestating	Lactating
NRC, 1988		
mg/d	95.0	265.0
mg/kg of diet	50	50
NRC, 1998		
mg/day	93.0	263.0
mg/kg of diet	50	50
NRC, 2012		
mg/day	210.0	596.6
mg/kg of diet	100	100

Effects of zinc supplementation on sow productivity and piglet performance

As previously mentioned, fetal growth and development is completely reliant on the dam. By providing an essential nutrient at critical developmental phases, fetal imprinting occurs (Ji et al., 2017). In circumstances where fetal crowding may occur, preparing offspring for survival through nutritional fetal imprinting strategies may be

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essential. Because Zn is an important, versatile nutrient, supplemental dietary Zn in gestation may reduce the incidence of stillborn piglets (Hill et al., 1983; Vallet et al., 2014), increase litter weaning weights (Payne et al., 2006), and even reduce pre-weaning mortality of low birth weight pigs (Vallet et al., 2014). Scientists hypothesized that the observed improvement in survival of small piglets may be due to Zn's diverse roles in antioxidant activity, protease activity, transcription, and carbonic anhydrase (Vallet et al., 2014). However, further investigation of Zn's role in these biological processes and the potentially positive effects of Zn in fetal imprinting are necessary to fully understand such responses.

Zinc is essential for maintaining a desired level of sow and piglet performance. However, the level of zinc supplementation may heavily affect long-term productivity. In an early investigation reported by Hedges et al. (1976), sows fed corn-soybean meal diets supplemented with 33 or 83 mg/kg over five parities did not exhibit any signs of zinc deficiency or any detrimental effects on farrowing performance. However, overall piglet growth performance was maximized for pigs farrowed by sows receiving the higher level of dietary zinc. A later study evaluated performance of sows fed diets containing 0, 50, 500, or 5,000 mg/kg supplemental Zn for two parities (Hill et al., 1983). Sows fed diets containing 5,000 mg/kg of zinc experienced greater incidence of osteochondrosis, fewer piglets weaned, and significantly smaller piglets at weaning than sows fed diets containing 0, 50, or 500 mg/kg of Zn. This clearly indicates that extremely high zinc supplementation is not necessary and negatively affects reproductive performance. Sows receiving any of the other dietary treatments (0, 50, or 500 mg/kg zinc) did not exhibit differences in the number of piglets born, born alive, or birth weights among piglets. Yet,

sows receiving no supplemental zinc had a greater number of abnormal pigs per litter. Therefore, dietary Zn supplementation between 50 and 500 mg/kg elicited comparable reproductive performance among sows. However, another trial involving only 12 sows evaluated gestation diets with no supplemental zinc and determined there were no differences in farrowing performance compared to sows receiving 50 mg/kg supplemental Zn (Kalinowski and Chavez, 1984). Although these experiments investigated various dietary Zn concentrations, few conclusions regarding optimal Zn supplementation can be determined, due to the inconsistent results among experiments.

Recently, Van Riet et al. (2018) conducted an experiment to evaluate performance of sows (n = 131) when fed dietary Zn concentrations of 0, 50, and 100 mg/kg. Despite obvious advancements in sow performance in the last 40 years, sows still exhibited no differences in the number of piglets born alive or birth weight of piglets across treatments. The inability to distinguish differences in performance among a range of dietary Zn concentrations mirrors results previously reported by Hill et al. (1983). Regardless of the lack of performance differences among ranges of dietary Zn concentrations in Van Riet et al. (2018), further investigation with a large population of modern sows is necessary to determine optimal Zn supplementation for lifetime sow and piglet performance.

Supplemental zinc in sow diets

We cannot assume that determined digestibility of Zn sources in growing-finishing pigs will be equal to that of reproducing females. Digestion and absorption of nutrients occurs in the small intestine and hindgut of the animal. Although pigs may have a fully developed small intestine at a bodyweight of about 20 kg, the hindgut of the

animal continues to develop after 150 kg body weight (Nielsen, 1962). Often, commercial pigs are marketed at or before 150 kg of bodyweight. Therefore, it is likely that reproducing sows, with body weights exceeding 150 kg, will have a more developed hindgut than finishing pigs which may alter digestibility of nutrients from what is reported for growing-finishing pigs. Digestibility values obtained from experiments with ileal cannulations in the small intestine of pigs may still be transferable to pigs with heavier body weights, such as sows (Furuya and Takahashi, 1980). However, digestibility values obtained from fecal digestibility experiments, which are much less labor intensive and more commonly used, must therefore consider body size of the animal (Shi and Noblet, 1994). Often, sows have superior digestibility of nutrients from commonly-used feedstuffs when compared to growing-finishing pigs (Fernandez et al., 1986). Feed offerings and rate of feed intake must also be considered when evaluating digestibility. Unlike growing-finishing pigs allowed *ad libitum* access to feed, gestating sows typically are offered feed only once daily. Availability of feed and nutrient intake may play a role in digestibility and utilization of those nutrients. When considering such impacts on digestibility, scientists confirmed that sows fed at maintenance levels had improved digestibility of energy and nutrients when compared to growing pigs fed ad libitum (Noblet and Shi, 1993). Nonetheless, extensive investigation of micronutrient digestibility in reproducing sows has not been completed. As a result, nutritionists cannot assume that reported digestibility values for Zn sources in young pigs will be exactly equal to that of gestating or lactating sows.

Summary

Zinc is a crucial component of diets for gestating and lactating sows and plays an essential role in proper growth and development of piglets. Although many zinc sources are available for supplementation in swine diets, digestibility and bioavailability for sources of zinc among sow diets have not been researched extensively. As a result, values for digestibility and bioavailability of zinc sources in growing-finishing swine diets are often assumed to be equal in diets for reproducing swine. Without full understanding of Zn utilization for gestating and lactating sows, nutritionists cannot accurately formulate diets to meet the animal's daily zinc requirement. Therefore, determining digestibility of both organic and inorganic zinc sources in practical diets for gestating and lactating sows is essential so that producers can both optimize sow performance and minimize costs. The experiment presented in Chapter 3 of this thesis investigates the digestibility of two zinc sources in sow diets.

Furthermore, as genetic lines continue to produce larger litters, a greater emphasis will be placed on survival and performance of piglets to increase the number of piglets weaned per litter. Such large litters often result in variability of birth weights, and increased number of low birth weight piglets. Until this variation can be reduced, methods to improve survivability of small piglets needs to be investigated. The experiment in Chapter 4 investigates the effects of supplemental zinc in late-gestation on pre-weaning survival of low birth weight pigs.

CHAPTER 3

Comparative digestibility of polysaccharide-complexed zinc and inorganic zinc in diets for gestating and lactating sows

SUMMARY

The primary hypothesis tested in this experiment is that digestibility of an organic zinc source from polysaccharide-complexed zinc is greater than inorganic zinc sulfate when sows consume high fiber diets containing corn dried distillers grains with solubles (DDGS). Gilts and sows (n = 32) were blocked according to parity and assigned randomly to one of four dietary treatments. Eight sows were assigned to each treatment in four replicate blocks. Dietary treatments consisted of: 1) Control (**ConZnSO$_4$**) – corn-soybean meal based diet + 100 ppm supplemental Zn from $ZnSO_4$; 2) Control PSZn (**ConPSZn**) – corn-soybean meal based diet + 100 ppm supplemental Zn from polysaccharide-complexed Zn; 3) **DDGS/ZnSO$_4$** – corn-soybean meal-40% DDGS gestation diet and a 30% DDGS lactation diet, with each containing 100 ppm supplemental Zn from $ZnSO_4$; 4) **DDGS/PSZn** – corn-soybean meal-40% DDGS gestation diet and a 30% DDGS lactation diet, with each containing 100 ppm supplemental Zn from polysaccharide-complexed Zn . A fifth dietary treatment was imposed using a subset of sows (n = 20) to determine basal Zn losses in gestating and lactating sows fed corn and soybean meal based diets containing no supplemental Zn. Nutrient balance experiments were conducted in both gestation and lactation to evaluate digestibility of Zn sources of the four dietary treatments, and to determine basal Zn losses during gestation and lactation when no supplemental dietary Zn was provided. The statistical model consisted of fixed effects of diet, Zn source, and their interaction, and

random effects of parity. Sows fed DDGS/ZnSO$_4$ had a greater ($P < 0.05$) number of pigs weaned per litter (13.4) compared with those fed ConZnSO$_4$ (10.8), ConPSZn (10.4), and DDGS/PSZn (11.9), resulting in decreased average piglet weaning weights (5.6 vs. 7.1, 6.3, 6.7 kg, respectively). These slight differences observed at weaning are not likely associated with dietary treatment, but rather due to numerical differences in piglets born per litter. Furthermore, overall piglet and litter gain among treatments was not different ($P > 0.05$). Estimated endogenous losses of Zn were used to adjust apparent total tract digestibility (ATTD) to true total tract digestibility (TTTD) of Zn in the four dietary treatment balance periods. There were no differences in Zn concentrations of urine, plasma, colostrum, or milk samples among treatments at any time of the experiment ($P > 0.05$). Gestating sows fed DDGS/PSZn exhibited improved ($P < 0.05$) ATTD, TTTD, and overall retention of Zn in comparison to both Control treatments, with the DDGS/ZnSO$_4$ treatment responses being intermediate. Lactating sows consuming diets without DDGS and supplemented with organic Zn exhibited the highest ($P < 0.05$) ATTD, TTTD, and retention of Zn, which were opposite to responses observed in gestation. Furthermore, ATTD, TTTD, and Zn retention for lactating sows consuming DDGS/PSZn were lower ($P < 0.05$) than all other treatments. Overall, it appears that stage of pregnancy and dietary fiber affect digestibility and retention of Zn, regardless of Zn source.

Key words: digestibility, gestation, lactation, sows, zinc source

INTRODUCTION

The zinc requirement of reproducing sows is not well established, but it is

estimated to be greater than that of growing pigs due to Zn needs for fetal growth, milk

synthesis, and repair of tissue damage during uterine involution. The current Zn

requirements, expressed on a diet concentration basis, for gestating and lactating sows are

100 mg/kg of diet (NRC, 2012). The Zn requirements, expressed on a daily basis, are 210

mg/d for gestating sows and 597 mg/d for lactating sows (NRC, 2012). However, the last

studies to determine dietary Zn requirements of sows were conducted about 40 years ago

(Hedges et al., 1976; Kirchgessner et al., 1981) with genetic lines that were vastly

different from those present in the commercial industry today. Very few studies have

been conducted since then to determine if these requirements are still adequate for

modern sows that produce larger, heavier litters (Stalder, 2018) under commercial

conditions.

Industry nutritionists have dramatically increased the safety margin above the

current requirement due to uncertainty of actual Zn requirements (Brugger and Windisch,

2017). This practice increases diet cost and leads to increased Zn excretion in manure

which can have negative environmental consequences over time. Excessive accumulation

of Zn in manure can be detrimental to the quality of soil and water (Jongbloed and Lenis,

1998). One of the goals in commercial pork production is to efficiently utilize dietary Zn

to minimize excretion while optimizing animal health and performance to achieve a

sustainable production system.

Changes in feed ingredients used in swine diets during the last two decades

further exacerbate the uncertainty regarding dietary Zn utilization. Often, producers

utilize grain co-products to minimize feed costs and meet nutrient requirements but some of these co-products, such as dried distillers grains with solubles (**DDGS**) or wheat middlings, may reduce Zn digestibility. Phytic acid or phytate, a major component of commonly used plant-based feed ingredients such as corn and soybean meal, strongly inhibits Zn absorption and decreases bioavailability (O'Dell, 1969; Cheryan, 1980; Mills, 1985; O'Dell, 1989; Lonnerdal, 2000). However, the use of phytase enzymes in diets can be effective in mitigating some of these negative interactions (Lei et al., 1993; Jongbloed et al., 2004). Other dietary components such as fiber, Ca, Cu, and protein source may also act as antagonists that negatively influence Zn utilization (Solomons, 1983; Baker and Ammerman, 1995; Solomons 2001). Ultimately, thorough investigation of Zn digestibility and absorption in modern sows must consider these antagonistic dietary factors.

Composition of supplemental Zn sources may also influence digestibility and absorption of Zn for swine. Inorganic Zn sources are digested into free ions before absorption in the gastrointestinal tract, but these free ions may react with other dietary components such as phytate, fiber, Ca, or Cu to form indigestible complexes (Close, 2010). Organic sources of Zn, such as amino acid and carbohydrate complexes, are absorbed via peptide or amino acid transport systems, which often increase absorption and decrease fecal Zn excretion (Nitrayova et al., 2012). However, the mechanism of absorption of a polysaccharide-complexed Zn source has not yet been determined. Previous investigation of Zn utilization in nursery and growing-finishing pigs have shown that organic Zn sources have greater digestibility, improved absorption, and decreased excretion when compared to inorganic Zn sources (Lee et al., 2001; Acda and Chae, 2002; Carlson et al., 2004; Liu

et al., 2014). Currently, no studies have been published regarding the nutritional value of organic polysaccharide-complexed Zn compared with an inorganic Zn source in diets for gestating and lactating sows.

Therefore, the objectives of this study were to determine and compare excretion, retention, apparent total tract digestibility (ATTD) and true total tract digestibility (TTTD) of polysaccharide-complexed Zn and Zn sulfate ($ZnSO_4$) in sow gestation and lactation diets with and without dietary antagonists. This objective tested the hypothesis that polysaccharide-complexed Zn will have greater digestibility and retention and less Zn excretion when compared to $ZnSO_4$. A second objective was to determine estimates of basal daily Zn losses of sows fed diets without supplemental Zn.

MATERIALS and METHODS

This experiment was conducted at the University of Minnesota's West Central Research and Outreach Center in Morris, Minnesota. The experimental protocol was reviewed and approved by the University of Minnesota Institutional Animal Care and Use Committee (IACUC# 1706-34924A). The experiment began in October, 2017 and concluded in December, 2017.

Animals, Housing, and Treatments

The experiment was conducted in two similar rooms in the same facility where each room contained 16 farrowing stalls. Sows were housed individually in farrowing stalls (1.52 x 2.13 m) from about d 75 of gestation through lactation and weaning of litters. Farrowing stalls were equipped with one stainless steel dry feeder and one nipple waterer on a partially slatted floor over a deep (2.4 m) manure collection pit. All heaters and ventilation fans were operated by an independent controller in each room that

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maintained room temperature between 15.5 and 18°C during gestation and lactation, and 21°C during farrowing. One heat lamp was placed in the creep area of each farrowing stall as a supplemental heat source for piglets during lactation.

On about d 75 of gestation, 32 sows (Topigs Norsvin, Burnsville, MN) were blocked according to parity and assigned within blocks randomly to one of four dietary treatments in a 2 x 2 factorial arrangement. Eight sows were assigned to each treatment and remained on their assigned dietary treatment over the entire experimental period, except during the low Zn balance feeding and collection period.

Dietary treatments consisted of: 1) Control – corn-soybean meal based diet + 100 ppm supplemental Zn from $ZnSO_4$ (**ConZnSO$_4$**); 2) Control polysaccharide Zn – corn-soybean meal based diet + 100 ppm supplemental Zn from polysaccharide-complexed Zn (**ConPSZn**); 3) High fiber control – corn-soybean meal-40% DDGS gestation diet and a 30% DDGS lactation diet, with each containing 100 ppm supplemental Zn from $ZnSO_4$ (**DDGS/ZnSO$_4$**); and 4) High fiber polysaccharide Zn – corn-soybean meal-40% DDGS gestation diet and a 30% DDGS lactation diet, with each containing 100 ppm supplemental Zn from polysaccharide-complexed Zn (**DDGS/PSZn**). Polysaccharide-complexed Zn was supplemented to dietary treatments using SQM Zinc (QualiTech, Chaska, MN). Corn DDGS (Glacial Lakes Energy LLC, Watertown, SD) with low vomitoxin concentration (< 1.2 ppm) was utilized in experimental diets. A fifth dietary treatment (**LowZn**) was imposed using a subset of sows (n = 20) to determine baseline Zn losses in gestating and lactating sows fed corn-soybean meal based diets containing no supplemental Zn.

Sows remained on their dietary treatments through the completion of the gestation and lactation balance periods (Figure 3.1). Immediately following gestation and lactation balance periods, 20 sows were randomly selected among those that did not exhibit any signs of infection from previous urinary catheter placement, and assigned to the LowZn dietary treatment before returning to their originally assigned treatment.

D 75 Gestation • Initiation of dietary treatments

D 85-87 • Gestation balance • n = 8 sows/diet

D 88-92 • Gestation LowZn adaptation period • n = 20 sows

D 93-95 • Gestation LowZn balance

D 96-109 • Sows return to initial dietary treatments

D 110 • Initiation of lactation diets • n = 29 sows

D 112-114 • Farrowing

D 7-9 Lactation • Lactation balance • n = 6 to 9 sows/diet

D 10-14 • Lactation LowZn adaptation period • n = 20 sows

D 15-17 • Lactation LowZn balance

D 21 • Weaning

Figure 3.1. Experimental timeline

Experimental diets were formulated to represent diets typically used in the commercial swine industry based on corn, soybean meal, and DDGS (Tables 3.1 and 3.2). Previous production data from sows in this herd were used in the NRC (2012) model to estimate nutrient requirements. The model output was then used to formulate diets that met or exceeded nutrient requirements during gestation and lactation, except for Zn. All diets were mixed in the same feed mill on-site and were offered to sows in meal form.

Sow and Piglet Performance

Sows were identified individually using ear tags and weighed at initiation of the experiment, within 24 h of expected farrowing date, within 24 h after parturition, and at weaning. Daily feed intake and instances of ill health were monitored and recorded daily. Sows were limit-fed 2.2 kg of feed once daily during gestation. Immediately following parturition and throughout lactation, sows were offered an increasing amount of feed twice daily to achieve *ad libitum* intake by d 4 of lactation. Instances of feed wastage were monitored and recorded if necessary. Sows were allowed *ad libitum* access to water throughout the experiment, which was supplied by one nipple drinker in each farrowing stall.

Measurements of sow performance at farrowing included: total number of piglets born, born alive, stillborn, mummified, and weaned per litter. Litter sizes were standardized as close to 11 piglets per sow as possible by cross-fostering piglets within treatment group within 24 h of birth. Piglets were processed according to the standard operating procedures of the farm and included administering injectable iron, needle teeth clipping, tail docking, and castration of males within 24 h of birth. All piglets were individually ear-tagged and weighed within 24 h of birth and again at weaning to determine total body weight gain during the nursing period. Piglets were weaned at 20.2 ± 1.6 d of age.

Zinc Balance During Gestation and Lactation

Two separate nutrient balance experiments were conducted on about d 88 and d 96 of gestation to evaluate digestibility of dietary Zn among treatments. Sows were fed

their respective diet containing 0.50% titanium dioxide. Sows were housed in individual stalls for sample collection and were allowed 7 days to adapt to their assigned diet before each 3-d collection period. Upon completion of the nutrient balance period, 5 sows per initial treatment (n = 20 total) were switched to the LowZn dietary treatment to undergo another balance period (about d 96 of gestation). Sows selected to receive the LowZn diet were allowed a 5-d acclimation period before a second 3-d collection period. During lactation, nutrient balance experiments were conducted at d 7 to 10 postpartum, and again on d 15 to 17 (LowZn balance) of lactation.

Fecal samples were collected once daily at 1500 h, placed in plastic bags, and stored separately at -20°C for all balance periods. At the end of each collection period, fecal samples were pooled by sow within balance period and weighed. To determine moisture content, samples were then dried in a forced draft oven at 55°C and weighed once daily for about 3 d or until samples maintained a constant weight. Once dried, feces were ground through a 1-mm screen and stored in Whirl-pak® bags until subsequent Zn and titanium analysis.

Urinary catheters were inserted into the urethra of each sow one day before each nutrient balance period. To begin this process, the entire vulva area of the sow was washed with an antiseptic (betadine). Next, sterile urinary catheters (Lubricath, 2-way, 30 mL balloon, 18 French; Bard Medical Canada Inc., Oakville, ON, Canada) were lubricated with a sterile lubricant and inserted flaccidly into the urethra. The lubricant was laced with lidocaine to prevent spasms in the urethra upon placement. While the sow was standing, the tip of the catheter was guided along the floor of the vagina by a technician's finger until it entered the urethra. The technician used sterile gloves for this

procedure. Once the tip was fully inserted into the urethra, the balloon was inflated with 30 mL of saline solution to retain the catheter in the bladder. Catheters were connected to polyvinyl tubing that dispensed urine into a closed vessel, allowing the sow to urinate as necessary. Urine was collected in a vessel that contained 20 mL of sulfuric acid to maintain urine pH $\leq$ 5. The use of sulfuric acid preserved the urinary nitrogen, prevented microbial contamination of the urine sample, and subsequently minimized the occurrence of ascending urinary tract infections. Catheters remained in place for 48 h and urine collection vessels were emptied and sub-sampled at 5% aliquots as necessary. At the end of the collection period, the inflatable cuff was deflated and the catheter was extracted from the bladder. Two urine samples (50 mL each) from each sow during each balance period were frozen and stored at -20°C for later Zn analysis. Sow body temperature, behavior, and occurrence of vaginal discharge were evaluated twice daily within each balance period to monitor for urinary tract infections. Any urine collections with instances of catheter complications such as infection or misplacement were not sampled or stored for later analysis.

Blood samples were collected on d 3 of each balance period via jugular venipuncture into heparinized Vacutainer™ tubes. Blood samples were placed immediately on ice after collection and then centrifuged at 1,400 x g to obtain plasma. Plasma samples were frozen and stored at -80°C for later analysis.

Colostrum samples (50 mL) were collected within 12 h of parturition from all functional teats by hand stripping teats and attempting to collect equal quantities of colostrum from each teat to obtain a representative sample. Milk was collected on the final day of both lactation balance periods by administering 10 IU of oxytocin IM to each

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sow before using a collection process similar to that described for colostrum. Immediately after collection, all colostrum and milk samples were frozen and stored at -20°C.

Digestibility Determinations and Milk Output

Values for apparent total tract digestibility (ATTD), true total tract digestibility (TTTD), and basal endogenous losses of Zn were calculated using the equations described by Stein et al. (2007). The equations were as follows:

$$\text{ATTD, \%} = [1 - (Zn_{feces}/Zn_{diet}) \times (M_{diet}/M_{feces})] \times 100;$$

$$Zn_{end} = Zn_{feces} \times (M_{diet}/M_{feces}); \text{ and}$$

$$\text{TTTD, \%} = \text{ATTD} + [(\text{total } Zn_{end}/Zn_{diet}) \times 100];$$

where Zn_{feces} represents fecal Zn concentration in mg/kg DM, Zn_{Diet} represents dietary Zn concentration in mg/kg DM, M_{diet} represents dietary indigestible marker (titanium) in mg/kg DM, M_{feces} represents fecal indigestible marker (titanium) in mg/kg DM, and Zn_{end} represents estimated endogenous Zn losses.

Milk energy output was estimated using an equation from NRC (2012):

$$\text{Milk energy output, kcal/d} = (4.92 \times \text{litter gain, g/d}) - (90 \times \text{number of pigs})$$

Estimated energy density of milk (Hurley, 2015) was used to estimate daily milk output. Energy density of milk used in calculations corresponded to the stage of lactation during which the balance experiments were conducted.

$$\text{Milk energy, kcal/g} = 1.29 \text{ or } 1.17$$

$$\text{Milk output, g/d} = \text{Milk energy output/Milk energy}$$

Sample Analysis

About 2 kg of each dietary treatment were collected at mixing and stored in a freezer at -20°C. Two randomly selected samples from each phase and treatment were sent to the University of Missouri Agricultural Experiment Station Chemical Laboratories (Columbia, MO) for proximate analysis and determination of Zn concentration. Feces, urine, plasma, colostrum, and milk samples were also analyzed for zinc concentration. Standard procedures (AOAC International, 2006) were followed for analysis of moisture (method 934.01), ash (method 942.05), EE (method 920.39), CF (method 978.10), CP (method 990.03) and zinc (method 985.01) concentrations. Dietary treatment and fecal samples were analyzed for titanium concentration according to procedures described by Myers et al. (2004).

Statistical Analysis

Data were evaluated for the presence of outliers, normal distribution, and heterogeneous variance among treatments. Outliers were deemed as any value greater than or less than two standard deviations from the mean, and were removed from the final analysis to achieve normal distribution of data and equal variances among treatments. Experimental data were then analyzed using the PROC GLIMMIX procedure of SAS (Version 9.4, SAS Institute Inc., Cary, NC) with a Gaussian distribution. Sow was considered the experimental unit. The statistical model included fixed effects of diet, Zn source, and their interaction, and random effects of parity. Treatment means were separated using the PDIFF option with the Tukey-Kramer adjustment for multiple comparisons.

Chi square analysis was used to determine the influence of treatments on categorical response variables such as piglet mortality over the entire experiment. All data are reported as least square means and considered statistically significant at $P < 0.05$ with $P < 0.10$ considered a trend.

RESULTS

Digestibility of Zn during Gestation

Sows consumed an identical amount of feed during the gestation balance period, regardless of dietary treatment (Table 3.3). Estimated daily fecal excretion of dry matter was greater ($P < 0.05$) for sows consuming diets containing DDGS with high fiber content compared with that of sows consuming corn soybean-meal based diets because of lower overall DM digestibility. Although daily fecal excretion increased ($P < 0.05$) for sows consuming DDGS diets, daily fecal Zn and Ti excretions decreased ($P < 0.05$), compared with sows consuming Control diets. Furthermore, there was an interaction ($P < 0.05$) of diet and Zn effects on concentrations of fecal Zn, where DDGS/PSZn was lower than all other treatments ($P < 0.05$). Consequently, ATTD, TTTD, and retention of Zn was greatest ($P < 0.05$) for DDGS/PSZn in comparison to both Control treatments, with DDGS/ZnSO$_4$ being intermediate. Therefore, despite lower dry matter digestibility for diets containing DDGS, ATTD and TTTD of Zn improved ($P < 0.05$) for sows consuming diets with high dietary fiber versus conventional corn and soybean meal diets. Furthermore, supplemental PSZn improved ($P < 0.05$) digestibility of Zn in gestating sows fed DDGS diets that contain higher concentrations of dietary fiber, but not in diets based on corn and soybean meal. Improved digestibility of Zn for diets with DDGS and PSZn supported reduced ($P < 0.05$) fecal Zn excretion and ultimately, improved Zn

retention, when compared to Control or $ZnSO_4$ diets. Additionally, total Zn excretion was greater ($P < 0.05$) for Control diets, resulting in improved Zn retention for sows fed DDGS diets. Overall diet and Zn source interactions affected Zn retention so that PSZn supplemented to DDGS diets allowed positive Zn retention in sows, unlike all other dietary treatments.

Digestibility of Zn in Lactation

Before the initiation of feeding lactation diets, a few sows were removed from the experiment that were confirmed not pregnant. The distribution of sows removed from the lactation balance trial were as follows: ConZnSO$_4$ = 2 of 8; ConPSZn = 0 of 8; DDGS/ZnSO$_4$ = 2 of 8; DDGS/PSZn = 0 of 8. An additional gilt that was not included in the gestation balance trials, but received the DDGS/PSZn treatment during gestation was added to the lactation balance trial. Therefore, the distribution of sows per treatment in the lactation balance trial were as follows: ConZnSO$_4$, n = 6; ConPSZn, n = 8; DDGS/ZnSO$_4$, n = 6; and DDGS/PSZn, n = 9.

Immediately after farrowing, sows were allowed an amount of feed that they would completely consume each day to best represent *ad libitum* feeding. However, there were differences in average daily feed intake (ADFI) among treatments (Table 3.4). Sows assigned to DDGS treatments consumed less ($P < 0.05$) feed compared with sows assigned to Control treatments. Consequently, sows consuming Control diets consumed a greater amount of daily Zn ($P < 0.05$) and tended to consume greater amounts of Ti ($P < 0.10$). Calculated daily fecal excretion of DM was not different across treatments. As expected and similar to responses in gestation, dry matter digestibility was reduced by DDGS inclusion in diets, resulting in greater ($P < 0.05$) DM digestibility for sows

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consuming corn and soybean-meal lactation diets. In addition, sows consuming DDGS

diets with high dietary fiber excreted less Ti ($P < 0.05$) and tended to excrete less Zn ($P <$

0.10), than lactation diets based on corn and soybean meal, which were similar to

gestation responses. Diet and Zn source interactions significantly affected ATTD, TTTD,

excretion, and overall retention of Zn ($P < 0.01$). Sows consuming diets without DDGS

and supplemented with organic Zn exhibited the highest ATTD, TTTD, and retention of

Zn, which was opposite to responses observed during gestation. However, when PSZn

was supplemented to diets with DDGS, ATTD, TTTD, and Zn retention were less ($P <$

0.05) than all other treatments. Despite slight differences in feed intake of sows in

lactation, there were no differences in estimated milk yield.

Sow Health and Performance

There were no differences in parity or bodyweight among sows fed any of the

four dietary treatments at initiation of the experiment (Table 3.5). Furthermore, no major

health challenges were experienced by sows or pigs assigned to this experiment. Pre-

weaning piglet mortality throughout the experiment was 12.5%, with stillborn piglets

representing the clear majority of this mortality. Distribution of stillborn piglets in

relation to the number of pigs born within each treatment were as follows: ConZnSO$_4$, 15

of 84 (17.8%); ConPSZn, 15 of 113 (13.3%); DDGS/ZnSO$_4$, 4 of 97 (4.1%); and

DDGS/PSZn, 8 of 137 (5.8%) (Chi-squared = 13.75; df = 3; $P = < 0.01$). The number of

piglet deaths prior to weaning was not different among treatments (Chi-squared 0.41; df =

3; $P = 0.94$) and was distributed within each treatment as follows: ConZnSO$_4$, 3 of 84

(3.6%); ConPSZn, 3 of 113 (2.6%); DDGS/ZnSO$_4$, 3 of 97 (3.1%); and DDGS/PSZn, 3

of 137 (2.2%).

Sows were monitored daily for elevated body temperatures by evaluating and recording rectal temperatures after farrowing and twice daily within balance periods. Some sows did experience transient fevers, often for 1 to 3 days, and consumed little or no feed, which was likely due to mild urinary tract infections from catheter placements. These sows (n = 2) were removed from the lactation LowZn balance period.

There were no significant differences among treatments in the total number of pigs born or number of pigs born alive per litter, but there were slight differences in piglet birth weight among treatments. However, it is not likely that these slight differences were associated with dietary treatment, but rather were due to numerical differences in the number of piglets born per litter. Sows assigned to the DDGS/ZnSO$_4$ treatment produced piglets with lower ($P < 0.05$) weaning weights than sows assigned to all other treatments, but there were no overall differences in piglet and litter daily gain among treatments.

Zinc Biomarkers

Composition and Zn concentrations of colostrum and milk were not different ($P > 0.05$) among diet, Zn source, or their interactions at any time of the experiment (Table 3.6). Similarly, there were no differences ($P > 0.05$) in Zn concentrations of plasma among dietary treatments at any time of the experiment (Table 3.7).

Baseline Zn Losses

When gestating sows were fed a diet without supplemental Zn, daily fecal Zn excretion and total daily Zn excretion exceeded the amount of Zn consumed, resulting in negative ATTD (-107.4%) and daily retention of Zn (-44.6 mg/d; Table 3.8).

Unlike responses of feeding diets without supplemental Zn to gestating sows, daily Zn intake of lactating sows exceeded the total Zn excretion, resulting in positive ATTD of Zn at 25.6%. Furthermore, after correcting for urine and milk Zn excretion, there was still positive retention of Zn (11.4 mg/d) during lactation.

DISCUSSION

Correct assessment of Zn digestibility using the indirect method relies heavily on accurate lab analysis of dietary Zn within treatments. Although slight variation within laboratory analysis is typically expected, we initially observed very high and inconsistent variation among analyzed Zn concentrations within the same diet subsamples. Recently, Jones et al. (2017) also observed high variation of Zn analysis within the same laboratory and estimated that at least 34 feed samples would need to be analyzed to have 95% confidence that analyzed Zn concentrations of a diet are within 4 ppm of the actual value. However, analyzing this many feed samples would be very costly, and may not be practical. To achieve accurate sample analysis results within 15 ppm, Jones et al. (2017) recommended that 2 to 5 feed samples must be analyzed. We suspect that procedures for subsample selection and uniform grinding of feed subsample within the laboratory may also influence analyzed Zn concentrations. To control this risk, diet samples were homogenized and ground (1.0 mm screen) prior to sample submission. As a result, we observed reduced variation of Zn concentration of dietary treatments.

Currently, there is no widely-accepted standard adaptation period or number of sample collection days for swine nutrient balance experiments (Jacobs et al., 2017). Furthermore, literature specific to diet acclimation periods for gestating sows is very limited. Gestating sows may have superior digestibility of nutrients due to limited feed

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intake, increased body weight (BW), higher rate of degradation of dietary fiber in the

hindgut and decreased rate of passage compared with growing-finishing pigs (Noblet and

Shi, 1993; Noblet and Shi, 1994; Le Goff and Noblet, 2001). These differences may

influence the diet acclimation period necessary for sows in a balance experiment utilizing

an indigestible marker, when compared to acclimation periods necessary for piglets or

growing/finishing pigs. Specifically, the decreased rate of passage for sows may increase

the adaptation period necessary to observe a consistent plateau of indigestible marker

fecal output. One study suggests that an adaptation phase of 7 d is adequate to achieve a

plateau of titanium excretion for sows (Jo and Kim, 2017). To confirm that an adaptation

period of 7 d was adequate for the desired plateau of fecal titanium (Ti) concentrations, a

subset of daily fecal samples (n = 12) were analyzed separately (data not shown).

Observations confirmed that desired fecal Ti concentrations were not achieved until at

least d 2 of the first balance experiment, which suggests that 8 d of adaptation is

necessary for gestating sows. Therefore, we only pooled and analyzed fecal samples from

d 8 and 9 of the gestation balance period for subsequent digestibility calculations.

Phytate, present in most swine feed ingredients, strongly impairs Zn absorption

(Lonneral, 2000), but the addition of the feed enzyme, phytase, can release trace elements

that may be bound in feedstuffs such as corn, wheat, soybean meal, wheat bran, and

wheat middlings (Jongbloed et al., 2004; Yu et al., 2018). Dietary treatments in this

experiment did not contain phytase. Therefore, the observed improvement in Zn

digestibility for gestation diets containing DDGS is not certain. However, researchers

have previously investigated digestibility of phosphorus (P) in growing pigs consuming

corn-SBM-DDGS diets and determined significant increases in ATTD, when compared

to diets without DDGS (Almeida and Stein, 2010). The improved ATTD of P in DDGS occurs when part of the phytate present in corn is hydrolyzed during ethanol and DDGS production (Stein and Shurson, 2009). We suspect that this process may similarly affect ATTD of Zn when DDGS is included in the diet.

Gestation

High fiber diets often reduce dry matter digestibility in adult sows (Le Goff et al., 2002; Holt et al., 2006). As a result, including DDGS in the experimental diets may have had an antagonistic effect on Zn digestibility in this experiment. However, due to higher dry matter digestibility in Control diets compared to DDGS diets, the negative ATTD, TTTD, and retention of Zn were not expected. Ultimately, daily excretion of Zn is affected by the relative proportions of indigestible Zn and endogenous losses of Zn, and exceeded dietary intake resulting in the calculated negative digestibilities and retentions observed.

Previously, scientists observed that the average mean retention time of feed in the gastrointestinal tract of sows was 81 h, but also reported great variability with ranges from 68 h to 95 h (Le Goff et al., 2002). Although an adaptation period of 7 d was allowed before balance periods, we confirmed that consistent analysis of the indigestible marker was not observed until d 2 of the collection period. Collection periods within this current experiment lasted only 72 h. As a result, one may hypothesize that the adaptation and collection periods for gestating sows in this experiment may not have been long enough for the indigestible marker to fully pass through the gastrointestinal tract, causing the observed negative digestibility of Zn among treatments. Increased retention time of feed in the gastrointestinal tract likely increased fermentation of dietary fiber in the

cecum (Williams et al., 2001; Jha and Berreocoso, 2015) which may have aided the observed increase of Zn digestibility within DDGS gestation treatments. To clarify this issue, further investigation of rate of passage for digesta and indigestible markers in meal-fed sows is necessary.

Total Zn excretion was reduced ($P < 0.05$) for diets containing organic Zn supplemented as PSZn, which is in agreement with the observed reduction in excretion of another organic Zn source compared to inorganic Zn sources previously determined by Nitrayova et al. (2012). There are discrepancies concerning digestibility and bioavailability of Zn sources (Hill et al., 1986; Wedekind et al., 1992; Wedekind et al, 1994; Close, 2003), yet studies conducted by researchers in poultry and growing-finishing pigs suggest that digestibility and bioavailability of organic Zn sources are generally equal to that of $ZnSO_4$ (Cao et al., 2000; Lebel et al., 2014). Although this may be true, it may not be so for gestating and lactating sows. The improved ATTD, TTTD, and retention of Zn for sows consuming DDGS-corn-soybean meal-based diets containing polysaccharide-complexed Zn in the current study confirm that digestibility of Zn from organic Zn may actually be superior to inorganic Zn for diets fed to reproducing sows containing DDGS. Furthermore, we suspect that reduced phytate among DDGS diets may have affected responses observed for Zn digestibility in gestation diets.

The primary route of excretion for Zn is through the feces (Poulsen and Larsen, 1995). Therefore, as expected, there were no effects of diet, Zn source, or their interaction on urinary Zn excretion among treatments. Urine collected from sows that exhibited signs of infection, contained blood in their urine, had a catheter that fell out, or had disconnected tubing from sample buckets were removed from the balance. Generally,

sows maintained good health status and only 3.7% of samples were unable to be analyzed for Zn content.

Stage of gestation may be an important factor to consider when evaluating digestibility and retention of Zn sources. This balance experiment was conducted from approximately d 85 to 87 of gestation. Often, swine producers begin to "bump feed" sows in late gestation beginning around d 90 of gestation until farrowing, to improve individual piglet birth weight (Goncalves et al., 2016). Feed intake was limited during the gestation balance trials. However, increased feed intake, or the ability to consume feed more than once per day, a practice some producers implement in late gestation, may affect digestibility and utilization of nutrients such as Zn (Cunningham et al., 1962; Shi and Noblet, 1993) since passage rate of digesta increases. Therefore, Zn utilization in later gestation under varying commercial practices must still be evaluated in the future.

Lactation

The observed reduction in Zn digestibility and increased fecal output from diets containing DDGS agree with conclusions from previously investigated impacts of high dietary fiber on nutrient digestibility (Wenk, 2001; Agyekum and Nyachoti, 2017). Apparent total tract digestibility and TTTD in lactating sows fed supplemental PSZn in corn-soybean meal based diets were superior to all other dietary treatments, similar to observations from an experiment evaluating organic and inorganic Zn sources in growing pigs by Liu et al. (2014). However, in contrast, $ZnSO_4$ was more digestible for lactating sows consuming diets containing DDGS, when compared to corn and soybean meal-based diets.

We speculate there may be a threshold of dietary fiber inclusion that actually decreases digestibility of organic Zn so much that the initially positive effects of PSZn in corn and soybean-meal diets become negatively affected when dietary fiber concentration exceeds some threshold. Further investigation is necessary to confirm this speculation. Nonetheless, it appears that organic Zn inclusion in corn and soybean-meal diets has the greatest ATTD, TTTD, and overall retention for lactating sows.

Baseline Zn Losses

Although we did not feed a completely Zn-free diet to truly measure endogenous losses of Zn, the negative retention of Zn observed in the gestation balance period may represent daily losses of Zn during gestation. Currently, the NRC (2012) suggests a daily Zn intake requirement of 210 mg/d for gestating sows. Gestating sows fed diets without supplemental Zn consumed 47.9 mg/d, yet excreted 44.6 mg/d. However, we cannot determine what portions of this Zn loss is due to endogenous Zn losses, or from poorly digestible Zn from commonly used feed ingredients.

Baseline losses of Zn likely still occur during lactation, however, these losses do not exceed dietary intake and utilization. The NRC (2012) suggests a daily Zn intake of 597 mg/d for lactating sows. Increased daily feed intake of lactating sows compared with gestating sows resulted in a greater daily Zn intake of 231.5 mg/d. Sows excreted 220.1 mg of Zn, therefore exhibiting slightly positive daily Zn retention (11.4 mg/d). Again, of the daily Zn losses, we cannot estimate what portion is due to poorly digestible Zn or endogenous losses.

Daily Zn intake requirements for gestating and lactating sows have not been evaluated for modern sows. Without full understanding of modern sow Zn requirements,

nutritionists may be overfeeding or underfeeding Zn. Estimating rate of Zn depletion in

sows may provide some guidance to evaluate current Zn feeding practices. Due to the

observed negative Zn retention in gestation, one may estimate that the rate of whole body

Zn depletion is greater for gestating sows compared to lactating sows. As a result, there

may be flexibility to fine-tune lactation sow diets to limit total Zn excretion without

increasing the rate of Zn depletion.

Zinc Biomarkers

Mineral requirements of sows are met by providing adequate amounts of

bioavailable forms in the diet relative to nutritional demands during gestation and

lactation. If the daily requirement exceeds dietary intake, body stores may be mobilized

to satisfy any deficit until body reserves become depleted. Depleted body mineral

reserves lead to reduced health and performance (Mahan, 1990). Zinc requirements

change throughout the reproductive cycle of sows, and as a result, many biomarkers of

Zn status fluctuate during gestation and lactation (Van Riet et al., 2015). Several

biomarkers may reflect Zn status of sows such as: plasma, liver, and milk concentrations

of Zn; metallothionein concentrations in blood; enzymes such as alkaline phosphatase

and superoxide dismutase; and concentrations of Zn in bone, kidney, and pancreas tissues

(McDowell, 2003). Unfortunately, there is no single, widely-accepted biomarker for

assessing Zn status in sows. Therefore, an assessment of a multitude of easily accessible

biomarkers such as plasma, milk, and growth performance of piglets should be

considered.

The lack of differences among plasma, colostrum, and milk Zn samples of sows,

regardless of dietary treatment, confirms that short-term Zn status was not impacted by

Zn or diet source. Sows could have mobilized Zn stores to maintain plasma, colostrum, milk Zn status so that short-term Zn status did not appear to be deficient. Overall, sows seemed to maintain a relatively similar Zn status throughout collection periods within the experiment.

Previous evaluation of Zn sources among sow diets have reported increased Zn concentrations in milk for sows consuming organic minerals (Acda and Chae, 2002), but this response was not observed in the current experiment.

The inability to distinguish differences in Zn concentrations of plasma among dietary treatments suggests a similar response to that of piglets and nursery pigs when fed inorganic or organic Zn sources (Case and Carlson, 2002; Schlegel et al., 2012). The lack of differences in plasma was not surprising, since blood sampling occurred at only one time point within each balance period. However, it is interesting that plasma Zn concentration during LowZn balance periods was numerically lower than all other collections. Although there is no single biomarker to evaluate Zn status of animals (Wood, 2000), plasma Zn is a widely accepted tool (Hambidge, 2003). The analyzed plasma Zn concentrations suggest that overall Zn status of sows was relatively similar across treatments.

CONCLUSION

Diets high in fiber, as supplemented by DDGS, reduced ($P < 0.05$) dry matter digestibility for sows in gestation and lactation; however, ATTD and TTTD of Zn decreased only during lactation. We are uncertain as to why a similar observation was not evident in gestation, but it appears that ATTD and TTTD of Zn, regardless of Zn source, improved under conditions of higher dietary fiber intake in gestation. Adding PSZn to

DDGS diets improved ($P < 0.05$) ATTD, TTTD, and overall retention of Zn in gestation compared with sows fed PSZn in corn-soybean meal diets. In contrast, PSZn supplementation in lactation DDGS diets significantly decreased ATTD, TTTD, and overall retention of Zn, when compared to a corn-soybean meal based diet or $ZnSO_4$. There were no differences in analyzed Zn concentrations of urine, plasma, colostrum, or milk at any time within the experiment, suggesting limited impact of dietary treatments on overall Zn status of sows throughout the trial.

Zinc excretion surpassed Zn intake for sows consuming diets without supplemental Zn in gestation. However, sows fed diets without supplemental Zn in lactation exhibited baseline Zn losses that did not exceed dietary intake and utilization. Of the Zn losses observed in both gestating and lactating sows, we cannot determine what portions is due to endogenous Zn losses, or from poorly digestible Zn. Extensive investigation of gestation and lactation Zn requirements for sows must be considered in the future to fully comprehend these observations and optimize Zn nutrition.

Table 3.1. Ingredient and nutrient composition of gestation diets (as-fed basis)[1]

Ingredient, %	Control ZnSO₄	Control PSZn	DDGS[2] ZnSO₄	DDGS[2] PSZn	LowZn
Corn	89.29	89.29	54.52	54.52	89.29
DDGS[2]	-	-	40.00	40.00	-
Soybean meal, 46.5% CP	7.71	7.71	2.88	2.88	7.71
Monocalcium phosphate, 21%	1.01	1.01	0.28	0.28	1.01
Limestone	0.99	0.99	1.32	1.32	0.99
Salt	0.50	0.50	0.50	0.50	0.50
ZnSO₄ premix[3]	0.50	-	0.50	-	-
PSZn premix[4]	-	0.50	-	0.50	-
Zn free premix[5]	-	-	-	-	0.50
TOTAL	100.00	100.00	100.00	100.00	100.00
Calculated nutrient composition:					
ME, kcal/kg	3,319	3,319	3,334	3,334	3,319
Crude protein, %	11.00	11.00	16.83	16.83	11.00
Crude fiber, %	2.43	2.43	4.49	4.49	2.43
Crude fat, %	3.72	3.72	5.22	5.22	3.72
NDF, %	9.36	9.36	17.73	17.73	9.36
SID Lys, %	0.38	0.38	0.38	0.38	0.38
SID Trp, %	0.09	0.09	0.10	0.10	0.09
SID Thr, %	0.33	0.33	0.45	0.45	0.33
SID Met + Cys, %	0.38	0.38	0.51	0.51	0.38
Calcium, %	0.60	0.60	0.60	0.60	0.60
Phosphorus, total %	0.52	0.52	0.51	0.51	0.52
STTD Phosphorus, %	0.26	0.26	0.26	0.26	0.26
Zinc added, ppm	100	100	100	100	0
Zinc total, ppm	123	123	146	146	23
Analyzed composition:					
Moisture, %	13.2	13.1	11.2	11.7	13.2
Crude protein, %	9.5	9.2	17.5	17.1	9.6
Crude fat, %	1.9	2.0	3.4	3.4	1.8
Crude fiber, %	1.7	1.7	3.6	3.5	1.8
Ash, %	4.4	4.4	5.0	4.7	4.5
Zinc total, ppm[6]	160.0	112.0	130.0	150.0	21.6

[1]Titanium dioxide used as indigestible marker added at 0.50% of the diet.

[2]Distillers dried grains with solubles.

[3]Contained the following nutrients per kilogram of premix: vitamin A, 3,742,468 IU; vitamin D₃, 748,493 IU; vitamin E, 40,827 IU; vitamin K, 220 mg; riboflavin, 716 mg; niacin, 1,918 mg; pantothenic acid, 2,315 mg; vitamin B₁₂, 7 mg; iodine, 66 mg; selenium, 66 mg; zinc (as zinc sulfate monohydrate), 44,092 mg; iron, 35,274 mg; manganese, 11,023 mg; copper, 8,818 mg.

[4]Premix as above but SQM Zn (QualiTech, Chaska, MN) was the source of zinc.

[5]Premix as above but containing no supplemental zinc.

[6]Average of 2 dietary sample analyses.

Table 3.2. Ingredient and nutrient composition of lactation diets (as-fed basis)[1]

Ingredient, % (as-fed basis)	Control		DDGS[2]		
	ZnSO$_4$	PSZn	ZnSO$_4$	PSZn	LowZn
Corn	75.62	75.62	52.35	52.35	75.62
DDGS[2]	-	-	30.00	30.00	-
Soybean meal, 46.5% CP	20.89	20.89	14.33	14.33	20.89
L-Lysine HCl	0.10	0.10	0.19	0.19	0.10
Monocalcium phosphate, 21% P	1.44	1.44	0.91	0.91	1.44
Limestone	0.95	0.95	1.22	1.22	0.95
Salt	0.50	0.50	0.50	0.50	0.50
ZnSO$_4$ Premix[3]	0.50	-	0.50	-	-
PSZn Premix[4]	-	0.50	-	0.50	-
Zn Free Premix[5]	-	-	-	-	0.50
TOTAL	100.00	100.00	100.00	100.00	100.00
Calculated nutrient composition:					
ME, kcal/kg	3,297	3,297	3,308	3,308	3,297
Crude protein, %	16.08	16.08	19.41	19.41	16.08
Crude fiber, %	2.69	2.69	4.18	4.18	2.69
Crude fat, %	3.58	3.58	4.73	4.73	3.58
NDF, %	9.39	9.39	15.64	15.64	9.39
SID Lys, %	0.78	0.78	0.78	0.78	0.78
SID Trp, %	0.16	0.16	0.15	0.15	0.16
SID Thr, %	0.50	0.50	0.55	0.55	0.50
SID Met + Cys, %	0.49	0.49	0.57	0.57	0.49
Calcium, %	0.70	0.70	0.70	0.70	0.70
Phosphorus, total %	0.66	0.66	0.64	0.64	0.66
STTD Phosphorus, %	0.35	0.35	0.35	0.35	0.35
Zinc added, ppm	100	100	100	100	0
Zinc total, ppm	128	128	146	145	28
Analyzed composition:					
Moisture, %	12.7	12.7	11.5	11.6	12.3
Crude protein, %	15.7	14.8	19.6	19.3	14.0
Crude fat, %	1.7	1.7	2.8	2.9	1.8
Crude fiber, %	2.1	2.0	3.3	3.5	1.9
Ash, %	5.4	5.5	6.0	5.9	5.1
Zinc total, ppm[6]	129.9	134.2	136.0	123.7	29.3

[1]Titanium dioxide used as indigestible marker added at 0.50% of the diet.

[2]Dried distillers grains with solubles.

[3]Contained the following nutrients per kilogram of premix: vitamin A, 3,742,468 IU; vitamin D$_3$, 748,493 IU; vitamin E, 40,827 IU; vitamin K, 220 mg; riboflavin, 716 mg; niacin, 1,918 mg; pantothenic acid, 2,315 mg; vitamin B$_{12}$, 7 mg; iodine, 66 mg; selenium, 66 mg; zinc (as zinc sulfate monohydrate), 44,092 mg; iron, 35,274 mg; manganese, 11,023 mg; copper, 8,818 mg.

[4]Premix as above but SQM Zn (QualiTech, Chaska, MN) was the source of zinc.

[5]Premix as above but containing no supplemental zinc.

[6]Average of 2 dietary sample analyses.

Table 3.3. Daily Zn balance, apparent total tract digestibility (ATTD), and true total tract digestibility (TTTD) of Zn for sows during gestation

	Control		DDGS			P-value		
Item	$ZnSO_4$	PSZn	$ZnSO_4$	PSZn	SE	Diet	Zn	Diet*Zn
No. of sows	8	8	8	8	-	-	-	-
ADFI, kg/d	2.22	2.22	2.22	2.22	-	-	-	-
Diet DM, %	86.8	86.9	88.7	88.3	-	-	-	-
ADFI, kg/d DM	1.93	1.93	1.97	1.96	-	-	-	-
Diet Zn, mg/kg DM	184.3	128.9	146.5	169.9	-	-	-	-
Diet Ti, mg/kg DM	3,443	3,314	3,087	3,115	-	-	-	-
Zn consumed, mg/d DM	355.6	248.9	288.9	333.4	-	-	-	-
Ti consumed, mg/d DM	6,645.6	6,401.1	6,089.9	6,112.1	-	-	-	-
Fecal excretion[1], kg/d DM	0.33[ab]	0.31[a]	0.38[b]	0.38[b]	0.02	< 0.01	0.62	0.63
Fecal Zn, mg/kg DM	1,401[a]	1,220[b]	911[c]	822[d]	37.8	< 0.01	< 0.01	0.02
Fecal Ti, mg/kg DM	20,682[a]	22,275[a]	16,029[b]	16,194[b]	1,136.2	< 0.01	0.33	0.41
Fecal Zn excreted[2], mg/d	462.5[a]	384.9[ab]	352.5[b]	320.2[b]	33.2	< 0.01	0.03	0.36
Fecal Ti excreted[3], mg/d	6,646.0[a]	6,401.0[b]	6,090.0[c]	6,112.0[d]	1.6	< 0.01	< 0.01	< 0.01
DM digestibility[4], %	82.9[xy]	83.9[x]	80.5[y]	80.4[y]	1.2	< 0.01	0.66	0.59
ATTD Zn[5], %	-31.0[a]	-52.9[a,x]	-21.9[ab,y]	4.1[b]	11.5	< 0.01	0.82	0.01
TTTD Zn[6], %	-3.2[a]	-13.1[a]	13.1[ab]	34.4[b]	11.5	< 0.01	0.54	0.09
Urine excretion[7], kg/d	13.1	12.2	12.2	8.1	4.0	0.53	0.54	0.70
Urine Zn concentration, mg/kg	0.3	0.8	0.5	0.6	0.2	0.99	0.19	0.32
Urine Zn excreted[8], mg/d	3.0	2.6	4.1	4.0	0.6	0.10	0.74	0.80
Total Zn excreted[9], mg/d	465.3[a]	387.3[ab]	356.5[b]	324.0[b]	33.2	< 0.01	0.03	0.36
Zn retained[10], mg/d	-109.7[a]	-138.4[a]	-67.6[ab]	9.4[b]	33.2	< 0.01	0.34	0.04

[xy]Means within a row with different superscripts differ ($P < 0.10$).

[1]Fecal excretion = $DM_{intake}*(Ti_{diet}/ Ti_{feces})$.

[2]Fecal Zn excreted = $Zn_{feces}*Output_{feces}$.

[3]Fecal Ti excreted = $Ti_{feces}*Output_{feces}$.

[4]DM digestibility, % = $[1-(Ti_{diet}/Ti_{feces})]*100$.

[5]ATTD Zn, % = $[1 - (Zn_{feces}/Zn_{diet})$ x $(M_{diet}/M_{feces})]*100$.

[6]TTTD Zn, % = $ATTD + [(Zn_{end}/Zn_{diet})*100]$.

[7]Represents urine output of the following sows per treatment: ConZnSO$_4$ = 5; ConPSZn = 3; DDGS/ZnSO$_4$ = 6; and DDGS/PSZn = 6

[8]Urine Zn excreted = $(Zn_{urine}*Urine_{output})$.

[9]Total Zn excreted = $(FecalZn_{excreted} + UrineZn_{excreted})$.

[10]Zn retained = $(Zn_{intake} - Zn_{excreted})$.

Table 3.4. Daily Zn balance, apparent total tract digestibility (ATTD), and true total tract digestibility (TTTD) of Zn for sows during lactation

Item	Control		DDGS		SE	P-value		
	$ZnSO_4$	PSZn	$ZnSO_4$	PSZn		Diet	Zn	Diet*Zn
No. of sows	6	8	6	9	-	-	-	-
ADFI, kg/d	6.71[a]	5.68[ab]	4.47[b]	4.83[b]	0.64	<0.01	0.43	0.09
Diet DM, %	87.3	87.3	88.4	88.5	-	-	-	-
ADFI, kg/d DM	5.85[a]	4.96[ab]	3.96[b]	4.27[b]	0.56	<0.01	0.43	0.10
Diet Zn, mg/kg DM	148.8	153.7	153.7	139.9	-	-	-	-
Diet Ti, mg/kg DM	3,425	3,139	3,368	3,518	-	-	-	-
Zn consumed, mg/d DM	810.7[a]	712.8[ab]	618.3[ab]	582.7[b]	60.7	0.01	0.28	0.61
Ti consumed, mg/d DM	18,662[a]	14,554[b]	13,549[b]	14,649[ab]	1,179.0	0.07	0.27	<0.01
Fecal excretion[1], kg/d DM	0.79	0.66	0.73	0.82	0.1	0.42	0.79	0.10
Fecal Zn, mg/kg DM	987[a]	889[b]	759[c]	798[c]	26.4	< 0.01	0.07	<0.01
Fecal Ti, mg/kg DM	25,788[a]	23,700[b]	18,369[c]	18,394[c]	296.1	< 0.01	< 0.01	<0.01
Fecal Zn excreted[2], mg/d	772.3[a]	600.1[ab]	547.3[b]	653.2[ab]	80.6	0.09	0.52	<0.01
Fecal Ti excreted[3], mg/d	19,965[a,x]	15,656[ab,y]	13,329[b]	15,028[b]	1,878.0	< 0.01	0.29	0.01
DM digestibility[4], %	86.7[a]	86.7[a]	81.6[b,x]	80.8[b,y]	0.2	< 0.01	0.14	0.07
ATTD Zn[5], %	11.6[a]	22.9[b]	10.0[a]	-8.6[c]	2.6	< 0.01	0.03	< 0.01
TTTD Zn[6], %	28.3[a]	39.0[b]	26.1[a]	9.1[c]	2.6	< 0.01	0.05	< 0.01
Urine excretion[7], kg/d	17.2	10.5	13.3	16.0	5.8	0.88	0.74	0.42
Urine Zn concentration, mg/kg	0.5	0.7	0.8	0.6	0.2	0.65	0.92	0.32
Urine Zn excreted[8], mg/d	6.2	8.1	9.4	6.8	1.7	0.60	0.86	0.23
Milk yield, kg/d	9.2	9.1	9.8	10.3	0.7	0.24	0.83	0.71
Milk Zn concentration, mg/kg	5.2	5.7	5.0	5.4	0.4	0.48	0.33	0.88
Milk Zn secreted[9], mg/d	51.7	51.9	49.0	54.5	5.7	0.99	0.63	0.64
Total Zn excreted[10], mg/d	830.2[a]	660.1[ab]	605.7[b]	714.5[ab]	80.6	0.10	0.55	< 0.01
Zn retained[11], mg/d	42.0[a]	100.9[b]	4.2[a]	-115.2[c]	19.1	< 0.01	0.02	< 0.01

[a-c]Means within a row with different superscripts differ ($P < 0.05$).

[x,y]Means within a row with different superscripts differ ($P < 0.10$).

[1]Fecal excretion = $DM_{intake}*(Ti_{diet}/Ti_{feces})$.

[2]Fecal Zn excreted = $Zn_{feces}*Output_{feces}$.

[3]Fecal Ti excreted = $Ti_{feces}*Output_{feces}$.

[4]DM digestibility, % = $[1-(Ti_{diet}/Ti_{feces})]*100$.

[5]ATTD Zn, % = $[1 - (Zn_{feces}/Zn_{diet}) \times (M_{diet}/M_{feces})]*100$.

[6]TTTD Zn, % = $ATTD + [(Zn_{end}/Zn_{diet})*100]$.

[7]Represents urine output of the following sows per treatment: ConZnSO$_4$ = 4; ConPSZn = 2; DDGS/ZnSO$_4$ = 5; and DDGS/PSZn = 5

[8]Urine Zn excreted = $(Zn_{urine}*Urine_{output})$.

[9]Milk Zn secreted = $(Zn_{milk}*Milk_{output})$.

[10]Total Zn excreted = $(FecalZn_{excreted} + UrineZn_{excreted})$.

[11]Zn retained = $(Zn_{intake} - Zn_{excreted})$.

Table 3.5. Effects of Zn source and diet on farrowing performance of sows

Item	Control ZnSO$_4$	Control PSZn	DDGS ZnSO$_4$	DDGS PSZn	SE	P-value Diet	P-value Zn	P-value Diet*Zn
Parity	3.2	2.7	2.2	2.8	0.6	0.43	0.82	0.41
Lactation length, d	21.2	19.2	19.8	20.7	0.6	0.94	0.36	0.03
Bodyweight, kg								
D80 Gestation	244.4	244.4	239.5	245.8	12.5	0.80	0.68	0.67
Pre-Farrow[1]	249.0	250.7	243.2	258.8	12.1	0.88	0.30	0.38
Post-Farrow[2]	234.3	237.3	237.3	241.6	11.4	0.54	0.53	0.91
Weaning	239.0	236.8	225.5	226.7	13.3	0.10	0.94	0.80
Farrowing Performance								
Total pigs born	14.2	14.0	16.2	14.2	1.2	0.38	0.41	0.49
Pigs born alive	11.5	11.6	15.0	13.2	1.3	0.07	0.54	0.51
Pigs weaned	10.8	10.4	13.4	11.9	1.0	0.03	0.31	0.56
Piglet birth wt., kg	1.31[ab]	1.29[a]	1.24[a]	1.41[b]	0.1	0.42	0.03	< 0.01
Litter birth wt., kg[3]	18.1	17.7	20.2	20.4	1.7	0.17	0.96	0.87
Litter start wt., kg[4]	14.4[a]	15.1[a]	19.0[b]	18.8[b]	1.4	< 0.01	0.84	0.76
Piglet weaning wt., kg	7.1[a]	6.3[b]	5.6[c]	6.7[ab]	0.3	< 0.01	0.26	< 0.01
Litter weaning wt., kg[5]	69.8[xy]	64.5[x]	75.1[xy]	82.6[y]	5.3	0.04	0.83	0.24
Piglet gain, g/d	251.3	243.6	211.5	241.9	12.4	0.11	0.37	0.14
Litter gain, g/d	2,615.3	2,582.0	2,830.5	2,918.9	207.2	0.20	0.90	0.77

[ab]Means within a row with different superscripts differ ($P < 0.05$).
[xy]Means within a row with different superscripts differ ($P < 0.10$).
[1]Sows were weighed 1 day prior to expected farrowing date.
[2]Sows were weighed within 24 h of completing farrowing.
[3]Total litter birth weight before cross-fostering.
[4]Total litter weight of live pigs after cross-fostering.
[5]Total litter weaning weight after cross-fostering.

Table 3.6. Colostrum and milk composition by treatment

Item	Control ZnSO$_4$	PSZn	DDGS ZnSO$_4$	PSZn	LowZn	SE	P-value Diet	Zn	Diet*Zn
Colostrum[1]									
Moisture, %	77.3	75.3	77.0	77.8	-	1.0	0.25	0.53	0.13
Crude protein, %	14.1	15.0	12.0	14.1	-	1.1	0.19	0.18	0.59
Crude fat, %	4.3	5.8	6.3	3.7	-	1.0	0.99	0.55	0.04
Zn, mg/kg	12.2	13.7	13.0	14.8	-	1.3	0.47	0.23	0.90
Milk									
Lactation[2]									
Moisture, %	79.1	79.9	79.9	79.8	-	0.9	0.66	0.70	0.57
Crude protein, %	5.3	5.7	5.0	5.3	-	1.0	0.14	0.13	0.81
Crude fat, %	9.7	8.6	9.5	9.1	-	1.1	0.90	0.47	0.77
Zn, mg/kg	5.2	5.7	5.0	5.4	-	0.4	0.48	0.33	0.88
LowZn Balance[3]									
Moisture, %	-	-	-	-	80.5	0.9	-	-	-
Crude protein, %	-	-	-	-	4.9	0.1	-	-	-
Crude fat, %	-	-	-	-	8.9	1.1	-	-	-
Zn, mg/kg	-	-	-	-	4.3	0.2	-	-	-

[1]Samples collected within 12 h after parturition.
[2]Samples collected on D10 of lactation.
[3]Samples collected on D17 of lactation.

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Table 3.7. Zinc concentration of plasma in sows during gestation and lactation

Item	Control		DDGS				P-value		
	$ZnSO_4$	PSZn	$ZnSO_4$	PSZn	LowZn	SE	Diet	Zn	Diet*Zn
Plasma Zn, mcg/dL									
Initial	69.6	67.0	66.8	72.1	-	2.9	0.67	0.62	0.16
Gestation[1]	70.0	75.0	75.3	73.3	-	3.6	0.54	0.63	0.25
Gest. LowZn[2]	-	-	-	-	57.9	2.2	-	-	-
Pre-Farrow[3]	78.3	77.8	79.8	83.5	-	4.7	0.17	0.56	0.42
Lactation[4]	86.0	76.7	83.2	79.7	-	3.5	0.98	0.08	0.41
Lact. LowZn[5]	-	-	-	-	49.9	2.4	-	-	-
Weaning[6]	86.7	97.6	89.8	90.3	-	5.7	0.68	0.28	0.32

[1]Samples collected on d 3 of gestation balance.
[2]Samples collected on d 3 of gestation LowZn balance.
[3]Samples collected 24 h prior to expected farrowing date.
[4]Samples collected on d 3 of lactation balance.
[5]Samples collected on d 3 of lactation LowZn balance.
[6]Samples collected on d 21 of lactation.

Table 3.8. Daily Zn balance, apparent total tract digestibility (ATTD), and true total tract digestibility (TTTD) of Zn for sows during gestation and lactation when fed diets not supplemented with Zn

Item	Gestation LowZn	SE	Lactation LowZn	SE
No. of sows	20	-	18	-
ADFI, kg/d	2.22	-	8.09	1.1
Diet dry matter (DM), %	86.8	-	87.7	-
ADFI, kg/d DM	1.93	-	7.10	0.9
Diet Zn, mg/kg DM	24.8	-	33.4	-
Diet Ti, mg/kg DM	3,422.0	-	3,158.0	-
Zn consumed, mg/d DM	47.9	-	231.5	7.8
Ti consumed, mg/d DM	6,601.1	-	21,890.0	740.0
Fecal excretion[1], kg/d DM	0.26	< 0.01	0.88	0.03
Fecal Zn, mg/kg DM	372.4	45.8	203.5	8.4
Fecal Ti, mg/kg DM	26,028.0	871.5	25,705.0	623.5
Fecal Zn excreted[2], mg/d	89.9	6.7	170.7	6.1
Fecal Ti excreted[3], mg/d	6,601.1	0.1	21,890.0	740.0
DM digestibility[4], %	86.6	0.5	87.6	0.3
ATTD Zn[5], %	-107.4	31.2	25.6	1.3
TTTD Zn[6], %	131.5	4.7	110.4	0.2
Urine excretion, kg/d	20.3	5.6	14.2	1.9
Urine Zn concentration, mg/kg	0.3	0.1	0.3	< 0.1
Urine Zn excreted[7], mg/d	2.6	0.3	3.9	0.3
Milk yield, kg/d	-	-	10.5	0.4
Milk Zn concentration, mg/kg	-	-	4.3	0.2
Milk Zn secreted[8], mg/d	-	-	45.5	2.6
Total Zn excreted[9], mg/d	92.5	6.7	220.1	6.1
Zn retained[10], mg/d	-44.6	6.7	11.4	2.8

[1]Fecal excretion = $DM_{intake}*(Ti_{diet}/Ti_{feces})$.
[2]Fecal Zn excreted = $Zn_{feces}*Output_{feces}$.
[3]Fecal Ti excreted = $Ti_{feces}*Output_{feces}$.
[4]DM digestibility, % = $[1-(Ti_{diet}/Ti_{feces})]*100$.
[5]ATTD Zn, % = $[1 - (Zn_{feces}/Zn_{diet}) \times (M_{diet}/M_{feces})]*100$.
[6]TTTD Zn, % = $ATTD + [(Zn_{end}/Zn_{diet})*100]$.
[7]Urine Zn excreted = $(Zn_{urine}*Urine_{output})$.
[8]Milk Zn secreted = $(Zn_{milk}*Milk_{output})$.
[9]Total Zn excreted = $(FecalZn_{excreted} + UrineZn_{excreted})$.
[10]Zn retained = $(Zn_{intake} - Zn_{excreted})$.

CHAPTER 4

Effects of supplementing transition sow diets with zinc on pre-weaning mortality and lifetime productivity of pigs under commercial rearing conditions

SUMMARY

The objectives of this experiment were to determine pre-weaning survival of low birth weight pigs when sows were supplemented with 3 dietary levels of zinc (Zn) in late gestation. Gilts and sows (n = 339) were assigned to one of three dietary treatments on d 75 of gestation based on parity. Dietary treatments were: 1) Control – sows fed a corn-soybean meal based diet containing 125 ppm total supplemental Zn as 75 ppm $ZnSO_4$ and 50 ppm AvailaZn™ (**CON**); 2) Intermediate – as Control + 240 ppm supplemental Zn as $ZnSO_4$ (**INT**); and 3) High – as Control + 470 ppm supplemental Zn as $ZnSO_4$ (**HI**). Final supplemental Zn concentrations of the three dietary treatments were as follows: 1) **CON** – 125 ppm; 2) **INT** – 365 ppm; and 3) **HI** – 595 ppm. Sows received dietary treatments from about d 75 of gestation until farrowing. Individual piglet birthweights were recorded within 12 h of parturition. Instances of piglet mortality were recorded daily. The statistical model considered fixed effects of treatment and random effects of parity. Piglets from sows fed the intermediate diet had greater ($P < 0.05$) birth weights than those fed CON (1.42 vs. 1.38 kg, respectively), while offspring from sows fed HI tended to have greater ($P < 0.10$) birth weights (1.40 kg). Furthermore, incidence of low birth weight pigs was less ($P < 0.05$) for sows consuming INT compared with sows fed CON and HI. Despite differences in birth weight, there were no differences ($P > 0.05$) in total pigs born, born alive, or weaned, nor differences in individual piglet gain or weaning weight across treatments. Mortality of low birth weight pigs was lowest ($P <$

0.05) for offspring from sows fed HI. Similarly, overall piglet mortality decreased ($P <$ 0.10) as dietary Zn content increased. A subset of pigs (n = 450, n = 150/treatment) were selected at weaning to evaluate sow dietary treatment effects on post-weaning performance. There were no significant treatment effects on carcass characteristics of market pigs. Overall, effects of supplemental dietary Zn at 365 and 595 ppm in late gestation improved pre-weaning survival of low birth weight piglets and reduced overall pre-weaning mortality.

Key words: carcass characteristics, mortality, pre-weaning, swine, zinc

INTRODUCTION

Sows in transition from gestation to lactation are in a period of dramatic physiological change due to increased nutrient demands of rapidly growing fetuses *in utero* to subsequent farrowing of offspring and lactation. These physiological changes require coordinated hormonal, nutritional, and management transitions to optimize piglet viability and post-natal growth. Inadequate preparation for these massive physiological changes could lead to increased stillbirth rate, increased number of low birthweight pigs (< 1 kg birth weight), and increased pre-weaning mortality of piglets. Pre-weaning mortality can be attributed to many factors such as low viability, trauma from crushing, starvation, or disease (Vaillancourt et al., 1990; Lay et al., 2002; Edwards and Baxter, 2015). In fact, it is common for commercial swine farms to experience pre-weaning mortality rates between 12 and 25% (Alonso-Spilsbury, 2007).

Birth weight strongly influences lifetime growth performance and subsequent carcass characteristics of market pigs (Lay et al., 2002; Douglas et al., 2013). Piglets weighing less than 1 kg at birth typically are at greater risk for mortality and poor lifetime growth performance compared to pigs with normal birth weights (Foxcroft et al., 2006; Diaz et al., 2017). Therefore, sows that produce pigs with birth weights greater than 1 kg have greater economic value for pork producers because they have enhanced post-natal piglet survival and growth performance. Post-natal management, environment, and nutritional interventions to improve piglet viability and growth have resulted in varying degrees of success (Deen and Bilkei, 2004; Douglas et al., 2014; Edwards and Baxter, 2015). A more effective strategy might be to intervene before farrowing to better prepare piglets for life outside the sow's uterus. Previously, researchers have considered effects

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of varying levels of dietary energy or amino acids in gestating sow diets to improve piglet birth weights or survival of low birth weight pigs, but with inconsistent results (Goodband et al., 2013).

Increasing dietary zinc supplementation may be a useful pre-natal intervention. Elevated dietary zinc concentrations can reduce incidence of stillborn pigs (Hill et al., 1983) and increase litter birth weight (Payne et al., 2006). Researchers clearly have demonstrated that zinc, copper, and manganese accumulate in high concentrations in the conceptus (Hostetler et al., 2003). Impaired accumulation of these trace minerals may negatively affect the piglet's chance of survival. Vallet et al. (2014) demonstrated, with a limited numbers of gilts, that elevated dietary zinc concentrations during late gestation reduced pre-weaning mortality of low birth weight pigs, but this observation has not been verified under large-scale commercial conditions. Therefore, the objectives of this study were to determine pre-weaning survival of piglets and lifetime performance of pigs weighing less than 1 kg at birth from sows fed increasing levels of dietary Zn approximately 45 d pre-partum.

MATERIALS and METHODS

This experiment was conducted in a commercial sow facility (1,200 sows; Schwartz Farms, Inc., Comfrey, MN). The experimental protocol was reviewed and approved by the University of Minnesota Institutional Animal Care and Use Committee (IACUC# 1083-35724A). The experiment began in May, 2018 and concluded in February, 2019.

Animals, Housing, and Treatments

Three consecutive weeks of production incorporating 339 total females (parity 0 to 7; PIC Camborough, Hendersonville, TN) were assigned based on parity to one of three dietary treatments at approximately d 75 of gestation. Treatments were assigned to a block of gestation stalls to avoid cross-contamination of treatments from one sow to adjacent sows. One "buffer" sow was placed at the end of each block of stalls to receive the same dietary treatment but was not included in the experiment. Sows were later moved to farrowing stalls within 3 d of expected farrowing date.

Dietary treatments consisted of: 1) Control – sows fed a corn-soybean meal based diet containing 125 ppm total supplemental Zn as 75 ppm $ZnSO_4$ and 50 ppm amino acid complexed Zn (as AvailaZn™; Zinpro Corp., Eden Prairie, MN) (**CON**); 2) Intermediate – as Control + 240 ppm supplemental Zn as $ZnSO_4$ (**INT**); and 3) High – as Control + 470 ppm supplemental Zn as $ZnSO_4$ (**HI**). Final supplemental Zn concentrations of the three dietary treatments were as follows: 1) **CON** – 125 ppm; 2) **INT** – 365 ppm; and 3) **HI** – 595 ppm. Gestation and lactation diet composition was fed based on the farm's standard operating procedures (Table 4.1). Dietary treatments were imposed by feeding 60 ml (45 g; INT) or 120 ml (90 g; HI) of the Zn supplement as a top-dress to provide 518 mg or 1,038 mg Zn, respectively, once daily to the feed hoppers each afternoon prior to feeding only for sows assigned to the INT and HI treatments (Table 4.2). Control sows did not receive any top-dressed Zn supplement. Sows remained on their assigned dietary treatment and received 2.2 kg of feed once daily until farrowing. Immediately after

farrowing, all sows were fed a common lactation diet and allowed ad libitum access to feed and water.

Sows were housed individually in stalls during gestation until approximately d 110 of gestation. Sows were then moved to individual farrowing stalls within farrowing rooms until weaning of litters. Each farrowing room contained 39 farrowing stalls. Farrowing stalls were equipped with one stainless steel feeder and one nipple waterer on a partially slatted floor over a deep manure collection pit. An independent controller within each farrowing room operated all heaters and ventilation fans. One heat lamp was placed in the creep area of each farrowing stall as a supplemental heat source for piglets.

Sow and Piglet Performance

Sows were identified individually using ear tags. Body condition and lameness scores were recorded at initiation of dietary treatments, at approximately d 110 of gestation, within 24 h of expected parturition, and at weaning. Body condition scores were determined using a body condition caliper placed at the last rib of the sow. Visual lameness scores were recorded and assigned as sows stood up within stalls according to the following scale: 1) Normal: sow standing with weight equally distributed on all feet; or 2) Lame: sow with arched back, weight unequally distributed on feet, difficulty or inability to stand.

Sow reproductive performance measurements included total number of piglets born, born alive, stillborn, mummified, and weaned per litter. Within 12 h of birth and prior to cross-fostering, all piglets were weighed individually and ear tagged. Ear tags were color-coded to match that of their dam's assigned dietary treatment. Litter sizes were standardized to 12 or 13 piglets per sow by cross-fostering within 24 h of farrowing.

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Cross-fostering within treatment was attempted, but was not controlled throughout

lactation. All piglets were processed according to the standard operating procedure

established by the farm within 24 to 48 h of birth. Piglet processing included tail docking,

needle teeth clipping, administering injectable iron, and castration of male piglets.

Incidence of stillborn and mummified piglets were recorded, but were not weighed. Any

pigs that died shortly before or during parturition, which was likely due to asphyxia or

dystocia, were classified as stillborn. Piglets were monitored daily for instances of

morbidity and mortality. Any dead piglets were weighed and recorded according to date,

piglet eartag ID, sex, and treatment. One day before weaning, individual piglet body

weights were determined and recorded to calculate total weight gain during the nursing

period. Piglets were weaned at approximately 18.1 ± 0.1 d of age.

A subset of about 15 litters per treatment (n = 150 pigs/treatment) of both low

birth weight (n = 50 pigs/treatment) and normal to heavy birth weight (n = 100

pigs/treatment) pigs were selected at weaning to monitor post-weaning growth

performance and subsequent carcass characteristics. Selected pigs were fed common,

nutritionally adequate diets throughout the entire growing-finishing period. All instances

of mortality were recorded. Pigs were tattooed individually before shipment and were

harvested at JBS Pork in Worthington, MN. Individual pig tattoo numbers were used to

collect hot carcass weight, backfat depth measured between the third and fourth rib, and

loin depth data. An optical probe (Fat-O-Meat'er™, Frontmatec Group, Denmark) was

used to determine backfat thickness and loin depth of all carcasses. The following

equation, as determined by JBS Pork, was used to calculate percentage carcass lean:

Percentage carcass lean = 58.86 - 0.61 × (backfat depth, inches) + 0.12 × (loin

depth, inches).

The following equations reported by NPPC (2000) were used to calculate percentage fat-

free carcass lean, and lean gain per day:

Percentage fat-free carcass lean (FFL) = [15.31 – (31.277 × backfat depth, inches)

+ (3.813 × loin depth, inches) + (0.51 × HCW, pounds)] / HCW × 100; and

Lean gain/day = (FFL at ending weight - FFL in feeder pig) / days on test.

Sample Analysis

Two random samples of the zinc top dress and gestation diet were collected at

initiation and throughout feeding of dietary treatments to each farrowing group. All

samples were stored at -20°C until shipment for analysis. Diet and top dress samples

were sent to Minnesota Valley Testing Laboratories, Inc. (New Ulm, MN) for proximate

analysis and determination of zinc concentration. Standard procedures (AOAC

International, 2006) were followed for analysis of moisture (Method 930.15), ash

(Method 942.05), fat (Method 2003.05), CF (Method BA6A-05), CP (Method 990.03),

calcium (Method 985.01), phosphorus (Method 985.01), and zinc (Method 985.01)

concentrations.

Statistical Analysis

Experimental data were analyzed using the PROC GLIMMIX procedure of SAS

(Version 9.4, SAS Institute Inc., Cary, NC) with a Gaussian distribution. Sow was

considered the experimental unit. Post-weaning data considered pig as the experimental

unit. The statistical model included fixed effects of dietary treatment, farrowing group,

and their interaction. Farrowing group was tested as a fixed effect for all variables but did

not influence performance or mortality variables, so it was removed from the final statistical model. Treatment means were separated using the PDIFF option with the Tukey-Kramer adjustment for multiple comparisons.

Chi square analyses were used to determine the influence of gestation dietary treatments on categorical response variables such as pre-weaning and post-weaning piglet mortality, lameness scores, and incidence of stillbirths and mummies. All data were reported as least square means and considered statistically significant at $P < 0.05$ with $P < 0.10$ considered a trend.

RESULTS and DISCUSSION

Sow Performance

Average sow parity and days on trial were not different across dietary treatments (Table 4.3). Average gestation length was greater ($P < 0.05$) for HI sows (115.6 d) compared with INT and CON sows (115.2 d). However, lactation length and days to first service were not different across treatments. At initiation of the experiment, body condition caliper scores were lower ($P < 0.05$) for CON sows compared with sows fed INT and HI diets. However, the observed scores across treatments were all within optimal ranges (Knauer and Baitinger, 2015). Body condition scores one day before farrowing and at weaning were not different across treatments. Sow farrowing performance including total number of pigs born, born alive, and weaned per litter were not different regardless of dietary treatment throughout the experiment. Sows farrowed about 14 total pigs born per litter, of which 13 to 14 pigs were born alive, and sows averaged slightly less than 11 piglets weaned per litter. Sows utilized in this experiment performed similarly to that of many U.S. commercial swine production systems (Knox et

al., 2013; Stalder, 2018). Instances of lameness throughout the experiment were rare (Table 4.4) and not different across treatments. When further evaluating farrowing performance of sows, there were no differences across treatments regarding the total number of stillborn or mummified pigs (Table 4.5).

Sows consuming the INT diet were less likely ($P < 0.01$) to produce pigs with low birth weights (≤ 1 kg) compared with sows consuming CON or HI treatments (Table 4.6). Sows producing litters of pigs with a high proportion of low birth weight piglets are typically at greater risk of pre-weaning mortality (Milligan et al., 2002; Kapell et al., 2011). The number of low birth weight pigs for sows consuming CON or HI treatments in this experiment were greater than those observed in a previous experiment conducted by Bergstrom et al. (2009), where incidence of small pigs per litter weighing less than 1 kg ranged from 8 to 13%. However, the number of low birth weight pigs for sows fed CON and HI treatments was similar to the 14.9% reported by Feldpausch et al. (2016). Overall, it appears that sows within this commercial production facility produced pigs similar to herds studied previously.

Piglet Growth Performance

Overall, piglets from sows fed the INT treatment had heavier birth weights than piglets from sows receiving CON treatment (Table 4.7). Although piglets from sows fed INT were larger at birth, there were no differences in weaning weight or total piglet gain across treatments. As a result, the advantage in initial birth weight for INT piglets was not maintained throughout the nursing period until weaning.

Growth performance of piglets was evaluated in the following three birth weight categories: 1) Low birth weight (≤ 1.00 kg); 2) Normal birth weight (1.01 to 1.75 kg);

and 3) Heavy birth weight ($\geq$ 1.76 kg). Low and heavy birth weights were determined to be within one standard deviation from the population mean birth weight. Total weight gain and weaning weight of low birth weight pigs were similar across dietary treatments. Furthermore, growth rate of heavy birth weight pigs was not different, regardless of the sow's dietary treatment. However, considering the normal birth weight category, piglets from sows fed INT in late gestation had heavier birth weights (1.41 kg) compared to piglets from CON or HI (1.38 kg) sows. The advantage of heavier birth weights for INT pigs was maintained at weaning (5.58 kg), and they were heavier ($P < 0.05$) than HI pigs (5.46 kg), with CON intermediate to the two treatments (5.50 kg). As a result, pre-weaning growth performance of piglets tended to improve when sows consumed diets containing 365 ppm of supplemental Zn in late gestation, in contrast to performance of pigs from sows consuming diets with 595 ppm of supplemental Zn.

Pre-weaning Piglet Mortality

Overall pre-weaning mortality tended to decrease when sows were fed increasing levels of supplemental Zn in late gestation ($P < 0.10$; Table 4.8). Mortality of low birth weight pigs decreased by 10 percentage points (38.3% to 28.1%) when supplemental Zn was included at 595 ppm compared with 125 ppm ($P = 0.05$). Researchers that previously investigated the inclusion of high dietary Zn supplementation during late gestation obtained similar results to those obtained in our experiment, whereas pre-weaning mortality of low birth weight piglet was reduced (Vallet et al., 2014). Our results appear to confirm that high supplemental Zn in late gestation may play a role in enhanced survivability of small piglets.

Zinc is a structural component in many enzymes and metabolic pathways essential for healthy pregnancy. Superoxide dismutase, a Zn-dependent enzyme, provides antioxidant defense for the placenta (Mistry and Williams, 2011), which may have reduced activity during instances of pregnancy complications such as stillbirth. Furthermore, we know that Zn ions are present at the active site of carbonic anhydrase, which is necessary for transport and regulation of carbon dioxide (Tu et al., 2012). Researchers hypothesized that additional Zn may increase carbonic anhydrase activity which may, in turn, build resistance to high CO_2 concentrations during birth (Vallet et al., 2014), potentially improving the chance of survival for piglets that would otherwise be stillborn. Researchers and producers are aware that newborn piglets are iron deficient at birth (Ullrey et al., 1960; Matrone et al., 1960), but optimal Zn status assessment of piglets at birth has not been evaluated. Therefore, high supplemental Zn in late gestation may affect Zn status of low birth weight pigs such that chances of survival improve.

Not only did mortality of low birth weight piglets decline, but mortality of heavy birth weight pigs also decreased ($P < 0.10$). Although we hypothesized that survival of low birth weight pigs would improve, we did not expect to observe improvements in survival of heavy piglets, as well. Piglets born at the end of the litter are at greater risk for asphyxiation or oxygen deficiency. Uterine contractions towards the end of parturition reduce the supply of oxygen to the fetus (Alonso-Spilsbury et al., 2005). High supplemental Zn in late gestation may increase concentrations of metallothionein, an intracellular Zn binding protein, in red blood cells. Caulfield et al. (2008) suggested that increased erythrocyte concentrations of metallothionein might support rapid proliferation and differentiation of red blood cells and provide protection from oxidative stress that is

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typically associated with increased oxygen demand during pregnancy. These responses may play a role in mitigating some of the detrimental effects of reduced oxygen supply to the fetus, so that there might be less overall oxygen deprivation. Therefore, the pig may be better prepared for survival outside of the maternal environment.

Biomarkers such as plasma and bone, and growth performance can be used to assess Zn status of swine. Metallothionein may also be an indicator for assessing Zn status during pregnancy or in periods of stress or trauma (Golden, 1989; Caulfield et al., 2008). Investigating effects of high dietary Zn in late gestation on piglet and sow metallothionein concentrations throughout pregnancy and at birth, in addition to some of the other biomarkers mentioned, may guide scientists to answers regarding Zn status of piglets and the observed reduction of small and heavy birth weight piglet mortality.

Pre-weaning mortality for pigs included in this trial was 13.5% overall, which is similar to that observed in other commercial facilities (Feldpausch et al., 2016), and 34.1% for low birth weight piglets (Table 4.9). Mortality of low birth weight piglets was slightly greater than that observed in other commercial facilities, but was not unusual (Bergstrom et al., 2009; Zeng et al., 2019; unpublished data). Furthermore, post-weaning mortality was not different across treatments (Table. 4.10).

Carcass Characteristics

Late gestation dietary Zn treatments had no effect on carcass characteristics of pigs, regardless of birth weight category (Table 4.11). There were no significant interactions between gestational Zn treatments and birth weight categories for any post-weaning performance or carcass characteristics. No studies have fully investigated the effects of increased supplemental Zn in late gestation on lifetime performance of

offspring. Nonetheless, it appears that additional Zn in late gestation did not affect carcass characteristics such as hot carcass weight (HCW), backfat (BF) depth, loin depth, or calculated carcass lean percentage, fat-free lean percentage, and total lean gain/day of pigs. Pigs born at low birth weights often exhibit reduced carcass quality and value at harvest (Rehfeldt et al., 2008; Fix et al., 2010). However, lightweight pigs from this study performed similarly to those of normal birthweights, regardless of dietary treatment. Therefore, it appears that survival and lifetime performance of small piglets in this experiment did not compromise carcass composition or risk economic losses that one may expect to occur with lightweight pigs.

CONCLUSION

Results of this experiment indicate that increasing supplemental dietary Zn intake of sows in the last 35 days of gestation decreased overall piglet mortality, mortality of heavy birth weight piglets, and mortality of low birth weight piglets. Subsequent growth performance and carcass characteristics of low birth weight pigs were similar to pigs from dams that did not receive increased supplemental Zn in late gestation. Therefore, there may be substantial value in utilizing increased supplemental Zn in late-gestation sow diets to maximize piglets' chance of survival and lifetime growth. However, further research evaluating sources of supplemental Zn that minimize fecal excretion and total barn output of fecal Zn must be considered.

Table 4.1. Ingredient and nutrient composition of sow diets (as-fed basis)

Ingredient, %	Gestation	Lactation
Corn	49.20	52.90
Wheat middlings	15.00	-
Soybean meal	2.50	26.96
DDGS[1]	30.00	15.00
Choice white grease	-	1.00
Limestone	1.70	1.50
Monocalcium phosphate, 21% P	0.45	0.80
Salt	0.45	0.45
L-Lysine HCl	0.23	0.32
L-Threonine	-	0.10
Choline chloride 60%	0.14	0.05
Dyna K	-	0.63
Sow pack[2]	0.08	0.04
Premix[3]	0.25	0.25
TOTAL	100.00	100.00
Analyzed nutrient composition:		
Moisture, %	13.4	15.7
Crude protein, %	16.0	19.2
Crude fat, %	4.3	4.3
Crude fiber, %	3.6	2.1
Ash, %	6.0	8.0
Calcium, %	1.01	1.94
Phosphorus, %	0.63	0.60
Zinc total, ppm	184.6	255.9

[1]Dried distillers grains with solubles

[2]Contains the following: direct-fed microbial (DFM), mycotoxin binder, yeast culture, and carnitine

[3]Contained the following nutrients per kg of premix: vitamin A, 4,409,240 IU; vitamin D_3, 1,587,326 IU; vitamin E, 26,455 IU; menadione, 1,764 mg; riboflavin, 3,307 mg; niacin, 19,842 mg; pantothenic acid, 13,228 mg; pyridoxine, 5,732 mg; vitamin B_{12}, 15 mg; folic acid, 661 mg; biotin, 88 mg; phytase, 132,277 FTU; zinc, 110,231 ppm (60% as $ZnSO_4$, 40% as AvailaZn, Zinpro, Eden Prairie, MN) ; iron, 97,003 ppm; manganese, 35,274 mg; chromium, 176 ppm; copper, 14,550 ppm; iodine, 485 ppm; selenium, 265 ppm.

Table 4.2. Ingredient and zinc composition of top-dress (as-fed basis)

Ingredient, %	Top-dress
Corn	95.8
Choice white grease	1.0
Zinc sulfate, monohydrate	3.2
TOTAL	100.0
Analyzed composition:	
Zinc total, ppm	11,530

Table 4.3. Effect of supplemental Zn in late gestation on farrowing performance of sows

Item	CON[1]	INT[2]	HI[3]	SE	P-value
No. of sows	112	112	115	-	-
No. of litters	108	104	110	-	-
No. of piglets	1,565	1,424	1,525	-	-
Parity	2.9	3.0	2.9	0.2	0.92
Days on trial	35.8	36.0	36.1	0.4	0.70
Gestation length, d	115.2[a]	115.2[a]	115.6[b]	0.1	< 0.01
Lactation length, d	22.4	21.9	22.3	0.3	0.45
Days to service	6.9	7.1	5.8	0.9	0.43
Sows mated within 7d post-weaning[4], %	85.9	83.3	89.8	-	0.48
Body condition score[5]					
D79 Gestation[6]	14.9[a]	15.5[b]	15.5[b]	0.2	0.03
Pre-Farrow[7]	13.1	13.3	13.3	0.6	0.68
Weaning	11.4	11.8	11.8	0.4	0.32
Farrowing performance					
Total pigs born/litter	14.7	13.8	14.2	0.4	0.23
Pigs born alive/litter	14.0	13.1	13.4	0.4	0.25
Pigs weaned/litter	10.7	10.3	10.7	0.3	0.26

[ab]Means within a row with different superscripts differ ($P < 0.05$)

[1]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$

[2]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$

[3]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$

[4]Calculated as: (number of sows mated within 7 d of weaning / total sows at weaning) x 100; Chi square = 1.46, df = 2

[5]Body condition scores evaluated at last rib via caliper

[6]Initiation of dietary treatments

[7]One day before expected farrowing date

Table 4.4. Effect of supplemental Zn in late gestation on prevalence of lameness of sows

Item	CON[1]	INT[2]	HI[3]	Chi square[4]	P-value
D79 Gestation				0.49	0.78
Lame	1	1	2		
Not lame	113	114	114		
Pre-farrow[5]				2.02	0.36
Lame	2	1	0		
Not lame	104	106	106		
Weaning				1.88	0.39
Lame	2	0	1		
Not lame	99	94	96		

[1]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$
[2]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[3]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[4]df = 2
[5]Presence of lameness evaluated one day before expected farrowing date

Table 4.5. Effect of supplemental Zn in late gestation on total number of stillbirths and mummified piglets

| Item | Treatment | | | Chi square | P-value |
	CON[1]	INT[2]	HI[3]		
Stillbirths	76	68	78	8.56[4]	0.20
Mummies	50	49	44	6.37[5]	0.61

[1]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$
[2]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[3]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[4]df = 6
[5]df = 8

Table 4.6. Effect of supplemental Zn in late gestation on total number of low birth weight piglets

Item	Treatment			Chi square[4]	P-value
	CON[1]	INT[2]	HI[3]		
Low birth wt. ($\leq$ 1.00 kg)	240	165	231	10.78	< 0.01
Normal birth wt. ($\geq$ 1.01 kg)	1325	1259	1294		
Total pigs born	1565	1424	1525		
Incidence of low birth wt., %[5]	15.3	11.6	15.1		

[1]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$
[2]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[3]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[4]df = 2
[5]Calculated as (number of low birth wt. pigs / total pigs born) x 100

Table 4.7. Effect of supplemental Zn in late gestation on piglet performance

| | Treatment | | | | |
Item	CON[1]	INT[2]	HI[3]	SE	*P*-value
Overall					
Piglet birth wt., kg	1.38[a,x]	1.42[b]	1.40[ab,y]	< 0.01	< 0.01
Piglet gain, g/d	227.0	226.5	229.7	1.5	0.28
Piglet weaning wt., kg	5.52	5.59	5.51	0.03	0.14
Piglet age at weaning, d	18.2	18.1	18.1	< 0.1	0.44
Total piglet gain, g	4,100	4,140	4,080	30	0.23
Low birth wt. (≤ 1.00 kg)					
Piglet birth wt., kg	0.83	0.84	0.83	< 0.01	0.75
Piglet gain, g/d	187.3	190.1	187.9	3.6	0.86
Piglet weaning wt., kg	4.41	4.44	4.34	0.06	0.58
Piglet age at weaning, d	18.9	18.8	18.5	0.1	0.12
Total piglet gain, g	3,532	3,559	3,475	67	0.66
Normal birth wt. (1.01 to 1.75 kg)					
Piglet birth wt., kg	1.38[a]	1.41[b]	1.38[a]	< 0.01	< 0.01
Piglet gain, g/d	227.0	230.1	225.6	1.7	0.16
Piglet weaning wt., kg	5.50[ab]	5.58[a]	5.46[b]	0.03	0.02
Piglet age at weaning, d	18.2	18.2	18.1	< 0.1	0.42
Total piglet gain, g	4,112[xy]	4,168[x]	4,073[y]	31	0.09
Heavy birth wt. (≥ 1.76 kg)					
Piglet birth wt., kg	1.94	1.94	1.96	0.01	0.26
Piglet gain, g/d	255.9	250.2	254.9	3.9	0.58
Piglet weaning wt., kg	6.42	6.29	6.44	0.07	0.28
Piglet age at weaning, d	17.6	17.5	17.6	0.1	0.75
Total piglet gain, g	4,485	4,349	4,473	69	0.33

[ab]Means within a row with different superscripts differ (*P* < 0.05)
[xy]Means within a row with different superscripts differ (*P* < 0.10)
[1]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$
[2]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[3]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$

Table 4.8. Effect of supplemental Zn in late gestation on mortality of pigs by treatment and weight classification[1]

Item	Treatment			Chi square[5]	P-value
	CON[2]	INT[3]	HI[4]		
Piglet Mortality					
Overall				5.41	0.07
Dead[6]	235	188	186		
Alive[7]	1,330	1,236	1,339		
Total pigs	1,565	1,424	1,525		
Mortality, %	15.0	13.2	12.2		
Low birth wt. (≤ 1.00 kg)				5.94	0.05
Dead[6]	92	60	65		
Alive[7]	148	105	166		
Total pigs	240	165	231		
Mortality, %	38.3	36.4	28.1		
Normal birth wt. (1.01 to 1.75 kg)				0.11	0.94
Dead[6]	127	120	112		
Alive[7]	987	938	909		
Total pigs	1,114	1,058	1,021		
Mortality, %	11.4	11.3	11.0		
Heavy birth wt. (≥ 1.76 kg)					
Dead[6]	16	8	9	5.20	0.07
Alive[7]	195	193	264		
Total pigs	211	201	273		
Mortality, %	7.6	4.0	3.3		

[1]Data presented as counts of pigs
[2]Diets containing 125 ppm supplemental Zn as AvailaZinc and $ZnSO_4 \cdot H_2O$
[3]Diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[4]Diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[5]df = 2
[6]Represents dead pigs from birth to weaning; does not include stillborn pigs
[7]Represents live piglets from birth to weaning

Table 4.9. Pre-weaning mortality of pigs by weight classification[1]

Item	
Overall	
Total deaths[2]	609
Total alive[3]	3905
Total pigs	4514
Mortality, %	13.5
Low birth wt. ($\leq$ 1.00 kg)	
Total deaths[2]	217
Total alive[3]	419
Total pigs	636
Mortality, %	34.1
Normal birth wt. (1.01 to 1.75 kg)	
Total deaths[2]	359
Total alive[3]	2,834
Total pigs	3,193
Mortality, %	11.2
Heavy birth wt. ($\geq$ 1.76 kg)	
Total deaths[2]	33
Total alive[3]	652
Total pigs	685
Mortality, %	4.8

[1]Data presented as counts of pigs
[2]Represents dead pigs from birth to weaning; does not include stillborn pigs
[3]Represents live piglets from birth to weaning

Table 4.10. Effects of supplemental Zn in late gestation on post-weaning mortality of pigs[1]

Item	Treatment CON[2]	INT[3]	HI[4]	Chi-square[5]	P-value
Overall					
Dead[6]	13	11	11	0.46	0.80
Alive[7]	122	134	130		
Total pigs	135	145	141		
Mortality	9.6%	7.6%	7.8%		
Low birth wt. ($\leq$ 1.00 kg)					
Dead[6]	3	4	1	2.50	0.29
Alive[7]	26	23	31		
Total pigs	29	27	32		
Mortality	10.3%	14.8%	3.1%		
Normal birth wt. (1.01 to 1.75 kg)					
Dead[6]	7	6	8	1.48	0.48
Alive[7]	61	100	77		
Total pigs	68	106	85		
Mortality	10.3%	5.7%	9.4%		
Heavy birth wt. ($\geq$ 1.76 kg)					
Dead[6]	3	1	2	< 0.01	0.99
Alive[7]	35	11	22		
Total pigs	38	12	24		
Mortality	7.9%	8.3%	8.3%		

[1]Data presented as counts of pigs
[2]Offspring from sow diets containing 125 ppm supplemental Zn as AvailaZn and $ZnSO_4 \cdot H_2O$
[3]Offspring from sow diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[4]Offspring from sow diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$
[5]df = 2
[6]Represents piglets that died from weaning to market
[7]Represents live piglets from weaning to market

Table 4.11. Carcass characteristics of pigs from sows by treatment and weight classification

Item	Treatment CON[1]	INT[2]	HI[3]	SE	*P*-value
Overall					
No. of pigs	122	134	130	-	-
Wean to slaughter, d	168.6	168.3	167.1	1.1	0.59
Hot carcass weight, kg	99.9	101.2	100.1	0.6	0.19
Backfat depth, mm	16.3	16.3	16.0	0.4	0.74
Loin depth, cm	6.9[xy]	6.8[x]	7.0[y]	< 0.1	0.07
Lean[4], %	57.2	57.1	57.5	0.2	0.42
FFL[5], %	53.5	53.5	53.8	0.2	0.54
Lean gain[6], g/day	314.5	319.4	320.3	2.6	0.23
Low birth wt. (≤ 1.00 kg)					
No. of pigs	26	23	31	-	-
Wean to slaughter, d	174.0	169.5	171.8	2.1	0.41
Hot carcass weight, kg	97.7	99.8	98.4	1.3	0.57
Backfat depth, mm	15.7	15.9	16.7	0.7	0.55
Loin depth, cm	6.7	6.5	6.8	0.1	0.26
Lean[4], %	57.3	56.9	56.8	0.5	0.76
FFL[5], %	53.8	53.5	53.1	0.5	0.62
Lean gain[6], g/day	301.5	315.3	305.1	6.0	0.25
Normal birth wt. (1.01 to 1.75 kg)					
No. of pigs	61	100	77	-	-
Wean to slaughter, d	169.1	168.3	167.2	1.4	0.66
Hot carcass weight, kg	100.1	101.6	101.1	0.8	0.39
Backfat depth, mm	16.7	16.5	15.9	0.5	0.46
Loin depth, cm	7.0	6.9	7.1	0.1	0.35
Lean[4], %	57.1	57.1	57.7	0.3	0.31
FFL[5], %	53.4	53.4	53.9	0.3	0.35
Lean gain[6], g/day	313.7	319.8	323.5	4.8	0.13
Heavy birth wt. (≥ 1.76 kg)					
No. of pigs	35	11	22	-	-
Wean to slaughter, d	163.7	165.8	160.2	2.2	0.28
Hot carcass weight, kg	101.0	101.3	98.8	1.3	0.33
Backfat depth, mm	16.3	14.9	15.2	0.9	0.53
Loin depth, cm	7.0	6.7	7.2	0.1	0.19
Lean[4], %	57.3	57.8	58.2	0.6	0.55
FFL[5], %	53.5	54.2	54.3	0.4	0.47

Lean gain[6], g/day	325.3	324.2	330.1	4.3	0.74

[xy]Means within a row with different superscripts differ ($P < 0.10$)

[1]Offspring from sow diets containing 125 ppm supplemental Zn as AvailaZn and $ZnSO_4 \cdot H_2O$

[2]Offspring from sow diets containing 365 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$

[3]Offspring from sow diets containing 595 ppm supplemental Zn as Control + $ZnSO_4 \cdot H_2O$

[4]Lean calculated as: $58.86 - 0.61 \times$ (backfat depth) $+ 0.12 \times$ (loin depth); JBS Pork

[5]Fat-free lean calculated as: $[15.31 - (31.277 \times \text{backfat depth}) + (3.813 \times \text{loin depth}) + (0.51 \times \text{HCW})] / \text{HCW} \times 100$; NPPC (2000)

[6]Lean gain calculated as: (FFL at ending weight - FFL in feeder pig) / days on test; NPPC (2000)

CHAPTER 5: OVERALL SUMMARY

Commercial sow production and subsequent piglet output is steadily expanding in the United States. Although litter sizes for commercial sows are increasing, pre-weaning mortality within production facilities is increasing, so that total barn output measured as piglets weaned per litter, is not improving as rapidly. Major causes for piglet mortality, especially in the initial days of life, include stillbirth, low viability, trauma from crushing, and starvation. Furthermore, birth weight variation of piglets from sows that produce such large litter sizes has increased, yielding greater number of low birth weight (< 1.00 kg) piglets born within litters. Until birth weight variation within piglet litters is controlled, it is essential to investigate methods to reduce high rates of pre-weaning mortality experienced by small piglets in commercial systems.

Zinc is an essential trace mineral for optimal reproductive performance of sows and the subsequent growth and development of piglets. However, available literature regarding dietary Zn requirements, digestibility, and bioavailability for modern reproducing swine is limited. Furthermore, effects of dietary antagonists such as phytate, fiber, and other mineral interactions complicate Zn absorption and utilization. Even more, there are discrepancies regarding classes of dietary Zn sources. As a result, industry nutritionists may not accurately formulate gestation and lactation diets and provide excess supplemental Zn in diets to provide a safety margin. This practice not only increases diet costs, but also risks excessive Zn output in manure that is applied later to cropland.

Results obtained from the experiment in Chapter 3 suggest that diets high in fiber, provided by DDGS, reduced ATTD and TTTD of Zn during lactation, but not in

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gestation. Apparent total tract digestibility and TTTD of Zn, regardless of Zn source, improved when gestating sows consumed DDGS diets high in fiber content. Additionally, gestating sows fed DDGS diets supplemented with organic Zn exhibited improved ATTD, TTTD, and overall retention of Zn, compared with sows fed diets based on corn and soybean meal. However, lactating sows fed DDGS diets supplemented with organic Zn exhibited reduced ATTD, TTTD, and overall retention of Zn, compared with sows fed corn-soybean meal diets or diets supplemented with inorganic Zn. There were no effects of supplemental Zn source, diet, or the interaction of Zn source and diet on overall Zn status of sows throughout the trial.

Furthermore, Zn excretion surpassed Zn intake for sows consuming diets without supplemental Zn in gestation. Alternatively, sows in lactation exhibited positive Zn retention, indicating that current NRC (2012) lactation Zn requirements are adequate. However, to truly investigate dietary Zn requirements of gestating and lactating sows, further research evaluating true endogenous Zn losses must be conducted.

Results obtained from the experiment in Chapter 4 indicate that increasing supplemental dietary Zn in late gestation positively influenced survival of piglets. Overall piglet mortality, mortality of heavy birth weight piglets, and mortality of low birth weight piglets decreased. Even more, subsequent growth and carcass performance of low birth weight piglets were not different than piglets from dams that did not receive high dietary Zn in the last 35 d of gestation. It appears that increased concentrations of supplemental Zn in diets for late-gestating sows maximizes survival of piglets, without compromising lifetime performance.

In conclusion, dietary Zn in gestating and lactating sow diets remains essential. Results from these experiments indicate that stage of the reproductive cycle, Zn source, and diet composition heavily influence digestibility, utilization, and retention of Zn. Additionally, further investigation of the true Zn requirements for modern commercial sows is still necessary to optimize sow performance and minimize fecal excretion of Zn. Answers from such investigations will guide producers to implement accurately formulated diets to meet reproductive demands of gestating and lactating sows. Strategic formulation of diets to include high concentrations of supplemental Zn in late gestation may reduce mortality of low and heavy birth weight piglets, as well as improve overall piglet survival. Although, further investigation of the biological mechanisms responsible for this reduction in mortality must be conducted.

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