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EVALUATING THE EFFECTS OF MEDIUM CHAIN FATTY ACIDS ON SOW
REPRODUCTIVE CHARACTERISTICS, OFFSPRING BIOLOGICAL
HEALTH MARKERS, AND GROWTH PERFORMANCE

BY

ANALICIA J. SWANSON

A thesis submitted in partial fulfillment of the requirements for the

Master of Science

Major in Animal Science

South Dakota State University

2022

THESIS ACCEPTANCE PAGE

Analia J. Swanson

This thesis is approved as a creditable and independent investigation by a candidate for the master's degree and is acceptable for meeting the thesis requirements for this degree.

Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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ABBREVIATIONS

AA	Amino acid
ADFI	Average daily feed intake
ADG	Average daily gain
BF	Backfat
BW	Body weight
CP	crude protein
d	day/days
g	Grams
G:F	Grain to feed ratio
GI	gastrointestinal
h	hour
i.m.	Intramuscular
IgA	Immunoglobulin A
IgG	Immunoglobulin G
IgM	Immunoglobulin M

Kg	Kilogram
lbs	pounds
m	Meter
MCFA	Medium chain fatty acid
MCT	medium chain triglyceride
mL	Milliliter
mm	Millimeter
n	Number
ppm	Parts per million
SAS	Statistical analysis system
SUP	supplemented dietary treatment
TRT	dietary treatment
UNSUP	control dietary treatment
μg	microgram
VH:CD	Villus height: crypt depth

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ABSTRACT

EVALUATING THE EFFECTS OF MEDIUM CHAIN FATTY ACIDS ON SOW
REPRODUCTIVE CHARACTERISTICS, OFFSPRING BIOLOGICAL HEALTH MARKERS, AND
GROWTH PERFORMANCE

ANALICIA J. SWANSON

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A sow's diet can influence and her offspring not only in the suckling period, but well after weaning. The objective of this study was to determine how supplementing medium chain fatty acids (MCFA) in gestation and lactation impacted offspring growth performance as well as the biological markers of pig quality from the suckling period until week 6 of the nursery period. A total of 77 primiparous and multiparous females and their 438 offspring were utilized in this study. Sows and gilts ($218.15 \text{ kg} \pm 32.15 \text{ kg}$ BW at d28 of gestation) were assigned to one of 2 dietary treatments: Control (UNSUP) and control + MCFA (SUP). At weaning, 438 piglets ($5.66 \pm 1.37 \text{ kg}$ BW) were allocated in a 2x2 factorial based on maternal diet (UNSUP or SUP) and post-weaning diet (UNSUP_{nurs} or SUP_{nurs}) in a 3-phase nursery pig feeding program lasting 42 days. Following birth of the first piglet and prior to suckling, colostrum was collected using gentle stripping from all teats for a total volume of 40 mL and at d 4 or 5 of lactation, a milk sample was collected. Microbial analysis of piglet fecal samples at d 10, d 24, and d 63 of age were completed to determine the relative proportion of *Lactobacillus*, *Escherichia coli*, and *Salmonella*. Colostrum and milk samples were analyzed for protein, lactose, total solids, and fat (Division of Regulatory Services, University of Kentucky, Lexington, KY). At weaning, one of the "average" piglets from the selected litters was

ethanized, and 10 cm of the ileum was removed and placed in a 15 mL conical tube containing 5 mL of 10% buffered formalin solution for histology analysis. Suckling piglets were weighed at d7 of age and at weaning. In the nursery period, pigs were weighed, and feed disappearance was measured at week 1 (for study 2), 2, 4 and 6. No effect of maternal diet was observed for sow BW at d 110 (228.63 ± 3.12 kg) or weaning (211.92 ± 3.15 kg), piglet birth weight (1.38 ± 0.05 kg), piglet wean weight (5.74 ± 0.11 kg), or litter size (14.40 ± 0.44). The SUP sows had greater daily feed intake ($P < 0.05$) compared to the sows fed the control diet. In the nursery phase there was no maternal diet supplementation effect for all measured response variables and no effect of nursery dietary treatment. At birth, UNSUP litters had a larger percentage of average piglets (69.75% UNSUP, 59.02% SUP), a lesser percentage of light piglets (15.41% UNSUP, 19.67%) and a lesser percentage of heavy weight piglets (14.84% UNSUP, 21.31% SUP; $\chi^2 < 0.01$). At weaning, UNSUP sows tended to have a larger percentage of average piglets (71.68% UNSUP, 66.60% SUP), a lesser percentage of light piglets (13.49% UNSUP, 18.24% SUP) and they had lesser heavy weight birth piglets (14.84% UNSUP, 15.16% SUP; $\chi^2 = 0.10$). Inclusion of MCFA in gestation and lactation increased sow lactation daily feed intake. Colostral fat content was not impacted by maternal dietary treatment ($P = 0.70$). Colostral protein content increased ($P = 0.04$) in SUP sows compared to UNSUP. Colostral lactose content in UNSUP sows tended to increase ($P = 0.06$) compared to SUP sows. UNSUP sows tended ($P = 0.07$) to have decreased colostrum total solids and decreased ($P = 0.04$) colostrum solids not fat content compared to SUP sows. Milk composition was not affected by dietary treatment. Similarly, colostrum immunocrit was not altered by maternal dietary treatment. Serum immunocrit was

decreased ($P = 0.01$) in SUP piglet serum compared to UNSUP piglet serum. Colostral IgG as well as milk IgG was not impacted by supplementation of MCFA in the maternal diet. Similarly, villus height, crypt depth, and their ratio at weaning was also not impacted by maternal dietary treatment. The *Lactobacillus* content from the suckling period (d 10 of age) tended to be a greater population ($P = 0.08$) in SUP piglets compared to UNSUP piglets. In the nursery period (39% and 19%) SUP pigs more respectively maintained the slightly greater proportion of *Lactobacillus* compared to UNSUP pigs. The addition of MCFA in the maternal dietary treatment improved feed intake, biological markers of the piglets such as colostrum quality and the gut microbiome in the suckling and nursery periods. In lactation, feed intake is a key limiting factor for milk output, thus MCFA may contribute to improved sow milk output. Colostral proteins is predominately made up of immunoglobulins, which aid in providing the piglet passive immunity. Increasing colostrum proteins could increase immunoglobulins for the piglet. *Lactobacillus* is known as a “good” bacterium and can indicate a healthy pig. Increasing the abundance of *Lactobacillus* in the gut, especially at weaning, can be a mechanism to ensure pigs get off to the proper start and grow in those first few weeks in the nursery.

CHAPTER 1

1.0 LITERATURE REVIEW

1.1 Modern Sow Production

Genetic selection and improvements in management, health, and nutrition have led to a dramatic increase in sow productivity. In 2019, pigs weaned per sow per year averaged 27.91 in the United States, with an even higher productivity in some of the major pork producing countries ranging from 28.09 in Brazil to 33.60 in Denmark (Agriculture and Horticulture Development Board, 2019).

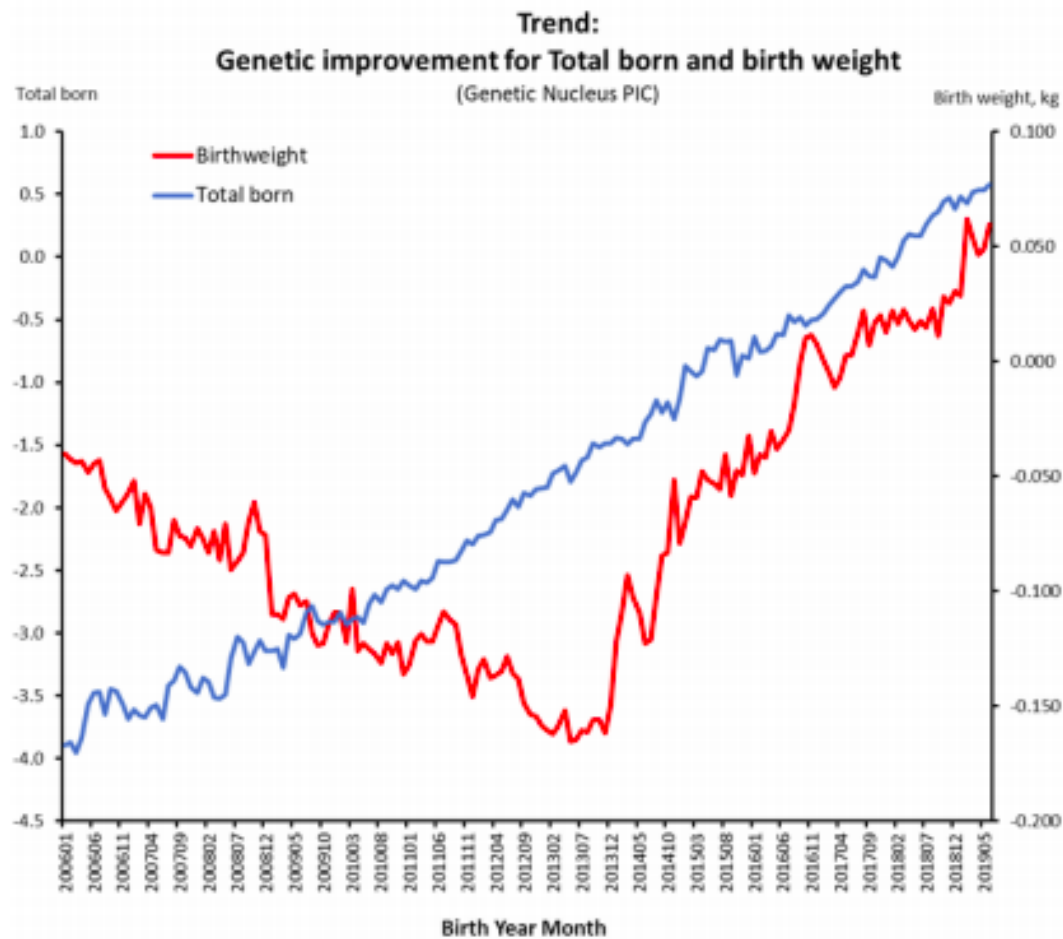


Figure 1.1 Genetic improvement trend for total pigs born and average pig birth weight at the nucleus level from Genus PIC (Case and Bloemhof, 2019).

Figure 1 from Genus PIC demonstrates that the genetic trend at the nucleus level for total pigs born has changed dramatically and that much of the increased number of pigs weaned per sow is a result of increased litter size. From the year 2006 to 2019, the trend for total born has steadily increased, resulting in an additional 1.5 pigs for total born per litter. This improvement in the total born initially led to a decrease in birth weight, as depicted in the figure above, and survivability; selection emphasis on piglet quality (i.e. alive at day 5; Case and Bloemhof, 2019) has reversed that trend at the nucleus herd level. The improvements that genetic selection has made has increased the metabolic demands on the highly prolific sow throughout gestation and lactation. This highly prolific sow is growing at a faster rate with lesser body fat and more lean body tissue (Tokach et al., 2019). These changes involving body composition and reproductive performance alters the nutrient demands.

1.2 Sow Longevity

A key component for efficient and profitable pig farming is sow longevity; however, the sow culling rate has steadily increased in recent years, suggesting that sow longevity is declining (Farmer 2015). A high percentage of sows in commercial breeding herds are being replaced after producing only one or two litters, a point at which the sows have yet to reach their maximum productivity or their replacement costs (Carroll, 2011; Stalder et al., 2003). These high removal rates are concerning for the swine industry and may indicate an animal welfare issue (Barnett et al., 2001). Approximately 70% of sow removals are the result of premature and unplanned culling mainly due to reproductive failure or leg issues (Engblom et al., 2011). In particular, early culling and high removal rates due to lameness can be associated with animal welfare (Engblom et al., 2007).

Lameness is often linked with pain and distress in the sow (Le et al., 2016). In the US, approximately 54% of sows are removed annually and the average parity at removal ranges from 3.3 to 3.8 (D’Allaire et al., 1987; Lucia et al., 2000). Recently, annual culling rates are between 35.7 and 49.5% in the USA, Spain, Sweden, and Japan (Tani et al., 2018). These early culling are detrimental to profitability as breeding females should be retained in the herd until at least the initial investment cost has been recovered (Farmer 2015). Stalder et al. (2003) and Carroll (2011) suggest that sows should remain in the breeding herd for at least three parities to have a positive return on investment; however, they also advise that the number of parities required to recoup the initial investment costs varies between farm as well as production system. Sehested (1996) stated that improving sow longevity by a single parity showed the same economic impact as improving all pork lean meat content by 0.5%; however, he also reported that the single parity increase had little effect beyond parity five. To put context to the value of one extra parity; in 2020, approximately 28.3 billion lbs of pork was produced in the US (Shahbandeh, 2021). An increase of 0.5% would be equivalent to 141,500,000 lbs pork at a value of \$0.7985/lb (01/09/2022

<https://www.cmegroup.com/markets/agriculture/livestock/lean-hogs.html#>)

that increase in pork is worth just under \$112 million. Sow longevity plays an important role in pig production systems. For example, the length of the sow’s productive life is directly correlated to the number of piglets produced during a sow’s lifetime (Farmer 2015). As summarized by Farmer (2015) the number of pigs produced per litter from first parity animals is less than the number of pigs produced by older sows. As well, early removal of sows from the herd results in higher infertility rates, less litters produced per

sow, lower mean litter size, lower number of pigs weaned per sow per year, and greater non-productive days (Anil et al., 2009; D'Allaire and Drolet, 1999; D'Allaire et al., 1987; Dourmad et al., 1994; Engblom et al., 2007; Friendship et al., 1986). Smits (2011) reported that piglets from first parity sows are lighter at birth and at weaning and have higher mortality risk and disease susceptibility compared with those piglets that are produced from older parity sows. When sows are culled, replacement gilts are needed to replace them. King et al. (1998) also reported that as the percentage of gilts in the breeding herd inventory increased by 1%, the average number of non-productive days increased by 2.6 days. Furthermore, the sow herd accounts for approximately 20% of feed expenses in a commercial farrow to finish operation, low removal rates could have a small direct effect on herd feed conversion due to changes in feed consumption because replacement gilts must be maintained 6-12 weeks before their first mating (Smits, 2011). When producers increase replacement rates, more gilts are needed to maintain breeding numbers, herd feed conversion (feed consumed per unit of carcass weight produced) will be increased or less efficient due to the increased amount of feed needed before breeding as well as the reduced output of pigs per year for gilts compared to sows (Smits 2011). It is important to be able to identify and to understand different factors and/or practices that impact sow longevity in order to increase the time a sow stays in the breeding herd and to increase farm profitability (Farmer 2015).

1.3 Importance of Parity Distribution in Relation to Sow Longevity

There is no clear definition of sow longevity in scientific literature, but it is usually referred to as the length of productive lifetime compared to the sow's lifespan (Farmer 2015). The parity distribution of a breeding herd will give an indication when sows are

removed from the herd and parity distribution will influence herd biological and economic performance (Farmer 2015). An optimal parity distribution is essential to maintain production performance and to minimize fluctuations in numbers of replacement gilts needed (Farmer 2015). Some incentives to have a parity distribution with sows of higher parities are larger litter sizes with heavier pigs from sows of older parities, fewer unproductive days, higher sow retention rates, lower replacement cost, increase in gross income as older sows have an increase in piglets born alive (D'Allaire et al., 1987; Farmer 2015; Stalder et al., 2000, 2003). Pinilla and Lecznieski (2014) created a definition of an 'ideal' parity distribution viewing it like a mathematical equation incorporating sow removal rate, gilt availability, current market prices, and feed costs. According to D'Allaire et al. (2012), the 'ideal' parity distribution for a breeding herd is based upon the maximum number of parities that sows are allowed before automatic culling occurs and annual culling rate, once these two values are known, the distribution percentage of sows culled in each parity is a linear function. Having an ideal parity distribution is critical to maintain consistency in performance as well as to avoid severe fluctuations in the number of replacement gilts (Farmer 2015). Table 1 presents the ideal parity distribution of a sow herd as reported by several researchers (Farmer 2015). Looking further into each paper that is listed in Table 1.1, it is clear to see that more attention is needed to aid in decreasing the cull rates of younger females. Morrison et al. (2002) used a push model to answer the question "what is my optimal parity distribution". The model was designed to flow in gilts at a steady rate to get an optimal parity distribution (Morrison et al., 2002). The model investigates factors that drive parity distribution, those factors being litter size, gilt prices, retention rate (low, medium, high),

sow cull value, and sow mortality on herd net income (Morrison et al., 2002). The purpose of the model was to determine the economically optimum distribution for an individual herd (Morrison et al., 2002). Morrison et al. (2002) helped many producers make sound culling decisions such as the optimal time to cull. The numbers produced for Morrison et al. (2002) below in Table 1.1 showed the optimized proposed distribution based on the factors mentioned above. Other variables play a role in culling rates, such as personal philosophies or research purposes. An example of a personal philosophy is having a high herd replacement rate because of the genetic improvements that come with the new replacement gilts. The SDSU sow herd is an example where parity distribution is a balance between reproductive performance and research needs. In this herd, sows are culled after parity 5 regardless of past performance. This helps to maintain a reasonably narrow parity structure which is important for the research trials conducted there.

Parity distribution plays a crucial role with herd performance and economic profitability. Establishing a replacement rate should be based on the goal's set for that sow farm. Taking advantage of the models mentioned above and the variables that factor into them, will help aid in determining an optimal parity distribution for a given herd.

Table 1.1 Recommended parity distribution (expressed in percentage of sows/parity)¹

Reference	Parity								
	0	1	2	3	4	5	6	7	8
Straw (1984)	20	18	17	16	15	14	-	-	-
Parson <i>et al.</i> (1990)	30	23	19	14	10	5	2	1	-
Muirhead and Alexander (1997)	17	15	14	13	12	11	10	5	3
Caroll (1999)	-	17	16	15	14	13	11	10	4
Morrison <i>et al.</i> (2002)	19.1	16.5	16.9	14.1	10.2	8.2	5.1	4.9	4.9
Pinilla and Leczneski (2014)	20	17	16	15	14	13	5	-	-

¹As reported by Farmer (2015)

1.4 Reasonings for Culling

Sow cullings can be classified into two categories: voluntary and involuntary.

Voluntary is when management makes a decision to remove a female from the herd. An example of this is SDSU practice of maximum parity of 5 based on research and teaching objectives rather than reproductive performance. Involuntary is when females must be removed due reproductive failure, lactation failure, or locomotion problems (Farmer 2015). The most frequent involuntary reason is reproductive failure (Farmer 2015), which accounts for roughly 20 – 43% (Boyle et al., 1998; D’Allaire et al., 1987; Dijkhuizen et al., 1989; Engblom et al., 2007; Hughes et al., 2010; Lucia et al., 2000).

Reproductive failure includes the failure to cycle, conceive, or farrow (Farmer 2015). Interestingly, sows that are removed for reproductive failure have lower numbers of piglets born alive per year and more non-reproductive days compared to sows that are culled for other reasonings (Sasaki and Koketsu 2011). Poor litter performance (made up of small litters born/weaned, low birth/wean weight, udder defaults, or poor mothering abilities) account for an average of 20-30% of culled sows (Boyle et al., 1998; D’Allaire et al., 1987; Dijkhuizen et al., 1989; Engblom et al., 2007; Hughes et al., 2010; Lucia et al., 2000; Tarrés et al., 2006). Feet and leg problems are second largest removal reason reported for young sows (Boyle et al., 1998; Engblom et al., 2007; Lucia et al., 2000), which makes feet and leg problems a major contributor to involuntary sow culling. Feet and leg unsoundness cause financial losses to the swine industry due to extra labor required for surveillance and treatment, veterinary treatment costs, reduced reproductive performance, and partial or total carcass rejections (Farmer 2015). Pluym et al. (2011) noted that severely lame sows that are euthanized on the farm implies a loss of slaughter

revenue and extra cost for euthanasia and/or destruction of the carcass. Interestingly, Grandjot (2007) reported that sows that were culled due to lameness produced 1.5 litters less than sows that were not lame throughout their productive lifetime. Feet and leg defects could also impair welfare because sows with severe conformation problems could have limited access to feed and water because of discomfort while standing or moving and be forced to modify normal behavior due to the limitations of movement (Fernández de Sevilla et al., 2008).

Diseases such as enteropathy, porcine reproductive and respiratory syndrome, pseudorabies, porcine enterovirus, swine influenza, chronic erysipelas, acute hemorrhagic ileitis, have become very common in sow herd (Loula, 2000; Stalder et al., 2004) and also contribute to reduced sow longevity. These diseases negatively impact length of the productive life cycle of sows due to some pathogens causing sows to abort and then producers face the decision of whether to cull these females or to retain them and take a chance on their ability to rebreed (Farmer 2015).

1.5 Gilts vs Sows on Farm Productivity

When a farm experiences high removal rates, non-productive days are increased which in turn increases the number of gilts in the herd, resulting in lower mean litter size and lower number of pigs weaned per sow per year (Hughes and Varley, 2003).

Replacement gilts bring a concern for passive immunity as well maintaining overall good health status for a herd. The introduction of replacement gilts into the breeding herd may pose many health challenges and risks among the existing females due to their immature immune system (Sanz et al., 2002). Sow farms that have a larger percentage of their breeding females as gilts experience more challenges with *Streptococcus suis*,

Actinobacillus suis, *Haemophilus parasuis*, *Mycoplasma hyopneumonia*, *Staphylococcus hyicus* and *Pasteurella* organisms (Sanz et al., 2002). Regardless of where replacement gilts originate from (i.e. farm source), isolation and acclimation are essential for the long-term productivity of the gilt in that breeding herd (Farmer 2015). Healthy gilts may appear to be healthy; however, they could have incubating infections or be a carrier of pathogens (Farmer 2015). Gruhot et al., (2017) looked at the economics of sow retention in the US and discovered that although older parity females have smaller born alive numbers in comparison to younger parities, they wean a greater percentage of their pigs. Retaining sows until later parities has economic potential over culling sows after parities 1-4, indicating a higher return over total cost (Gruhot et al.,2017). Although older sows produce and sell fewer pigs per year than younger parities, the cost associated with producing a weaned pig is lowest when sows are culled after parities 5-9 (Gruhot et al., 2017).

However, introduction of replacement gilts into a sow herd is inevitable and a critical component of the overall production cycle that needs attention. It has been argued that replacement gilts are the foundation of a production system's efficient breeding program (Ketchum and Rix 2006). It is these replacement gilts that will become the older parity sows so the more effectively they are 'brought into' production the more likely they will be retained within the herd (Nikkilä et al., 2013). The successful introduction of high-quality eligible breeding gilts into the herd is frequently underestimated as an important tool for sow lifetime longevity (Patterson and Foxcroft, 2019). A proper gilt management program addresses several key components such as: birth traits that determine the efficiency of replacement gilts, effective selection of the most fertile females,

management programs that provide a consistent supply of eligible gilts within the appropriate weight, physiological maturity, and positive metabolic state at the time of breeding (Patterson and Foxcroft, 2019). Execution of a breeding management program that recognizes the link between proper gilt management and sow lifetime productivity is achievable and cost effective (Patterson and Foxcroft, 2019). When a system evaluates sow cullings, the hidden factors mentioned above, and the economics of those factors, need to be considered.

1.6 Sow Feeding Program and Longevity

Nutritional methods can be utilized to manipulate sow longevity (Farmer 2015). Examples of this can be decreasing amino acid (AA) intake to reduce lean to fat ratio or increase dietary vitamins and trace minerals that are needed for bone structure and tissue development (Farmer 2015; Kitt 2010). Backfat thickness is a reflection of the total fat content of the sow (Mullan and Williams 1990), and backfat thickness is described as an objective indicator of body condition of sows (Charette et al., 1996). Excessive loss of body protein and backfat during late gestation and throughout lactation is correlated with an increased percentage of stillborn piglets (Maes et al., 2004) and reduced litter sizes and decreased growth (McKay 1993; Clowes et al., 2003). It also correlates to sow longevity by resulting in a prolonged wean-to-estrus interval, declines in conception rates, ultimately resulting in a shorter productive lifetime (De Rensis et al., 2005; Serenius et al., 2006). After confirmation of pregnancy, the feeding program is based on individual sow body condition which is an indirect measure of sow body composition. Body condition is closely monitored, if excessive the sow could experience dystocia and further issues throughout lactation. Numerous studies have indicated that sows with

higher backfat at farrowing will have lower feed intake during lactation compared to sows with lower backfat at farrowing (Tokach et al., 2019). If a sow is under conditioned in gestation, detrimental consequences could be shown in the subsequent reproductive cycle. An example of this is an extended wean-to-estrus interval (De Rensis et al., 2005) or reduced litter size and growth (McKay 1993; Clowes et al., 2003). Therefore, gestation diets need to control body weight gain and also supply enough nutrients to support sow body maintenance, fetal growth and development, as well as mammary development. Gestating females are limit fed ranging from 1.8 kg to 2.3 kg of feed per day to monitor sow body condition (Shannon, 2011). Sows need to maintain optimal backfat thickness and body condition in the latter part of gestation and into lactation to sustain subsequent reproductive performance (Tummaruk et al., 2000, 2001, 2007; Houde et al., 2010). Energy is demanded for development and growth of the fetus, placenta, uterus, and mammary tissues, as well as the deposition of lipids and proteins (NRC 2012). When an increase in energy intake has occurred in late gestation, it can positively affect the fetal growth as well as maternal weight gain; however, there are often problems associated with it, such as, negatively impacting the subsequent lactation period (NRC 2012). Additionally, if feed intake in gestation is increased, it can be correlated to reduced energy intake as well as greater sow body weight loss during that subsequent lactation period (Williams 1985; Weldon et al., 1994). The weight a female gains in gestation is a result of maternal protein and lipid deposition and conceptus gain (NRC 2012). Mineral supplementation is vital for aiding in the development of soft and hard tissues (Farmer 2015). Some of the minerals of particular importance include calcium, phosphorus, magnesium, manganese, zinc, and copper. Calcium is needed to achieve growth,

development, and maintenance of the skeleton (Farmer 2015). Phosphorus is also critical for bone formation and mineralization of the skeleton (Crenshaw 2001). Magnesium aids in bone strength and integrity and nerve transmission (Patiente and Zijlstra 2001). Zinc contributes to growth, development, reproduction and metabolic activity (Hill and Spears 2001). Manganese is needed for fertility and growth (Underwood and Suttle, 1999).

Gilt body composition made up of backfat thickness, loin depth, and body weight at the time of the first breeding can affect sow longevity (Farmer 2015). In order to maximize a gilt's lifetime productivity, she needs to build up body reserves that will allow for a long reproductive life (Farmer 2015). The key to success for gilt development is stemmed by slowing down protein deposition and building fat, mineral, and other nutritional reserves that can be utilized in the first lactation when nutrient need is not being met by feed intake (Stalder et al., 2007). Gilts with backfat thickness >18 mm in late gestation, remained in the herd until at least the fourth parity compared to leaner gilts with backfat <10 mm (Brisbane and Chenais 1996). However, gilts at the time of their first farrow who have a backfat thickness >19 mm are at greater risk of culling (Farmer 2015). Although lactation only represents 15 to 20% of the sow's reproductive cycle, it is the most metabolically demanding stage of production (Tokach et al., 2019). During lactation, the priority of the sow is to sustain milk production for the large and fast-growing litter of piglets, but nutrients required to meet milk production are not often attained by voluntary feed intake (Tokach et al., 2019). This limitation is noted above, when addressing sows losing backfat and body weight in lactation and that negatively impacting her subsequent reproductive performance and ultimately sow lifetime productivity. The mobilization of body fat and protein reserves is considered critical to

support milk production, factoring in the size of her litter, her previous milk performance, and body condition, in high-producing sows. Lactating sows utilize up to 70% of their dietary crude protein for milk protein synthesis (Pedersen et al., 2016), when there is insufficient dietary crude protein milk yield may be suppressed which can cause substantial body weight loss and backfat (Strathe et al., 2017a). However, there is question as to whether sow tissue mobilization is an obligatory process or not (Pedersen et al., 2019). This idea of obligatory tissue mobilization could impact measure of lactating sow feed efficiency, where sow feed efficiency is based on the female's productivity from energy directly from the diet instead of the energy, she receives from body stores (Pederson et al., 2019). Pederson et al. (2019) also points out that lactation diet formulation should account for the fact that sows utilize dietary energy more efficiently for milk output compared to growing pigs using it for whole body energy retention.

The energy requirement of the modern lactating sow has substantially increased with the increased number of piglets that nurse (Tokach et al., 2019). When the energy requirement for lactation surpasses energy intake, lactating sows will mobilize body fat and protein (Strathe et al., 2017b). It is known that energy mobilization is exponentially higher in the modern high-yielding sows (Strathe et al., 2017b). Milk production represents 65% to 80% of the energy requirement of lactating sows (NRC 2012). The energy concentration of lactation diets is an important determinant of energy consumption and is typically modified by the inclusion of fats, oils, or fibers in the diet (Tokach et al., 2019). An increase in dietary energy can have both positive and negative effects on sow lactation feed intake (Xue et al., 2012). For example, dietary energy

concentrations of 12.8 to 13.4 MJ ME/kg in the lactation diet can increase total sow energy intake and lessen weight loss and increase litter growth (Xue et., 2012). However, there is a threshold where if dietary energy is 13.8 to 14.2 MJ ME/kg it can result in decreased feed intake and energy intake (Xue et al., 2012). Literature shows that even when additional energy is supplemented in lactation, sows will still prioritize her lactation needs and partition the energy for milk and milk fat output (Rosero et al., 2015). Changes in the energy balance during lactation can have long-term effects on sow reproduction and longevity (Dourmad et al., 1994). The energy requirement for the lactating sow are characterized by metabolizable energy intake for body maintenance and milk production (NRC 2012). Although lactation feed intake is *ad libitum*, energy intake is often not sufficient to support milk production (NRC 2012).

Amino acid requirements are driven by the need for milk production and sows will utilize as much as 70% of dietary protein for milk synthesis (Pedersen et al., 2016). When sows experience excessive body protein mobilization in lactation, this can decrease subsequent litter size due to reduced follicular development (Clowes et al., 2003). Gourley et al. (2017) discovered that increasing dietary AA's led to the reduction of protein loss in lactation. Reducing protein mobilization in lactation improves reproductive cycling which ultimately improves sow longevity and productivity. Strathe et al. (2017a) discovered that increasing dietary standardized ileal digestible (SID) crude protein (CP) to 850 grams/day improved litter ADG by increasing the milk protein content and milk yield. They also report that this increase in SID of CP, body weight was minimized and that the subsequent reproductive cycle was not negatively impacted (Strathe et al., 2017a). Tokach et al. (2019) state lactation diets need a minimum

inclusion of 13.5% to 14.3% digestible CP. When the supply of AA and CP are close to the sow's requirements, milk protein output is increased and muscle protein mobilization are minimized (Strathe et al., 2017a; Gourley et al., 2017; Pedersen et al., 2019). When the number of suckling piglets increase per sow, the essential AA requirements in milk and mammary tissue increase as well (Kim et al., 2001). The first 3 limiting essential AA for milk production, based upon a corn-soybean meal lactation diet, are lysine, threonine, and valine (Kim et al., 2001; Soltwedel et al., 2006). The AA profile in milk and mammary gland tissue becomes a major factor to influence the ideal AA profile for lactating sows (Kim et al., 2001). Mobilization of large amounts of AA from sow body tissues can influence the AA profile required in the diet composition (Kim et al., 2001). Similarly, sows that experience minimal body tissue loss result in a different order of AA depletion (Kim et al., 2001).

Understanding what drives milk production and limits sow body weight loss and backfat thickness is critical for sow longevity. Further, altering colostrum protein composition via the sow diet can impact the development of the gut of the piglet (Theil et al., 2014).

1.7 Development of the Gut in Piglets

During gestation, prenatal gut development acquires structural and functional competencies that helps the piglet for life after birth (Farmer 2015). Gut development starts early on in a piglet's life, starting during the fetal period and continuing through the first months after birth (Farmer 2015). Prior to birth, the gut of the neonate is known to be void of microbes (Guevarra 2019). The gastrointestinal tract (GI) is not only an important organ for digestion, absorption, and metabolism of dietary nutrients, but it is

also the largest immune organ in the body (Liu 2015). In swine production, pigs encounter numerous pathogenic and nonpathogenic challenges, which results in activation of the GI immune system (Liu 2015). With recent transformational changes in molecular technologies and strategies, the pig gastrointestinal microbiota and the roles the gut microbiota plays with health and well-being of the animals has been more intensively studied (Guevarra 2019; Kim and Issacson 2015). The gut microbiota provides the pig improved energy harvesting capacity, the production of volatile fatty acids, and enhanced resistance against pathogenic bacteria (Guevarra 2019; Kim and Issacson 2015; Stokes 2017; Yang et al., 2017). Normal microbiota (no detection of inflammation or pathogens) has significant effects on intestinal morphology (Liu 2015). The sow's microbiota is shared by the piglet during the first days of postnatal life (Bauer et al., 2006), therefore, maternal environment factors (diet composition, antibiotic treatment) that induce changes in maternal microbiota have huge effects on piglet gut physiology (Farmer 2015). Specific discussion of microbial gut succession in the young pig is detailed in a subsequent section.

1.7.1 Suckling Piglet Gut Immune Response

In developing management and nutritional strategies to maximize growth performance and health of pigs, it is critical to consider the effect of inflammation on gastrointestinal function (Liu 2015). Activation of the GI immune system leads to the production of a diverse set of specialized cells and signaling molecules, especially pro-inflammatory cytokines (Liu 2015). Over-production of these cytokines results in intestinal mucosal injury and dysfunction, and consequently results in poor growth (Liu 2015). When pigs suffer from intestinal infections (such as enterotoxigenic *Escherichia*

coli infection) they typically have lower feed intake and slower gain (Liu 2015). It is important that the GI is activated to handle invading pathogens; nutritional strategies that avoid excessive activation are important means to improve the efficiency of production. Inflammation is a fundamental aspect when considering the function of the GI tract. A healthy GI tract is known to be in a constant state of “controlled” inflammation because of the large population of bacteria found in the lumen, dietary antigens, and toxins (Liu 2015). When different intestinal infections (such as enterotoxigenic *E. coli* and *Salmonella typhimurium*) occur, inflammatory responses are increased, and intestinal morphology functions are damaged (Boyer et al., 2015; Liu 2015; Scharek-Tedin et al., 2013; Xiao et al., 2014). There is literature to support that inflammation induced by various factors causes drastic morphologic changes to the intestine (Liu 2015). In addition to digestive and absorptive function, inflammation can have a detrimental effect on the intestinal barrier function (Liu 2015). The intestine is the first barrier for nutrients and luminal components and has a central role in determining postnatal defense (Farmer 2015). The mucosal immune system is practically absent in the neonatal piglet even when the systemic immune tissue is well developed (Inman et al., 2005). Intestinal barrier function is commonly described as the capacity of the GI epithelium to prevent penetration by luminal bacteria and dietary allergens into the mucosa (Liu 2015). In the lumen, gastric acid and pancreatic fluid degrade bacteria and antigens (Liu 2015). The microclimate close to the epithelium is composed of the unstirred water layer, glycocalyx and mucus layer, which prevents bacterial adhesion and contains antimicrobial products secreted by Paneth cells (Liu 2015). Below this layer are epithelial cells separated by junctions that represent homo- and heterotypic binding of extracellular tight junction

proteins (Liu 2015). The intestinal epithelium is constructed of a monolayer of epithelial cells including columnar epithelial cells, Paneth cells, goblet cells and M cells (Liu 2015). These cells cover the mucosa and play a central role in intestinal mucosal barrier and host immune response (Liu 2015). The Paneth cells synthesize and secrete antimicrobial peptides such as lysozyme and defensins, these peptides have antimicrobial activity against various potential pathogens (Liu 2015). The goblet cells secrete mucus which acts as an antimicrobial and forms a charged gel that acts as a physical barrier (Liu 2015). Both cells together play an important role in limiting bacterial access to the epithelial surface. The intestine changes its surface by growing in length, circumference and in villous size (Farmer et al., 2015). Cells produced in the crypt regions differentiate and mature as they migrate along the crypt to the villous axis (Farmer 2015).

1.7.2 Development of the Suckling Piglet Gut

Colostrum stimulates intensive growth of the small intestine within the first two days of life (Farmer 2015). This rapid growth is caused by endocytosis of immunoglobulins, mucosal hyperplasia, and protein synthesis (Simmen et al., 1990; Burrin et al., 1992; Zhang et al., 1997).

The digestion and absorption of nutrients are of particular importance in a neonate whose nutrient requirements are enormous to support the rapid growth and high metabolic rates (Farmer 2015). Colostrum intake increases enzymatic maturation of the gut (Jensen et al., 2001). The ingestion of colostrum triggers specific effects on the processing of the brush border enzyme known as lactase-phlorizin hydrolase (Farmer 2015). Lactase-phlorizin hydrolase is the leading disaccharidase in the jejunal brush border membrane for the piglet and it is responsible for hydrolysis of lactose to galactose

and glucose (Corring et al., 1982; Henning 1987; Zhang et al., 1997). Piglets absorb glucose and galactose after lactase digestion and lactase activity will peak at birth (when colostrum is consumed) and decrease thereafter (Farmer 2015). As the piglet becomes older, the enzymes maltase and sucrase will increase as well (Farmer 2015).

For a suckling piglet, the maternal environment impacts the development of their GIT (Farmer 2015). There are several factors such as sow diet composition or antibiotic treatment that are primarily associated with the maternal environment that have an impact on the development of the piglet during the suckling phase (Farmer 2015). If antibiotics are administered to the sow during the last week of gestation, it can affect intestinal development in offspring for a period of at least 7 weeks (De Greeff et al., 2020). When antibiotics are administered to the sow, it effects the microbial colonization and development of the gut in her offspring (De Greeff et al., 2020). The treated sow's bacterial populations were changed in the vaginal microbiota at parturition and the population of bacteria in the sow feces at day 1 of lactation (De Greeff et al., 2020). Literature suggests that it's likely that the piglet's microbiota is colonized from maternal microbiota either via the vagina or from the ingestion of maternal feces or colostrum (Blaser and Dominguez-Bello, 2016; De Greeff et al., 2020). Consequently, antibiotics administered to the sow could impact early colonization of the neonate by influencing the first colonies in the GIT of the piglet (Schulfer and Blaser 2015; De Greeff et al., 2020). Piglets from sows that were treated with antibiotics showed increased expression of genes involved with the processes of "tight junctions" and "immunoglobulins" after parturition (De Greeff et al., 2020). Upregulation of those genes can lead to intestinal epithelium that closes rapidly in piglets from treated sows (De Greeff et al., 2020). Feed additives have

been investigated, and used, to improve pig health and productivity via intestinal microbiota manipulation (Nowland et al., 2021). These dietary additives include phytochemicals (Walker et al., 2019), essential oils (Ruzauskas et al., 2020), organic acids (Oh et al., 2019), probiotics (Shu et al., 2001), minerals (Højberg et al., 2005), and medium chain fatty acids (Lan and Kim 2018). The maternal gut microbiota can influence the suckling piglet's maturation and function of the mucosal immune system, such as the bactericidal Paneth cell activity, IgA production, and intraepithelial lymphocyte development to prevent inflammatory responses that can disrupt the barrier function (Chowdhury et al., 2007; Hooper, 2004; Farmer, 2015).

1.7.3 Microbial Succession of the Piglet Gut

The microbial composition and ecological succession of the intestinal microbiota in early life of the piglet is shaped by several factors (Guevarra 2019). However, the piglet will rapidly undergo shifts to an extremely dense microbial population that eventually experiences a microbial succession and establishes an adult-like microbial community or microbiome (Guevarra 2019). A microbiome is defined as a collection of microorganisms (bacteria, archaea, fungi, and viruses) (Nowland et al., 2021). Colostrum and milk contain a variety of bacteria and prebiotics that assist in intestinal development (Bian et al., 2016; Chen et al., 2018a; Nowland et al., 2021). There are multiple bacteria positively correlated with young pig growth performance. For example, species of the *Bacillus* genus improve daily gain and reduce feed conversion ratio (Wang et al., 2018); *Bacteroidetes* increased abundance in diarrhea resistance in suckling piglets and lighter pigs in the post-wean period (Dou et al., 2017; McCormack et al., 2017). Species of the *Ruminococcaceae* genus improve growth of suckling piglets and is higher in piglets not

displaying post-wean diarrhea (Mach et al., 2015; Dou et al., 2017; Gaukroger et al., 2020) and *Lactobacillus* species are associated with increased feed efficiency, gain, and anti-inflammatory activity (Gaukroger et al., 2020; McCormack et al., 2017; Hillman 2001). There are multiple other species that can be found in the piglet gut microbiome that provide added benefit to the piglet (Nowland et al., 2020). Microbial succession has the potential to impact animal health and productivity (Tannock, 2005). The piglet's gut ecosystem is made up of numerous bacterial colonists in competition with each other (Chen et al., 2018b). In the suckling period of a piglet's life, the taxonomic groups found are: Firmicutes, Bacteroidetes, and Proteobacteria (Chen et al., 2018b). Weaning is an extremely stressful event for the pig, and it can lead to disruption of the microbiome and decreased health status as well as growth performance (Guevarra et al., 2018). In the post-wean or nursery period, the diet shifts to a dry form, altering the gut microbiome. *Lactobacillaceae* species appeared in the suckling piglet around day 3 of age and remained throughout the suckling period (Petri et al., 2010). Guevarra et al., (2018) investigated the microbial succession in the pig after weaning and interestingly discovered that after weaning there was an increase in the genus *Prevotella*. *Prevotella* is linked to the fermentation of plant-derived non-starch polysaccharides to short-chain fatty acids (Ivarsson et al., 2014) Guevarra et al., (2018) also discovered that there were genes associated with bacterial heat shock responses which is a response to stress and are needed for successfully adapting to changes in the environment and bacterial habitat (Yura et al., 1993). Guevarra et al., (2018) also noted that there was higher abundance of gene groups that were linked with oxidative stress. The weaning process significantly changes the composition and functionality of the gut microbiome (Guevarra et al., 2018).

1.8 Passive Immunity

Piglets are considered immunodeficient at birth and are dependent upon a supply of specific and non-specific immune factors present in maternal colostrum and milk for immune protection, development, and survival (Salmon et al., 2009). This transfer of immune protection from mother to offspring is known as passive immunity. Once piglets are born, they will be exposed to various pathogens within their environment and they will begin to protect themselves against these pathogens using immune components produced themselves, this is their active immunity. During gestation, the female's placenta blocks the transfer of immunoglobulins to the neonate (Rooke and Bland 2002) thus passive transfer via colostrum is critical. Between 24 to 36 h after birth, the gut will begin to close, making it critical for absorption of intact immunoglobulins to occur before closure (Rooke and Bland 2002). Within 24 h after birth, serum IgG concentrations in suckling piglets are often like that of their dam because of the passive transfer of intact IgG from colostrum (Farmer 2015). Colostrum contains immune cells and immunomodulatory factors that play a role in the response to pathogens and that help maturation of the piglet's immune system (Salmon et al., 2009; Farmer 2015). The examples used of immunomodulatory factors including ferritin which led to an increase in IgM and the inclusion of Freund's adjuvant (immune booster) led to an increase in IgA (Salmon et al., 2009). Immunoglobulins such as IgG, IgA, and IgM have different routes of absorption (Rooke and Bland 2002). Immunoglobulin G is the most important immunoglobulin during the first few weeks of life (Kielland et al., 2015). IgG is absorbed via the gastrointestinal tract 24 to 48 h after birth (Sjaastad et al., 2012). Colostral IgG concentration varies between individual sows (Kielland et al., 2015), and ranges from

48.0 to 95.6 g/L (Klobasa and Butler 1987; Tuchscherer et al., 2002; Svendsen et al., 2005; Couret et al., 2009; Foisnet et al., 2010; Bovey et al., 2014). Immunoglobulin A is needed to locally produce specific antibodies at the mucosal level. IgM is produced in smaller amounts compared with IgG and is a first line defense response to antigens (Mehra et al., 2006). Colostrum production plays a crucial role for passive immunity.

1.9 Colostrum and Milk Production

As mentioned previously, colostrum and milk production play an essential role in ensuring piglet survival and growth (Farmer 2015). Piglet mortality is typically highest during the first 3 days after parturition (Tuchscherer et al., 2000) and literature suggests that early pre-wean mortality is primarily due to low consumption of colostrum (Edwards, 2002; Le Dividich et al., 2005). Colostrum yield is a limiting factor for piglet survival during the first few days following parturition (Farmer 2015). Colostrum yield is highly variable among sows and ranges vary from 1 to more than 6.0 kg (Devillers et al., 2007; Quesnel, 2011). An increase in litter size results in a lesser mean birth weight, which indicates that there is an increased proportion of light weight piglets (Quiniou et al., 2002). With the lightweight piglets at birth, there is a concern for piglet viability and survival, piglets that weigh less than one kg at birth have a very low chance of making it to weaning (Quiniou et al., 2002). Piglets that weigh less than one kg, may have difficulty suckling due to physical size difference between their mouth and the sow teat or they do not have the energy to suckle resulting in an inadequate colostrum intake increasing pre-wean mortality. There is little literature that provides knowledge on colostrum yield and its influencing factors. Because of the difficulty in measuring colostrum intake and colostrum production, an equation has been developed to estimate colostrum intake based

on growth from the birth of the first piglet to 24 h after and the assumption that total colostrum intake of the litter is a reasonable estimate of colostrum production (Devillers et al., 2004b). Litter growth from birth to day 3, 5, or 7 of lactation is directly related with consumption of colostrum during the 24 h after birth (Farmer 2015).

During the past few decades, improving sow prolificacy and carcass merit of market hogs have been the main objectives for genetic selection programs, which has led to an increase of 2 to 4 piglets per farrowing over past 20 years (Farmer 2015). However, the hyperprolific sow lines that are being utilized currently, suggest that both colostrum as well as milk yield are even more limiting (Farmer 2015). The increased litter sizes have resulted in greater demand for colostrum and milk, as well as immunoglobulins, to meet the needs of the additional piglets. The amount of colostrum that is ingested is highly variable between piglets and is dependent upon the sow's capabilities to supply enough colostrum and on the piglet's ability to suckle (Devillers et al., 2011).

Colostrum and milk differ in timing of secretion and composition (Farmer 2015). Influencing colostrum composition to increase piglet gain via the maternal diet, is a factor to consider when piglet growth is part of a determination for a sow to be culled. Colostrum is the first secretion of the mammary glands which is largely synthesized before the onset of parturition (Farmer 2015). Colostrum is characterized by high concentrations of IgG compared to that of milk and contains lower concentrations of lactose and lipids than milk (Farmer 2015). Colostrum is defined as the mammary secretions ingested by piglets until 24 h after the onset of parturition, transient milk is produced after colostrum until around day 4 of lactation, and mature milk from day 10 of lactation (Devillers et al., 2004a). The modern hyperprolific sow can sometimes endure a

longer farrowing duration which can ultimately decrease colostrum yield (Hasan et al., 2019). The decrease in colostrum yield, can bring down sow productivity which can result in a culling, therefore hurting the lifetime potential of that female. The content and composition of colostrum fat is dependent upon the diet fed throughout gestation which means manipulation of the diet may be used to impact the colostrum fatty acid content and/or composition (Schmidt and Herpin 1998; Dividich et al., 2005). Colostrum is defined as the synthesis of milk-specific constituents and the transfer of IgG into lacteal secretions (Quesnel and Farmer 2019). Colostrum quantity and quality can be influenced by sow characteristics such as endocrine status, nutrition, immune stress, and litter characteristics (Quesnel and Farmer 2019) as well as parity. Parity 2 and 3 sows tended to produce more colostrum (4.3 kg) than primiparous (3.4 kg) and older sows (3.6 kg) (Devillers et al., 2007).

At birth, piglets are exposed to a cold environment, thus activation of thermoregulatory mechanisms is key for maintenance of homeostasis (Farmer 2015). Unlike other species, newborn piglets lack thermogenic brown adipose tissue, and overall lipid content of the newborn piglet is relatively low (less than 2%; Seerley and Poole, 1974). Therefore, colostrum intake is needed to replenish the hepatic and muscle glycogen stores used for thermogenesis for piglet survival and growth (Theil et al., 2011). Hepatic and muscle glycogen make up the main body stores for heat-producing nutrients for oxidation and these energy stores are depleted within 12 to 17 h after birth in the absence of colostrum intake (Theil et al., 2011). Colostrum also helps adapt the piglet to the new environment by supplying digestive enzymes and by stimulating energy metabolism and thermoregulatory mechanisms (Herpin et al., 2005)

Sow milk production is stimulated by stimuli that originates from suckling piglets which indicates that it is dependent upon litter size and weight (Farmer 2015). The removal of milk from the mammary glands is the most important aspect for the maintenance of milk secretion (Auldist et al., 2000). Total milk production is related to a number of factors including, the number of functional teats, the suckling intensity, and the resultant milk removal from individual glands (Auldist et al., 2000). For example, total milk production increases linearly by approximately 0.6 kg of milk per day per additional suckling piglet (Noblet et al., 1998). Milk intake on a per piglet basis decreases as litter sizes increases (Kim and Hurley 2001). A sow's litter size needs to be equivalent to her functional teat capacity to ensure she is reaching her maximum milk production through that lactation. If teat capacity isn't filled, there could be detrimental loss to those non-suckled glands, hurting subsequent milk output.

Nursing and suckling behavior is a complex sequence of events (Thodberg and Sørensen 2006), with a pre-massage phase, milk let-down phase, post-ejection massage phase, and nursing (Whittemore and Fraser, 1974; Fraser, 1980). The udder massage stimulates the subsequent milk production in the massaged gland (Algers and Jensen 1985). Within a suckling event, milk is not available to the piglets continuously and is only let down during ejection periods which only last for about 10 to 15 seconds (Fraser, 1980).

Sow milk production over the entire lactation period can be described by four phases (colostral, ascending, plateau, and descending phases) (Farmer 2015). During the ascending phase of lactation typically day 2 to day 10, nursing frequency doubles going from 17 to 35 nursings per day (Jensen et al., 1991). Campbell and Dunkin (1982)

discovered that the quantity of milk obtained at each nursing period also increases from 29 g to 53 g between the first and third week of lactation. On day 4 of lactation total milk production ranges anywhere from 5 kg to 10 kg per day (average 8 kg/day) (Toner et al., 1996). In modern swine production, most sows do not reach the descending phase of lactation because they are weaned during the plateau phase (Farmer 2015). When piglets are removed at weaning, milk will accumulate in the alveoli, triggering milk stasis (Kim and Hurley 2001). At weaning, mammary involution takes place where the parenchymal tissue is undergoing extreme regression (Farmer 2015). The dramatic loss of tissue DNA in nonsuckled glands shows that there is cell loss occurring (Kim and Hurley 2001). When litter size doesn't match sow functional teat capacity, this will result in a lesser milk output, resulting in a reason to be potentially culled and hurting the individual sow's longevity and productivity.

1.10 Mammary Development

Primary mammary growth and development occurs during lactation (Kim and Hurley, 2001). The mammary glands on sows are located in two parallel rows along the ventral body wall from the thoracic region to the inguinal area (Farmer 2015). The thoracic, abdominal, and inguinal glands are attached to the ventral body wall by adipose and connective tissue and each gland is separate and distinctive from other adjoining glands (Turner, 1952). Mammary development occurs at three distinctive periods, and this is when management, nutritional and hormonal strategies can be utilized to try to stimulate mammogenesis (Farmer 2015). Strategies to improve colostrum quality and increase Ig content have been widely investigated because both are sensitive to nutritional changes (Quesnel and Farmer, 2019). For example, the fatty acid content of colostrum depends

upon the amount of lipids that are provided to the gestating sow in late gestation (Farmer and Quesnel, 2009). In pregnant gilts and sows, quantitative development of the mammary glands is slow in the first two-thirds of gestation, while almost all accumulation of mammary tissues and DNA occurs in the last third (Hacker and Hill, 1972; Kensinger et al., 1982; Sorensen et al., 2002). Ji et al. (2006) reported that there is a significant increase in weight of mammary gland tissue from day 45 of gestation until around day 112. Mammary development and suckling density play a crucial role in sow longevity, improper management techniques in lactation can lead to a decrease in milk production in the subsequent lactation periods. Lower milk production could hurt litter growth performance, which is a culling indicator in most commercial farms. Proper management of litter size based upon the individual and her functional teat count as well as her suckling density is a necessary to ensure maximum mammary development for multiple lactations.

1.11 Feeding Medium Chain Fatty Acids to Sows

With the impressive improvement of sow reproductive performance (litter size, piglets born alive), the need to extend sow longevity is crucial and needs to be explored. As noted above, dietary manipulation via nutrient concentration or feed additives is a tool to aid improvement of sow longevity. One such nutritional strategy that can potentially influence both the sow and her offspring is the inclusion of medium chain fatty acids. Medium chain fatty acids (MCFA) and monoglycerides have emerged as potential feed additives due to the key molecular features and versatile functions, including inhibitory activity against viral and bacterial pathogens (Jackman et al., 2020). Fatty acids with an aliphatic tail of six to twelve carbon atoms are classified as MCFA, which occur naturally

as medium-chain triglycerides (MCT) in milk fat and various feed stuffs (such as coconut oil or palm oil). Both MCFA and MCT have specific nutritional and metabolic characteristics, including rapid digestion, passive absorption, and obligatory oxidation. Medium chain fatty acids are building blocks of MCTs, they are created after lipase breakdown of triglycerides *in vivo* to yield MCFAs and monoglycerides (Jackman et al., 2020). Medium chain fatty acids can be utilized directly by the enterocytes for energy production and thereby help to support the integrity of the intestine for young piglets (Guillot 1993). Medium chain fatty acids are known to be absorbed according to a pathway which in some respect differs from that of long-chain fatty acids (Guillot 1993). Medium chain triglycerides are assumed to undergo a rapid hydrolysis by lipases because of their solubility and motility at the lipid drop interface compared with long-chain triglycerides (Greenberger 1966). Medium chain fatty acids are then transported as non-esterified fatty acids into the portal blood stream and reach the liver directly, providing an easy supply of energy (Hashim 1964). Medium chain fatty acids and MCT have been suggested to improve gut health under inflammatory conditions, however the evidence in pigs is limited (Liu 2015). Medium chain fatty acids and MCT have antimicrobial and antiviral activity in the gastric lining and small intestine of pigs (Zentek et al., 2012). Zentek et al. (2012) also reported that low dietary MCFA supplementation affected gastric microbial ecology, decreased propionic, butyric and valeric acid concentrations, and increased acetic acid concentration in the small intestine of weanling piglets. In addition, Messens et al. (2010) reported that MCFA inhibited *Salmonella typhimurium* in an *in vitro* simulation of the cecum. An important benefit of directly using free MCFA is that they exhibit antimicrobial properties and thus can potentially inhibit viral and

bacterial pathogens in the feed to reduce risk of disease transmission (Jackman et al., 2020). Lan and Kim (2018) investigated supplementation of MCFA blends in sow diet 42 days before farrowing through to weaning (28 days after parturition). Supplementation showed no effect on sow body weight, backfat thickness, or feed intake (Lan and Kim, 2018). Piglets were weighed on d 14, 21, 28 and body weight was significantly higher in piglets from supplemented sows compared to control (Lan and Kim, 2018). Sow fecal microbiota was impacted by MCFA supplementation where an increase in *Lactobacillus* and a decrease in *E.coli* was detected (Lan and Kim, 2018). A similar response in piglet fecal microbiota at the same time points as their mothers (farrowing and weaning) was reported (Lan and Kim, 2018). This study shows that providing MCFA in the maternal diet in gestation and lactation improved suckling piglet body weight and gut microbial composition. Altering the count of *Lactobacillus* suggests the gut of the piglet from the supplemented sow had greater concentration of beneficial bacteria. Świątkiewicz et al., (2020) investigated supplementing two oils (rapeseed or coconut (form of medium chain triglyceride) during late gestation and lactation and the effects on offspring performance. This study also investigated piglet growth after weaning, supplementing piglets from day 7 of age until day 84 of age with either 0.3% MCT or 0.3% caprylic acid (Świątkiewicz, et al., 2020). The results of this study showed that type of oil fed to sows did not affect the reproductive characteristics, including birth weight of piglets, however, piglet mortality was improved by supplementation of coconut oil. The coconut oil supplementation influenced fatty acid profile in the milk, as well as beneficial effects on IgM and IgG milk levels (Świątkiewicz et al., 2020). With respect to offspring performance, growth was improved by either MCT or caprylic acid supplementation

(Świątkiewicz et al., 2020). Gatlin et al. (2002) investigated the effects of supplemental dietary fat fed as either MCT or long chain triglycerides on reproduction and lactation performance and body condition. Sows were assigned to one of 3 dietary treatments consisting of: 1) no supplementation 2) 10% MCT inclusion 3) 10% inclusion of long chain triglycerides (Gatlin et al., 2002). Born alive was not affected by dietary treatment, however, sows that were supplemented had more mummies and stillborns (Gatlin et al., 2002). Lactation feed intake of sows that were supplemented was 10% lower compared to control sows (Gatlin et al., 2002). Sow body condition was improved by supplementation and their offspring had greater gain and heavier weaning weights (Gatlin et al., 2002). Overall, supplementation of MCFA in free form or in triglyceride form showed a positive improvement on either the sow or her offspring.

1.12 Medium Chain Fatty Acids: mode of action

Medium chain fatty acids are antimicrobial agents that can disrupt the phospholipid bilayer surrounding membrane-enclosed pathogens such as bacteria and lipid bilayer-enveloped viruses (Jackman et al., 2020). Medium chain fatty acids can inhibit bacterial growth or induce bacterial cell lysis and cell killing (Yoon et al., 2018). Jackman et al. (2020) reported that MCFAs have more potent inhibitory activity against Gram-positive bacteria than Gram-negative bacteria due to Gram-positive having simpler, singular lipid bilayer cell membrane structures, while Gram-negative have more complex inner and outer cell membranes. Medium chain fatty acids can also disrupt a variety of lipid bilayer-enveloped virus particles compromising infectivity (Jackman et al., 2020). On the other hand, MCFAs are completely inactive against non-enveloped viruses (Jackman et al., 2020). In sow diets, MCFA may act through multiple mechanisms to positively

influence sow and piglet health. It can help aid in killing pathogens in the feed before it the sow digests the feed. It also can help defend against pathogens inside the gut. An example of this was described above where the gut microbiota had shifted to a more beneficial bacterial profile. This mode of action acts similarly for the suckling piglet and in the nursery periods. In the swine industry, MCFA are also emerging as potential growth performance enhancers (Jackman et al., 2020).

Besides previously stated benefits about including MCFA to enhance piglet health and performance, MCFA also can serve as a source of dietary fatty acids in supplemental oils. Oils such as coconut oil are naturally occurring source of MCT and can be used as a source of dietary energy and MCT, but the inclusion cost is a factor that limits the usage of these oils in the maternal diet.

Medium chain fatty acids aid in establishing a healthy gut microbiome with bacteria that aid in piglet health and growth in the suckling period. The inclusion of MCFA has the potential to improve colostrum and quality, allowing the piglet a better chance at survival. These benefits from supplemental MCFA in the maternal diet has the potential to make an impact on sow longevity and piglet health.

CHAPTER 2

2.0 Supplementing Medium Chain Fatty Acids throughout gestation and lactation to improve sow and piglet performance

2.1 Abstract

A study was conducted to investigate growth and reproductive performance traits following supplementation of MCFA from d 28 of gestation throughout lactation and in weaned pig diets. A total of 77 sows and gilts (218.15 kg \pm 32.15 kg BW at d28 of gestation) were assigned to one of 2 dietary treatments: Control (CON) and control + MCFA (SUP). Response variables measured were sow body weight (BW), sow daily feed intake, litter characteristics at birth and weaning, piglet BW, and piglet weight distribution at birth and wean. At weaning, offspring were allotted to pens balanced by weight and litter within maternal dietary treatment. Pens of pigs received the same dietary treatment as the sow during the suckling phase in a 3-phase feeding regimen (phase 1: d0-d6, phase 2: d7-20, phase 3: d21-42) for 42 d. Variables measured were pig BW, daily gain, feed intake, gain:feed. Performance response variables were analyzed as randomized complete block using the Mixed model procedure of SAS v9.4 with sow and pen as experimental unit. A total of 32 sows and her offspring were utilized for biological collections and analysis. From these litters, a total of 5 piglets (1 heavy, 1 light, 3 average) were selected based upon litter average weight \pm 1 standard deviation. Piglet BW distributions among categories at birth and weaning and the change in category from birth to weaning were calculated

. At birth, 'light' represented piglets <1.07 kg, 'middle' represented piglets 1.07 – 1.68 kg, and 'heavy' represented piglets >1.68 kg. At weaning, the 'light', 'middle' and 'heavy' categories were as follows: <4.43, 4.43 – 7.05, and >7.05 kg, respectively.

Distribution of pigs in each weight category and percent changed was analyzed using the proc Freq method of SAS, within main effects of supplementation. No effect of maternal diet was observed for sow BW at d 110 (228.63 ± 3.12 kg) or weaning (211.92 ± 3.15 kg), piglet birth weight (1.38 ± 0.05 kg), piglet wean weight (5.74 ± 0.11 kg), or litter size (14.40 ± 0.44). Medium chain fatty acid sows had greater daily feed intake ($P < 0.05$) compared to the sows fed the control diet. In the nursery phase there was no maternal diet supplementation effect for all measured response variables and no effect of nursery dietary treatment at birth, UNSUP litters had a larger percentage of average piglets (69.75% UNSUP, 59.02% SUP), a lesser percentage of light piglets (15.41% UNSUP, 19.67%) and a lesser percentage of heavy weight piglets (14.84% UNSUP, 21.31% SUP; $\chi^2 < 0.01$). At weaning, UNSUP sows tended to have a larger percentage of average piglets (71.68% UNSUP, 66.60% SUP), a lesser percentage of light piglets (13.49% UNSUP, 18.24% SUP) and they had lesser heavy weight birth piglets (14.84% UNSUP, 15.16% SUP; $\chi^2 = 0.10$). Inclusion of MCFA in gestation and lactation increased sow lactation daily feed intake. In lactation, feed intake is a key limiting factor for milk output, thus MCFA may contribute to improved sow milk output.

Key Words: lactation, medium chain fatty acid, nursery, piglet, sow

2.2 Introduction

Pathogen status of a sow herd plays a critical role in sow longevity as well as reproductive performance (Farmer 2015). Common pathogens that can disrupt a sow herd are porcine reproductive and respiratory syndrome, pseudorabies, porcine enterovirus, porcine epidemic diarrhea virus, swine influenza, chronic erysipelas, *Mycoplasma hyopneumonia*, and *Streptococcus suis* (Loula 2000; Sanz et al., 2002; Stalder et al., 2004). Some potential feed alternatives under investigation to help sows combat incidences of increased pathogen pressure, are essential oils, organic acids, antimicrobial peptides as well as MCFA. As noted in Chapter 1.0, MCFA can act as antimicrobial agents and inhibit bacterial growth or induce bacterial cell lysis and cell killing (Yoon et al., 2018; Jackman et al., 2020). Feed additives have various potential uses for swine, including, improving growth performance, contributing to improving feed utilization as well as improving pathogen status in the gut (Zentek et al., 2012; Liu 2015; Lan and Kim 2018; Jackman et al., 2020). Previous literature suggests that MCFA supplementation of sow diets in late gestation and lactation shortened wean-to-estrus interval (Chen et al., 2019). Supplementation of MCT had beneficial impacts on piglet serum IgG and IgM levels and lowered piglet mortality (Swiatkiewicz et al., 2020). Nursery weight gain was higher in pigs from MCFA-supplemented sows (Swiatkiewicz et al., 2020). In the nursery Swiatkiewicz et al. (2020) examined 4 experimental diets (control, supplementation at 0.3% mixture of MCT, 0.3% caprylic acid, or 0.51% lauric acid). This study looked at growth performance of pigs up to 12 weeks after birth. Lan and Kim (2018) supplemented MCFA in gestation and throughout lactation and looked at the impact it made on reproductive characteristics as well as growth and biological parameters, such as

the gut microbiome. These studies suggest that feeding the MCFAs into the maternal diet provides added benefits to not only herself, but her offspring. There are currently several products that have been developed by commercial companies with various combinations of MCFAs. There is variability among the content of MCFA and what inclusion level is needed to impact growth performance as well as other biological characteristics. The previous studies mentioned above investigated the supplementation of MCFA in gestation and lactation but did not further supplement in the nursery period or follow throughout the entire nursery period to see if there were any carry-over benefits from maternal supplementation. The objective of this study was conducted to look at the connection between supplementation in the maternal diet from conception through weaning on growth of offspring up to the grower phase.

2.3 Materials and Methods

The South Dakota State University Institutional Animal Care and Use Committee approved the protocol used in this experiment (IACUC # 17-072A, 18-046A). The study consisted of four sow breed groups, groups 1 and 2 (study 1) were conducted from January to August 2020 and groups 3 and 4 (study 2) were conducted from November 2020 to May 2021. In the results and discussion, responses of each group of sows and their offspring will be referred to as either from study 1 or 2.

2.3.1 *Animal management, diets and feeding*

On day 28 of gestation, a total of 77 mixed parity sows (PIC 1050 Landrace x Yorkshire) were used at South Dakota State University's Swine Research and Education Center. Sows were blocked by BW within parity category (gilts, parity 1, parity 2+) and allotted to 1 of 2 dietary treatments consisting of a control diet (UNSUP) or 0.3%

inclusion of MCFA blend (SUP; Table 2.1 and 2.2). In this study, the blend contains lauric acid, caprylic acid, formic acid, propionic acid, acetic acid, benzoic acid, sorbic acid, and citric acid. These dietary treatments were provided throughout gestation and lactation and formulated to meet or exceed nutrient requirements in gestation and lactation according to NSNG (2010). Diets were manufactured at the South Dakota State University feed mill (Brookings, SD). Feed intake was recorded for individual sows during gestation and lactation. Sows were housed in gestation crates from time of breeding up until pregnancy was confirmed (day 28 ± 3). Sows were housed in pens (5.49 to 6.10 m per sow) from confirmation of pregnancy until d110 of gestation and feed provided via an electronic feeding system (Mannebeck; PigTekPig Equipment Group, Milford, Indiana, USA) unless they were removed and placed into an individual gestation stall (0.61 m x 1.98 m). If sows were placed into a gestation stall, they were hand-fed their respective diet for the remainder of gestation. The farrowing house was equipped with individual farrowing crates (1.83 m x 2.43 m) containing an electronic feeding hopper (Gestal 3G; Jyga Technologies, Greeley, KS, USA) that allowed for daily intake up to 20% above the set lactation curve for ad libitum intake. Feed was provided in 6 meals/day at 3-hour intervals that began at 0500 daily. If a female was deemed either a poor eater (i.e., feeder was full of feed at first AM check at least 2 consecutive days or feed dispensed was <50% of targeted intake according to parity specific lactation curve) or was eating well above the curve (feed dispensed was 20% above targeted intake according to parity specific lactation curve) feed amount dispensed was decreased or increased, respectively. The farrowing crates were equipped with nipple waterers for sows and piglets, and heat mats and heat lamps for piglets. Sows and gilts were

supervised during farrowing by a trained technician and the assigned graduate research assistant 24h/d from birth of first piglet to the last piglet born within the sow group. Sows and piglets were checked twice daily by the graduate research assistant following the completion of farrowing up until weaning. A 1 ml intravulval injection of Dinoprost tromethamine (Lutalyse, Zoetis, Pasippany, NJ) was administered at d 116 of gestation to females that had yet to farrow.

Cross fostering occurred within maternal treatment groups only and litters were equalized to 12 to 15 piglets depending on functional teat count within 48 hours by either cross fostering or removal. Removals included piglets that were deemed to be runts (≤ 600 g at birth) and fallbacks that were taken off test and placed on milk replacer (Birthright baby pig milk replacer, Ralco, Marshall, MN) using milk decks (Birthright milk deck, Ralco, Marshall, MN). Fallbacks were defined as piglets who appeared small or thin and had an ADG of ≤ 30 g from birth to time of weighing. Animals whose gain ranged from 35 to 70 g were reweighed within three days to determine if placement into milk deck was needed. Within 24 h after birth of the first piglet, all piglets received a 2 mL intramuscular (i.m.) injection of iron dextran (Uniferon 200, Pharamacosmos, Watchung, NJ), 1 mL oral dosage of Ponazuril (Marquis, Merial, Duluth, Georgia), and if birth weight was under 1 kg, piglets received a 0.25 mL i.m. injection of Excede (Zoetis, Pasippany, NJ) and a 1 mL oral dosage of First Pulse D (Ralco, Marshall, MN). Piglets weighing ≤ 0.50 kg (1.1lbs) were euthanized using blunt force. On d 2 of age piglets were assigned to a birth weight category based on individual litter average weight. Categories consisted of light, middle, heavy and determination of categories within a litter was accomplished by determining the average litter weight ± 1 standard deviation. Weight

distribution at birth were as follows: low (<1.02 kg), middle (1.02-1.65 kg), and heavy (>1.65 kg). Weaning weight distributions were determined similarly to birth weight and were as follows: low (<4.43 kg), middle (4.43-7.05 kg), and heavy (> 7.05 kg). “Change to wean” was defined as the weight category change from birth to weaning to determine the percentage of animals that maintained, improved, or declined a category. On d3 of age, heat lamps were turned off, piglets were processed (tail docking, tattooing, and castration) and vaccinated with 1 mL i.m. injection of Circumvent PCV-M G2 (Merck Animal Health, Madison, NJ). Young boars who appeared small or thin were processed at 5 to 6 days of age as a measure to prevent further health decline. Individual piglets or litters identified with scours after d 3 were treated with 1 mL oral dose of Spectinomycin (Spectogard Scour-chek, Bimdea, Oakbrook Terrace, IL) twice daily for two days. Two weeks following the completion of farrowing, all piglets were vaccinated with 1 mL oral drench of *Escherichia coli* vaccine (Entero-vac, ARKO Laboratories, Jewell, IA). At weaning, all piglets received a booster dose (1 mL i.m. injection) of Circumvent PCV-M-G2 (Merck Animal Health, Madison, NJ).

2.3.2 Weaning procedure and dietary treatments

At weaning, 438 piglets (5.66 ± 1.37 kg BW) were allocated in a 2x2 factorial based on maternal diet (UNSUP or SUP) and post-weaning diet (UNSUP_{nurs} or SUP_{nurs}) in a 3-phase nursery pig feeding program (Table 2.1 and 2.2) lasting 42 days. Piglets were weaned into groups ranging from 10-16 pigs/pen within maternal treatment resulting in 18-20 pens/maternal treatment. Pens were balanced by weight and litter as best as possible. Piglets that were reared in milk decks were not weighed at weaning, weaned into separate pens, and deemed off-test. Feed and water were offered ad libitum.

Individual veterinary treatment of pigs was administered on a per pig basis and medication type, dosage amount, and reason for treatment was recorded. Pigs who were removed from the trial due to poor health, death, or euthanasia were recorded with date and weight at removal. All pigs and facilities were checked daily by the assigned graduate research assistant during the course of the study.

2.3.3 Data collections, chemical analyses, and calculations

Sow BW was measured at d 28 of gestation, d 110 of gestation, within 24 hours after parturition, and at weaning; back fat (BF) was measured by sow body condition caliper score at last rib at d 28 of gestation, d 110 of gestation and at weaning.

Feed samples were collected throughout gestation and lactation as well as the post-wean period from the feeders at the farm. Samples were pooled by stage (early, mid, late gestation/lactation/nursery phase) and experimental diet. Feed samples were mixed and placed into whirl-pak bags (Nasco, Fort Atkinson, WI) and shipped for analysis. Diets were analyzed for CP, moisture, ash, ether extract and crude fiber (Table 2.1) (Experiment Station Chemical Laboratories, University of Missouri – Columbia) (Crude Protein LECO, Crude Fat, Moisture, Ash, Crude Fiber Kjeldahl).

When a piglet was born, time of birth was recorded, pigs were dried off using a desiccant (Arbocel R-#44; Pipestone Veterinary Services, Pipestone, MN) and paper towels. Umbilical cords were tied and shortened to approximately 10 cm in length. Additionally, pigs were given an individual ear tag for identification, weighed and sex recorded before placing them next to the sow. Stillborn and mummified fetuses were also weighed, sexed (where possible) and time of birth was recorded. During parturition, after 60 min had elapsed with no farrowing progress evident, trained technicians followed the

proper sleeving protocol and recorded it on the data sheet (# of piglets pulled, clean sleeve, after birth expulsion, etc.). The farrowing process was deemed complete when no new piglet had been born after 1 h, no evidence of piglet after sleeving and placenta expulsion had been observed. Within 24 h proceeding farrowing completion, sows were weighed to measure weight of conceptus and placenta, blood loss, and fecal loss.

In sow groups 1 and 2, at 24 h after birth of the first piglet in each litter, piglets were individually weighed to calculate colostrum intake using the equation:

$$CI = -106 + 2.26 WG + 200 BWB + 0.111 D - 1,414 WG/D + 0.0182 WG/BWB$$

Where, WG represents 24 h piglet weight gain in grams, D is duration of colostrum suckling in minutes, and BWB is body weight at birth in kg as described by Theil et al (2014).

Suckling piglets were also weighed at d7 of age and at weaning. After weaning, pigs were weighed and feed disappearance was measured in week 1 in study 2 only, 2, 4 and 6.

2.3.4 Statistical analysis

Data was analyzed using the MIXED model procedure of SAS (Version 9.4, SAS Inst. Inc., Cary, NC) and checked for assumptions using ANOVA, where dietary regimen within parity category (0, 1, 2+) was a fixed effect, sow was the experimental unit, and block was the random effect. Due to the range in parity structure within treatment groups, sows were assigned to one of three parity groups, parity 0 (gilts) parity 1, or parity 2+. Main effect of parity and parity by dietary treatment interactions were also evaluated. For post-weaning performance, wean room was included as a random effect. Results were

considered significant at P -value ≤ 0.05 and tendencies were detected at $0.05 \leq P \leq 0.10$. Weight categories and category change were analyzed using the Freq procedure in SAS (Version 9.4, SAS Inst. Inc., Cary, NC).

2.4 Results

2.4.1. *Farrowing and Suckling Growth Performance*

Sow BW in gestation, lactation and at weaning was not affected by maternal dietary treatment (Table 2.3). However, sow body weight was impacted ($P < 0.01$) by parity (Table 2.3) where parity 0 females weighed less at all time points compared to parity 1 and 2+ females and parity 1 females weighed less than parity 2+ sows at all time points. There were no interactions among parity category and sow dietary treatment. As illustrated in Table 2.3, sow lactation feed intake increased with the inclusion of MCFA in the diet ($P < 0.05$). Similarly, feed intake was greater in parity 2+ than parity 1 females which was greater than gilts ($P < 0.05$). Litter born alive, stillborn, total born, mummies, piglets weaned, kg weaned, or piglet pre-wean mortality was not affected by maternal treatment (Table 2.3). No effect of maternal dietary treatment was observed for birth weight, d 7 of age, or at weaning (Table 2.3). Daily gain for piglets in the suckling period was not affected by maternal treatment (Table 2.3). Colostrum intake and yield was not impacted by maternal treatment (Table 2.3). Parity did play a factor in piglet BW on d7 of age ($P < 0.01$); piglets from parity 1 sows were heavier than piglets from gilt and parity 2+ litters which is similarly reflected in ADG. The interaction between sow reproductive performance and study is reported in Table 2.4. There were limited interactions between study and MCFA supplementation for sow performance, litter characteristics, and litter growth. There was an interaction for mummies, pigs weaned,

and daily gain during the suckling period where in study 1, number of mummies tended to be greater ($P < 0.10$) in UNSUP litters and in study 2 number of mummies was greater in SUP litters. Alternatively, in study 1 number of pigs weaned was lesser ($P < 0.05$) in SUP litters and greater in SUP litters in study 2. In study 1, there was no difference in daily gain of piglets from birth to wean, in study 2, SUP pigs tended ($P < 0.10$) to have greater gain. In study 1 sows were heavier ($p < 0.01$) at all weigh periods compared with sows from study 2. Feed intake was greater ($p < 0.01$) in study 2 than study 1. There were no differences between studies for total born, mummies, or liveborn. The number of piglets weaned tended to be greater ($p = 0.10$) in study 2 compared with study 1. Kilograms weaned per sow was greater ($p = 0.02$) in study 2. There were no differences among piglet survivability across studies. Piglet body weight gain at birth, d 7, or weaning was not different across study.

Offspring weight distribution at birth and weaning was impacted by sow dietary treatment. At birth, there was a greater proportion ($P < 0.01$; Figure 2.1) of piglets in the heavy category from SUP litters and more piglets in the average category for UNSUP litters. At weaning, there tended to be a greater proportion ($P = 0.10$), Figure 2.2) of piglets in the light category and heavy from SUP litters and more piglets in the average for UNSUP litters. The categorical change had a similar response where a greater proportion ($P < 0.03$, Figure 2.3) of piglets from SUP litters fell back 1-2 SD. At birth, study 1 had a greater proportion of light category piglets and lesser proportions among the middle and heavy categories ($P < 0.01$, Figure 2.4). At weaning, the category distributions were similar to that observed at birth ($P = 0.01$, Figure 2.5). There were no

differences detected for categorical changes between the two studies ($P = 0.39$, Figure 2.6).

2.4.2 Post-wean Growth Performance

Piglet BW, average daily feed intake, as well as gain to feed ratio at weaning, week 2, week 4, and week 6 was not affected by maternal treatment or post-weaning dietary treatment (Table 2.5). There was no interaction between maternal and post-weaning dietary treatment. There were minor differences among piglet performance by study (Table 2.6). However, study 1 pigs weighed less at week 2 compared to study 2 ($P < 0.01$), but study 1 pigs tended to have increased gain overall in the nursery ($P = 0.06$). Average daily feed intake was lesser at week 2 ($P < 0.01$), week 4 ($P = 0.02$), and overall, in the nursery ($P = 0.02$) resulting in greater ($P < 0.01$) overall gain:feed ratio in study 1.

2.5 Discussion

The objective of this study was to assess the impact of MCFA on sow reproductive performance as well as her offspring in the early nursery period. In the current study, there were minimal effects of MCFA supplementation in sow gestation and lactation diets on sow performance including BW and reproductive performance measures at birth. Other studies similarly noted minimal effects of MCFA supplementation in sow diets on sow reproductive response variables (Chen et al., 2019; Świątkiewicz et al., 2020). However, in this study, sow lactation feed intake was improved with supplementation of MCFA. While this didn't result in an increase in piglet BW, sow lactation intake is often not sufficient to meet the nutrient demand for milk output (Noblet et al., 1998; Eissen, 2000) and the observed increase in sow intake with MCFA inclusion may reduce the need for body tissue mobilization and/or increase milk

output demanded by the suckling piglet. In the current study, MCFA supplementation in sow or nursery diets did not appear to improve piglet performance; however, Lan (2018) reported improved suckling pig growth from sows receiving MCFA supplemented diets. The type of MCFA blend differs amongst commercial products, the active ingredients in Lan and Kim (2018) were: 17% fumaric acid, 13% citric acid, 10% malic acid, 1.2% capric acid, 1.2% caprylic acid, and 57.6% Kaolin ($2\text{SiO}_2 \cdot \text{Al}_2\text{O}_3 \cdot 2\text{H}_2\text{O}$). In this study, the blend contains lauric acid, caprylic acid, formic acid, propionic acid, acetic acid, benzoic acid, sorbic acid, and citric acid. The blend amount as well as the fatty acid length alters the mechanism on how it is absorbed as well as stored. In Study 1, a flu outbreak occurred during the latter half of gestation that impacted overall reproductive performance and growth in suckling and nursery periods as evidenced by the number of mummies, stillborns, and kg weaned experienced in Study 1. A flu outbreak can lead to more susceptibility to other infections as well as hurt reproductive performance (Gumbert et al., 2020). The gut microbiome can impact health status; supplementation of MCFA via the maternal diet has the potential to increase beneficial bacteria. There was no interaction between maternal dietary treatment and study even though the outbreak impacted overall performance, MCFA did not appear to make a measurable difference.

2.6 Conclusions

In conclusion, dietary supplementation of MCFAs in sows as well as their offspring impacted sow lactation feed intake, making it a useful strategy to improve milk output that is demanded by the litter. The inclusion of MCFA had little impact on reproductive characteristics or on the suckling piglet in terms of gain throughout the

lactation period. However, MCFA supplementation in gestation result in more piglets in the heavy category which may suggest higher quality of piglets.

Table 2.1 Experimental diet formulations¹

Item, %	Gestation	Lactation	Nursery ²		
			Phase 1	Phase 2	Phase 3
Corn	81.18	66.04	35.45	53.88	65.84
Soybean meal, 46.5%	14.54	29.85	19.45	27.71	30.35
Dried Whey	-	-	25.00	10.00	-
Fish meal	-	-	7.00	3.00	-
HP 300	-	-	7.00	-	-
Monocalcium phosphate	1.84	1.76	0.50	1.1	1.41
Zinc Oxide	-	-	0.42	0.28	-
Limestone	1.31	1.22	0.32	0.85	1.02
Salt	0.5	0.5	0.03	0.03	0.35
Nursery Vitamin premix	-	-	0.05	0.05	0.05
Sow Vitamin premix ³	0.05	0.05	-	-	-
Trace Mineral premix ⁴	0.15	0.15	0.15	0.15	0.15
L-Lysine HCl	-	-	0.26	0.32	0.4
L- Threonine	-	-	0.11	0.12	0.15
DL – Methionine	-	-	0.20	0.15	0.15
L-Tryptophan	-	-	0.03	0.03	0
Soybean oil	-	-	3.70	2.00	-
Formulated content					
Dry matter	89.5	89.6	92.0	90.6	89.6
ME, kcal/kg	1486.0	1490.0	1593.0	1544.0	1500.0
Crude Protein	13.5	19.4	23.8	21.0	20.2
Crude Fat	3.6	3.5	6.7	2.5	2.9
Ca, %	0.89	0.89	0.85	0.85	0.75
P, %	0.72	0.76	0.78	0.73	0.69
Phos avial-swine	0.44	0.44	0.55	0.45	0.37
SID Lys, %	0.55	0.97	1.51	1.31	1.25
SID Met, %	0.21	0.28	0.57	0.46	0.43
SID TSAA, %	0.43	0.57	0.88	0.76	0.73
SID Thr, %	0.42	0.62	0.94	0.81	0.78
SID Trp, %	0.12	0.2	0.28	0.25	0.21

¹Experimental diets consisted of control (UNSUP) and SUP where MCFA was supplemented at 0.3 % in each diet.

²Feed budget provided per kg/pig: 1.96 (phase 1), 5.91 (phase 2), 12.73 (phase 3).

³Provided vitamins A (22,028,589.2 IU/kg), D3 (3,304,729.3 IU/kg), E (110,231.1 IU/kg), B12 (88.2 mg/kg), menadione (8,818.5 mg/kg), riboflavin (19,841.6 mg/kg), D-pantothenic acid (121,254.2 mg/kg), niacin (110,231.1 mg/kg), folic acid (2,204.6 mg/kg), pyridoxine (6,613.9 mg/kg), thiamine (6,613.9 mg/kg), and biotin (341.7 mg/kg).

⁴Provided copper (1.10%), manganese (2.94%), selenium (200 ppm), and zinc (11%).

Table 2.2 Proximate analysis of gestation and lactation experimental diets¹

	Gestation UNSUP		Gestation SUP		Lactation UNSUP		Lactation SUP	
	Study 1	Study 2	Study 1	Study 2	Study 1	Study 2	Study 1	Study 2
CP	13.90	13.03	12.90	13.33	20.18	19.59	19.51	17.49
Moisture	13.07	13.05	13.20	13.15	13.35	13.20	12.82	12.75
Crude Fat	2.36	2.22	2.13	2.22	2.05	2.20	2.04	1.89
Crude Fiber	2.10	2.26	1.91	1.87	2.20	2.22	2.29	2.45
Ash	4.97	4.92	5.14	4.95	5.70	5.58	5.82	5.36
Nursery phase								
	Phase 1		Phase 2		Phase 3			
	UNSUP	SUP	UNSUP	SUP	UNSUP	SUP		
CP	23.85	22.94	18.25	20.35	20.86	21.05		
Moisture	8.85	9.02	10.95	10.98	11.98	12.21		
Crude Fat	5.30	5.38	3.85	3.68	2.61	2.54		
Crude Fiber	1.95	1.73	2.05	1.73	2.55	2.31		
Ash	6.76	6.94	6.55	5.71	5.04	5.32		

¹ Proximate analysis conducted on subsample of each diet at periodic intervals throughout gestation and lactation. Values represent the average of pooled samples, there was a total of 24 samples across the duration of the trial.

Table 2.3 Reproductive performance of sows across 4 groups provided diets with or without MCFA supplementation

Item	Dietary treatment		Parity			SEM	P-value ¹	
	UNSUP	SUP	0	1	2+		Diet	Parity
Sows on test ² , n	40	37	12	20	45			
Body weight, kg								
Initial (d30 of gestation)	186.2	183.0	153.9 ^a	181.8 ^b	218.2 ^c	3.00	0.56	<0.01
d110	230.6	226.7	208.5 ^a	226.3 ^b	251.1 ^c	3.12	0.49	<0.01
Farrow ³	215.6	216.7	191.9 ^a	215.3 ^b	241.3 ^c	3.40	0.85	<0.01
Wean	211.1	212.7	181.4	207.4	246.9 ^c	3.15	0.77	<0.01
Feed intake, kg								
Total quantity	121.2	131.7	98.4 ^a	131.8 ^b	149.1 ^c	3.2	0.06	<0.01
ADFI	6.13	6.58	5.18 ^a	6.59 ^b	7.29 ^c	0.13	0.05	<0.01
Litter size, n								
Total Born	16.1	15.6	15.0	16.2	16.3	0.48	0.60	0.45
Liveborn	14.5	14.3	13.6	15.1	14.5	0.44	0.76	0.36
Stillborn	1.12	0.97	0.98	0.83	1.31	0.23	0.72	0.50
Mummies	0.42	0.38	0.43	0.29	0.49	0.12	0.86	0.67
Pigs Weaned	13.0	12.7	12.8	13.3	12.5	0.27	0.51	0.21
Kg Weaned	74.0	72.6	69.8	77.7	72.51	1.99	0.67	0.18
Survivability ⁴ , %	91.6	92.5	96.8	91.1	88.28	0.03	0.86	0.41
Piglet body weight, kg								
Birth	1.37	1.39	1.38	1.42	1.34	0.05	0.59	0.29
d7	2.60	2.55	2.60 ^a	2.70 ^b	2.43 ^c	0.05	0.54	0.02
Wean	5.76	5.71	5.51	5.92	5.76	0.11	0.82	0.34
Daily gain, kg								
Birth to d7	0.18	0.17	0.17 ^a	0.18 ^b	0.16 ^c	0.004	0.21	<0.01
Birth to wean	0.22	0.22	0.22	0.23	0.22	0.004	0.56	0.36
Colostrum intake, g ⁵	412.31	419.19	-	-	-	8.44	0.59	-
Colostrum yield, g ⁶	5644.46	5202.83	-	-	-	291.28	0.30	-

¹There were no interactions between dietary treatment and parity.

² Two sows aborted before farrow date, one sow died due to farrow complications, one sow was euthanized in lactation due to a twisted gut, two gilts were removed due to late farrow dates. Removed sows consisted of 1 and 3 for Control and MCFA, respectively.

³ Weighed within 24h after farrowing.

⁴ Calculated on a litter basis as $\text{total weaned piglets} / \text{total piglets born alive} * 100$.

⁵ Colostrum intake (only for study 1 pigs where piglets were weighed after birth of first piglet) for the individual piglet was calculated using the equation described in section 2.3.3 (Theil et al., 2014).

⁶ Colostrum yield was calculated as a summation of the individual piglet intake in each litter

Table 2.4 Reproductive performance of sows across 4 groups provided diets with or without MCFA supplementation and their interaction¹P-value represents interactions between study and maternal dietary treatment.

Item	Study 1		Study 2		P-value ¹	Study		P-value
	UNSUP	SUP	UNSUP	SUP		1	2	
Sows on test, n	23	15	17	22				
Body weight, kg								
Initial (d30 of gestation)	189.3	188.1	183.0	178.0	0.69	188.7	180.5	<0.01
d110	237.8	233.7	223.3	219.7	0.97	235.8	221.5	<0.01
Farrow	221.0	227.1	210.2	206.2	0.34	224.1	208.2	<0.01
Wean	218.5	222.5	203.7	203.0	0.63	220.5	203.3	<0.01
Feed Intake, kg								
Received Quantity	117.2	124.9	125.1	138.5	0.57	121.0	131.8	0.03
ADFI	5.95 ^a	6.30 ^a	6.32 ^a	7.03 ^b	0.19	6.04	6.67	<0.01
Litter Size, n								
Total Born	16.69	14.85	15.41	16.35	0.07	15.77	15.88	0.88
Liveborn	14.45	13.29	14.58	15.26	0.18	13.87	14.92	0.12
Stillborn	1.60	1.34	0.63	0.59	0.76	1.47	0.61	0.02
Mummies	0.64 ^w	0.23 ^{wy}	0.21 ^y	0.54 ^{yw}	0.05	0.43	0.37	0.76
Pigs Weaned	13.14 ^a	11.87 ^b	12.89 ^{ab}	13.53 ^{ac}	0.02	12.51	13.21	0.10
Kg Weaned/Sow	70.93	68.14	77.15	77.05	0.66	69.54	77.10	0.02
Survivability, %	94.0	92.0	89.0	93.0	0.50	93.0	91.0	0.77
Piglet body weight, kg								
Birth	1.30	1.38	1.43	1.40	0.22	1.34	1.41	0.11
d7	2.50	2.54	2.71	2.55	0.23	2.63	2.52	0.18
Wean	5.50	5.77	6.01	5.65	0.08	5.64	5.83	0.30
Daily gain, kg								
Birth to d7	0.17	0.17	0.18	0.17	0.41	0.17	0.17	0.44
Birth to wean	0.21 ^w	0.22 ^{xw}	0.23 ^x	0.21 ^w	0.08	0.22	0.22	0.44

Table 2.5 Weaned pig performance across 4 sow groups (1 nursery room per sow group) provided diets with or without supplementation of medium chain fatty acids¹

Item	Sow Dietary Trt				Piglet Dietary Trt			
	UNSUP	SUP	SEM	P-Value	UNSUP	SUP	SEM	P-Value ²
BW, kg								
Week 2	7.26	7.07	0.30	0.60	7.11	7.22	0.18	0.78
Week 4	12.57	12.29	0.39	0.69	12.53	12.34	0.35	0.81
Week 6	20.82	20.53	0.43	0.71	20.73	20.62	0.39	0.90
ADG, kg								
Week 2	0.13	0.13	0.01	0.99	0.13	0.14	0.01	0.79
Week 4	0.38	0.37	0.02	0.76	0.38	0.37	0.02	0.72
Week 6	0.60	0.61	0.02	0.90	0.60	0.61	0.02	0.98
Overall	0.35	0.35	0.01	0.81	0.35	0.35	0.01	0.83
ADFI, kg								
Week 2	0.30	0.33	0.02	0.45	0.34	0.28	0.02	0.21
Week 4	0.58	0.59	0.02	0.72	0.57	0.59	0.02	0.69
Week 6	0.95	0.99	0.02	0.46	0.98	0.96	0.02	0.59
Overall	0.57	0.59	0.02	0.45	0.58	0.57	0.01	0.77
G:F								
Week 2	0.63	0.60	0.10	0.86	0.54	0.70	0.09	0.42
Week 4	0.67	0.64	0.04	0.74	0.69	0.62	0.04	0.48
Week 6	0.64	0.62	0.02	0.72	0.63	0.64	0.02	0.77
Overall	0.63	0.60	0.01	0.27	0.62	0.62	0.01	0.95

¹ Weaned pigs were placed into a 2x2 factorial experimental design for the 6-week nursery period (3 phase nursery).

² There was no interaction between sow dietary treatment and nursery treatment on weaned pig growth performance.

Table 2.6 Nursery performance of pigs from Study 1 and Study 2 fed diets with or without supplementation of medium chain fatty acids

Item	Study 1		Study 2		<i>P</i> -value	Study 1	Study 2	SEM	<i>P</i> -value
	UNSUP	SUP	UNSUP	SUP					
BW, kg									
Week 2	7.18	7.40	7.04	7.04	0.77	7.29	7.04	0.15	0.47
Week 4	12.26	12.36	12.80	12.32	0.71	12.31	12.56	0.29	0.72
Week 6	21.01	20.97	20.45	20.27	0.94	20.99	20.36	0.33	0.42
ADG, kg									
Week 2	0.11	0.11	0.15	0.17	0.61	0.11	0.16	0.01	<0.01
Week 4	0.36	0.36	0.40	0.37	0.72	0.36	0.39	0.01	0.50
Week 6	0.62	0.61	0.59	0.60	0.75	0.62	0.59	0.02	0.53
Overall	0.37	0.36	0.34	0.34	0.89	0.36	0.34	0.01	0.06
ADFI, kg									
Week 2	0.16	0.15	0.51	0.42	0.37	0.16	0.47	0.02	<0.01
Week 4	0.53	0.55	0.61	0.63	0.96	0.54	0.62	0.01	0.02
Week 6	0.96	0.92	1.01	0.99	0.83	0.94	1.00	0.02	0.15
Overall	0.55	0.54	0.62	0.60	0.95	0.55	0.61	0.01	0.02
G:F									
Week 2	0.68	0.59	0.40	0.80	0.21	0.63	0.60	0.07	0.85
Week 4	0.69	0.64	0.69	0.61	0.82	0.66	0.65	0.03	0.84
Week 6	0.66	0.67	0.59	0.61	0.97	0.66	0.60	0.02	0.14
Overall	0.66	0.66	0.57	0.58	0.77	0.66	0.57	0.01	<0.01

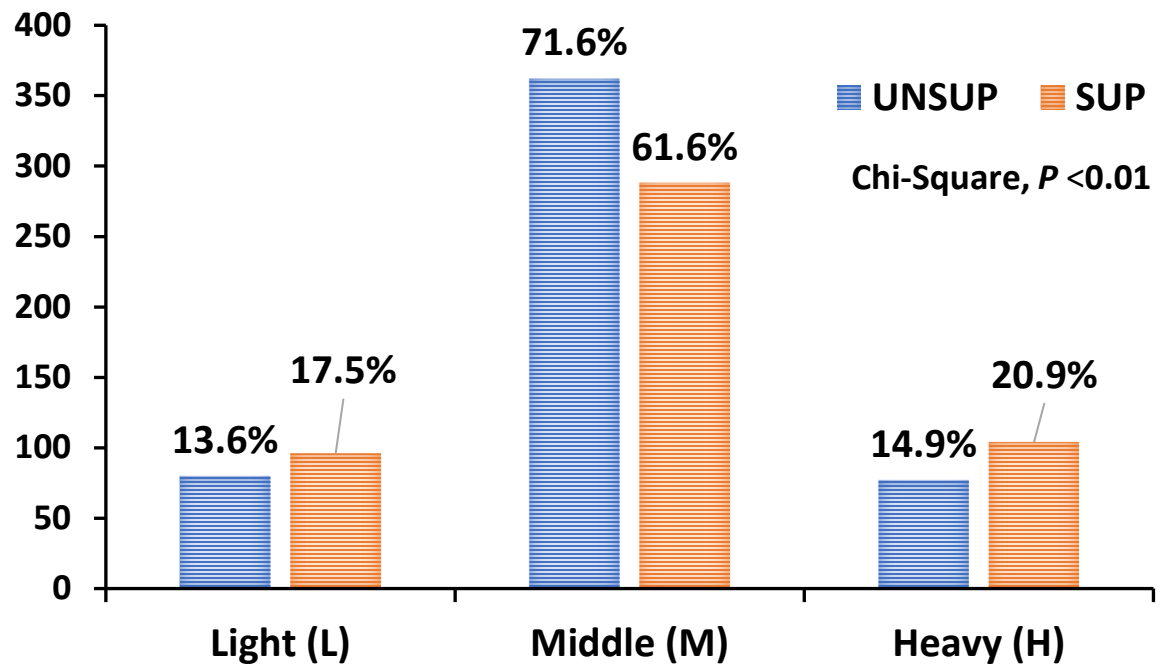


Figure 2.1. Birth weight categorical distribution among litters. MCFA blend was included at 0.3% in the maternal diet. At birth, 'light' represents piglets <1.05 kg, 'middle' represents piglets 1.05 – 1.68 kg, and 'heavy' represents piglets >1.68 kg.

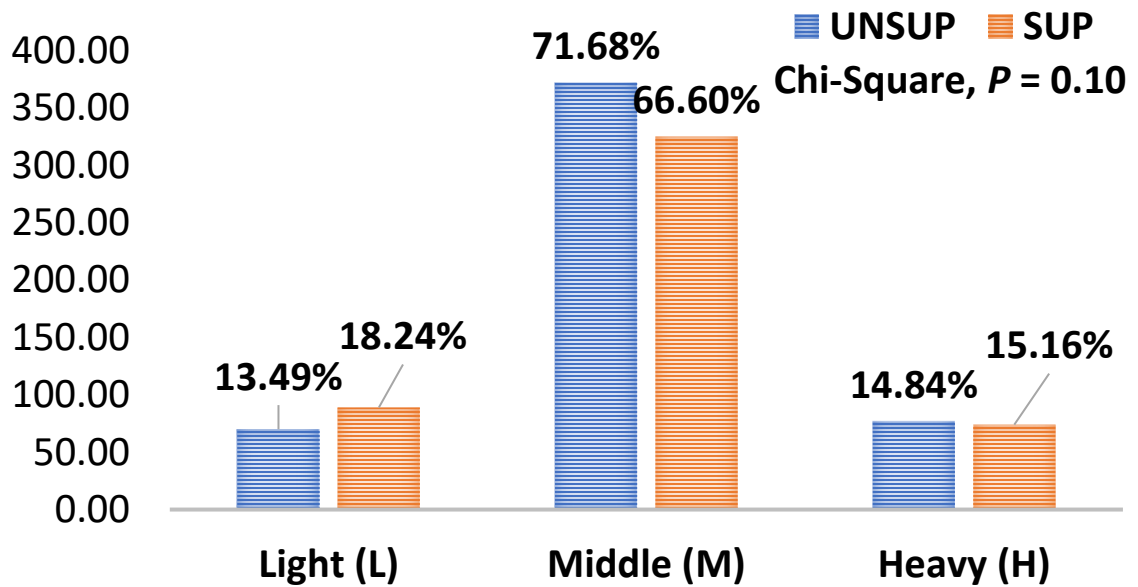


Figure 2.2. Wean weight categorical distribution among litters, ‘light’, ‘middle’ and ‘heavy’ represents piglets <4.43, 4.43 – 7.05, and >7.05 kg, respectively from sows fed diets without (UNSUP) or with medium chain fatty acid supplementation at 0.3% (SUP) in gestation and lactation.

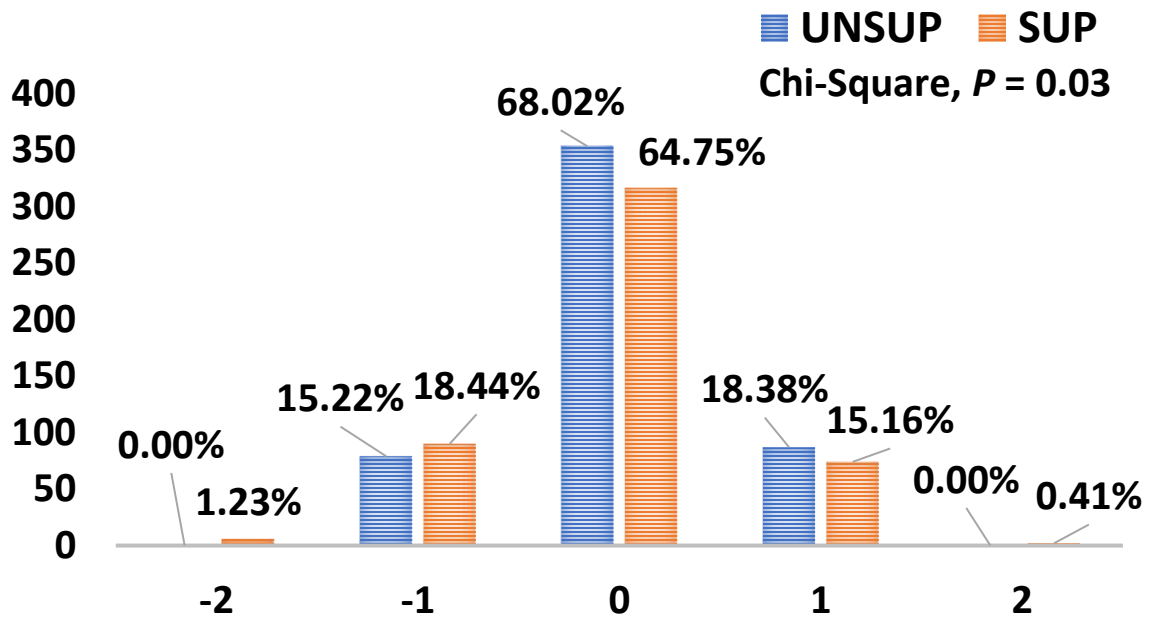


Figure 2.3. Categorical change distribution among litters from sows provided diet without (UNSUP) or with medium chain fatty acid supplementation at 0.3% (SUP) in gestation and lactation.

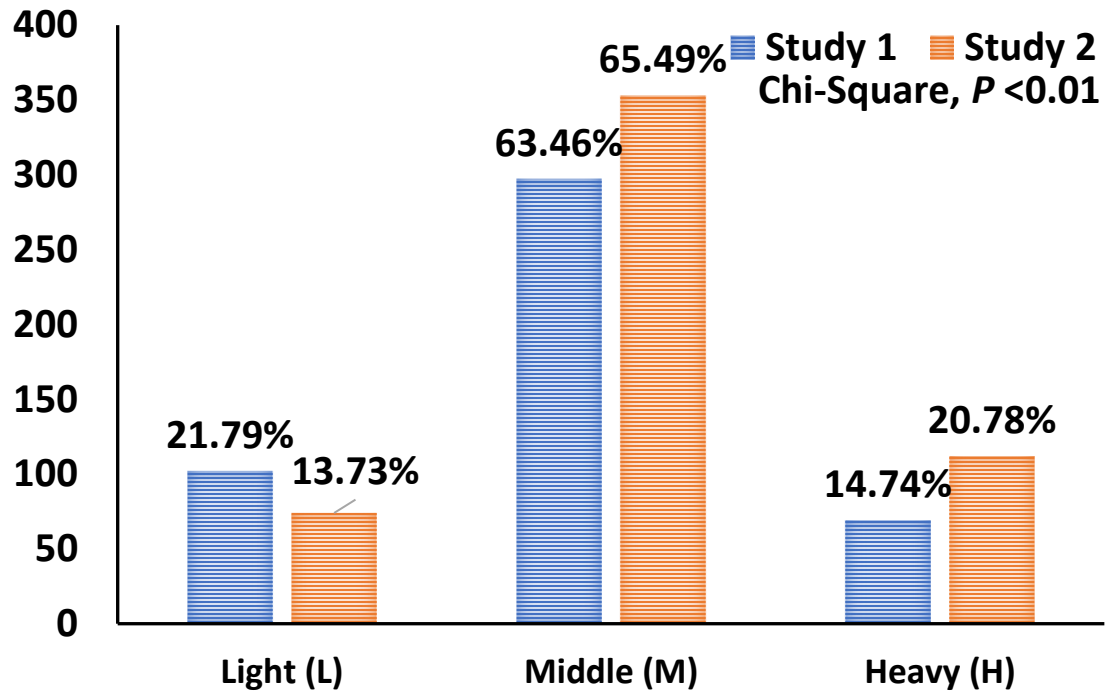


Figure 2.4. Birth Weight Categorical Distribution among litters from study 1 and 2. Sows in study 1 experienced a flu outbreak during gestation.

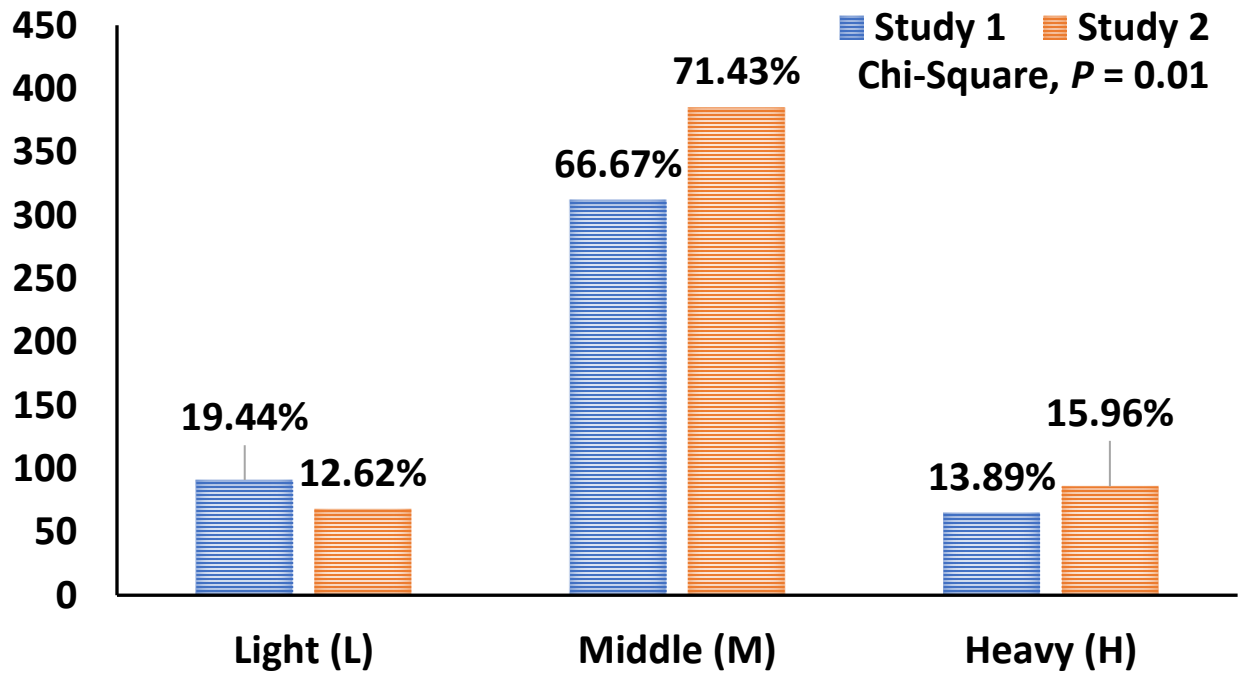


Figure 2.5. Wean Weight Categorical Distribution among litters from Study 1 and 2. Sows in Study 1 experienced a flu outbreak during gestation.

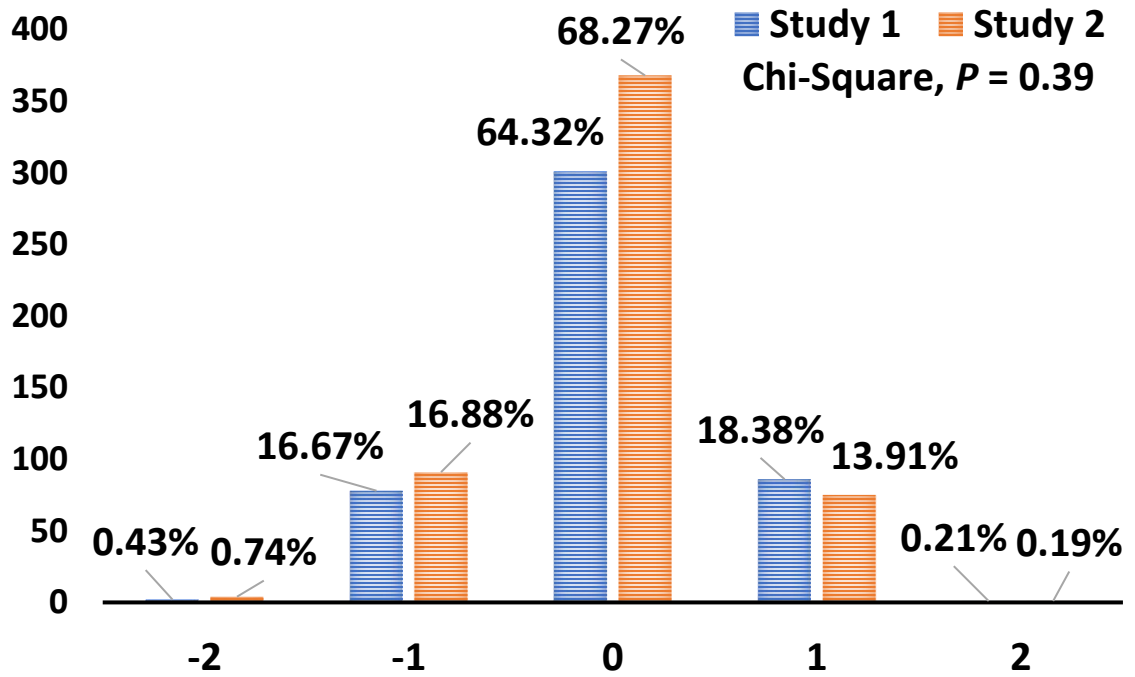


Figure 2.6. Categorical Change Distribution among litters from Study 1 and 2. Sows in Study 1 experienced a flu outbreak during gestation.

3.0 The impact of supplementation of MCFA in sow diets on offspring biological markers and gut health

3.1 Abstract

The objective of this study was to evaluate the impact of diet supplementation of MCFA throughout gestation, lactation, and the nursery phase on biological markers of piglet quality and gut health. Out of the 77 sows described in Chapter 2.0, 32 sows (16 in each of UNSUP and SUP treatment groups) and their offspring were utilized for biological collections and analysis. From each of these litters, 5 piglets (1 heavy, 1 light, 3 average) were selected based upon litter average weight \pm 1 standard deviation. At birth, 'light' represented piglets <1.07 kg, 'average' represented piglets $1.07 - 1.68$ kg, and 'heavy' represented piglets >1.68 kg. Biological variables measured included: piglet serum immunocrit, colostrum and milk composition, IgG in sow colostrum, small intestinal morphology, microbial content of piglet feces in the suckling and nursery stages. The data was analyzed as a randomized complete block using the Mixed procedure of SAS, with sow and weaned pen as experimental unit. UNSUP piglets had higher ($P = 0.01$) piglet immunocrit values (0.16) compared to SUP piglets (0.14). Colostral fat was not impacted by dietary treatment. Colostral protein content was greater ($P=0.04$) in SUP sows compared to UNSUP. The colostral lactose content tended to be greater ($P = 0.06$) in UNSUP sows compared to SUP. Colostral total solids content tended to be greater ($P = 0.07$) in SUP sows than UNSUP. Solids not fat was greater ($P = 0.04$) in SUP sows than in UNSUP sows. Milk composition was not impacted by dietary treatment. Lactobacillus content in piglet feces from SUP sows on d 10 tended to be higher ($p = 0.08$), in the nursery SUP pigs had no statistical differences in percentages

compared to UNSUP pigs on week 2 of the nursery ($p = 0.64$) and on week 6 ($p = 0.66$). Including MCFA in either the maternal or nursery diet, improved colostrum quality and increased beneficial bacteria in the gut microbiota.

Key Words: medium chain fatty acids, gut microbiome, immunocrit

3.2 Introduction

Pigs encounter numerous pathogenic and nonpathogenic bacterial challenges, which result in activation of the gastrointestinal tract (GI) immune system (Liu 2015). The GI is not only an important organ for digestion, absorption, and metabolism of dietary nutrients, but it is also the largest immune organ in the body (Liu 2015). The gut microbiota provides the pig with many functionalities to improve energy harvesting capacity, the production of volatile fatty acids, and enhanced resistance against pathogenic bacteria (Guevarra 2019). Gut development begins early in a pig's life, starting during the fetal period and continues through the first months of postnatal life. Activation of the GI immune system leads to the production of a diverse set of specialized cells and signaling molecules, especially proinflammatory cytokines (Liu 2015). Over-production of these cytokines results in intestinal mucosal injury and dysfunction, and consequently results in poor growth performance (Liu 2015). The sow's microbiota is shared to a large degree by the offspring during the first few days of postnatal life (Bauer et al., 2006); therefore, factors such as diet composition and antibiotic treatment that induce changes in maternal microbiota may have huge effects on piglet gut physiology (Farmer 2015). The current study looks at how supplementation of

MCFA in the maternal diet influences biological markers of piglet quality. Colostrum and milk quality, small intestinal morphology, milk and colostral IgG, sera and colostral immunocrit, and the relative presence of specific bacterium in the piglet feces throughout the suckling period and into the nursery period were considered relevant biological markers of pig growth and quality in this study. There are several factors associated with the maternal environment that have an impact on the development of the piglet during the suckling phase (Farmer 2015). The intra-uterine environment is not only important for fetal development and survival, but it also impacts the post-wean development and health of pigs (Morise et al., 2008). Prior to the birth of the piglet, the gut of the neonate is presumed to be void of microbes but will rapidly undergo shifts to an extremely dense microbial population that continuously experiences microbial succession and establishes an adult-like microbial community (Guevarra 2019). If maternal diet can improve intestinal bacterial composition such as greater proportion of *Lactobacillus* in the young piglet, or lessen *Salmonella*, that sets the piglet immune system up for success. Lan and Kim (2018) investigated supplementation of MCFA blends in sow diet 42 days before farrowing through to weaning (28 days after parturition). Sow fecal microbiota was impacted by MCFA supplementation where an increase in *Lactobacillus* and a decrease in fecal *E.coli* was detected (Lan and Kim, 2018). A similar response was found in the piglet fecal microbiota at the same time points as their mothers (farrowing and weaning).

Colostrum and milk production play an essential role in ensuring piglet survival and growth. There is evidence to support that maternal diet composition may influence colostrum and milk composition which can lead to changes in the gut functions in piglets (Farmer 2015). Świątkiewicz et al., (2020) reported that coconut oil supplementation

influenced fatty acid profile in the milk, as well as improved milk IgM and IgG levels (Świątkiewicz et al., 2020).

The importance of this study was to get a better understanding of how sow feeding strategy may be used to enhance biological markers in the suckling and post-weaning periods of growth.

The objective of this study was to evaluate the impact of diet supplementation of MCFA throughout gestation, lactation, and the nursery phase on biological markers of piglet quality and gut health.

3.3 Materials and Methods

3.3.1 Animal management, diets and feeding

Details on sow groups, diets formulations, sow and weaned pig daily care are provided in Chapter 2.0, sections 3.1 and 3.2. 5/20 sows tested positive for *Streptococcus Suis* when nasal samples were collected on day of allotment (d 28 of gestation).

3.3.2 Data collections, chemical analyses, and calculations

In concert with BF at d 28 (pregnancy confirmation) of gestation, d 110 of gestation, and at weaning, blood samples were collected from 10 sows/treatment in study 1 and from 6 sows/treatment in study 2 via jugular venipuncture using a 3.81 cm x 20 gauge bleeding needle into a no additive blood collection tube (BD Vacutainer, Franklin Lakes, NJ). Samples were kept on ice at collection, centrifuged at 5,000 x g for 10 minutes, transferred to 1.5 mL microcentrifuge tubes (Thermo Fisher Scientific, Waltham, MA) and then stored at -80°C for later analysis. Nasal swabs were collected at d 28 of gestation, d 110 gestation and at weaning; sterile swabs (BD BBL Culture Swab,

Franklin Lakes, NJ) were placed approximately 5 cm into a nasal cavity, rotated clockwise twice and repeated in the other nasal cavity. Swabs were placed in microcentrifuge tubes and stored at -20°C. Sow fecal samples were collected on d 28 of gestation, d 110 of gestation and at weaning using clean disposable gloves and lubricant to collect approximately 10 mL stool from each sow. Samples were stored at -20°C for later analysis.

Following birth of the first piglet and prior to suckling, colostrum was collected using gentle stripping from all teats for a total volume of 40 mL in sterile conical tubes (Fisher Scientific, Pittsburgh, PA). At d 4 or 5 of lactation, a milk sample was collected; piglets were removed for one hour, then 2 mL of oxytocic principle (Oxytocin, Aspen Veterinary Resources, Liberty, MO) was administered (2.54 cm x 20 ga needle) intravaginally and the same technique and total volume previously noted for colostrum was utilized. Piglets were returned to the dam following collections. Colostrum and milk were stored at -20°C until further use. On d2 of age, a 1 mL blood sample was collected from the mammary vein of five piglets from each litter based on birth weight category (one 'light, three 'average' and one 'heavy' piglet). Samples were centrifuged at 5,000 x g for 10 minutes, transferred to 1.5 mL microcentrifuge tubes (Thermo Fisher Scientific, Waltham, MA) and then stored at -80°C for later analysis.

Serum and colostrum immunocrit ratio were based on the method of Vallet et al. (2013). Briefly, serum was diluted in a 1:1 ratio with 40 % ammonium sulfate in distilled water. The newly diluted sample was loaded into a microcapillary tube and placed into a hematocrit centrifuge (MX12 PCV Centrifuge, LW Scientific, Lawrenceville, GA) and centrifuged at 12,000 x g for 10 minutes. The length of the Ig precipitate and the length of diluted colostrum was measured and divided to determine the immunocrit ratio (IR). In

conjunction with piglet sera samples, immunocrit was evaluated in colostrum. A modified methodology from Vallet and Miles (2017) was used. Briefly, colostrum samples were diluted in a 1:1 ratio with 1 % bovine serum albumin (1 mL: 9mL Saline; Fisher BP6751) in 0.9 % saline. In duplicate, diluted colostrum samples were combined with 40 % (wt/vol) ammonium sulfate in distilled water to precipitate immunoglobulins and then loaded into a hematocrit centrifuge and centrifuged at 12,000 x g for 10 minutes. Immunocrit ratio was determined as the ratio of the precipitate length divided by the total length of diluted colostrum, then doubled to account for prior colostrum dilution. Colostrum and milk samples were analyzed for protein, lactose, total solids, and fat (Division of Regulatory Services, University of Kentucky, Lexington, KY). Microbial analysis of piglet fecal samples at d 10, d 24, and d 63 of age were completed to determine the relative proportion of *Lactobacillus*, *Escherichia coli*, and *Salmonella*.

Isolation of fecal microbial DNA occurred with the use of DNeasy PowerSoil Kit (MO BIO Laboratories, Qiagen, Venlo, Netherlands). Quantitative polymerase chain reaction (qPCR) was performed based on the methods of Ortman et al (2020) in duplicate using 10 ng of microbial DNA per individual reaction, which also included SsoAdvanced Universal SYBR Green Supermix reagen (BioRad, Hercules, CA, USA) (1X final concentration) as well as target specific forward and reverse primer pairs (4 μ M/primer). The qPCR was performed on a Mx3005P Thermal Cycler (Stratagene, San Diego, CA, USA), starting with a hot start (3 min, 98°C), followed by 40 cycles of a ‘fast-2 step’ protocol, with each cycle consisting of a denaturation (10 s 98°C) step and an annealing/synthesis (30 s, 60°C) step. Threshold cycle (Ct) values were determined by the MxPro qPCR Software (Stratagene, San Diego, CA, USA), using default settings. The relative proportion of *Lactobacillus* was calculated based upon an equation: $\Delta\Delta C_t = C_{tLactobacillus\ sample} - C_{tUniversal\ sample}$.

At weaning, one of the “average” piglets from the selected litters was euthanized via cervical dislocation using a penetrating captive bolt gun. After euthanasia, the entire small intestine was removed, midpoint located, and 10 cm of the ileum was removed and placed in a 15 mL conical tube containing 5 mL of 10% buffered formalin solution for slide mounting and later histomorphology analysis. Samples for histology analysis were sent to the Animal Disease Research and Diagnostic Laboratory at South Dakota State University for staining with hematoxylin and eosin. Villous height (VH; from the top of the villi to the villous-crypt junction) and crypt depth (CD; from the villous-crypt junction to the base) were measured at 4x magnification using a microscope (Micromaster[®], Fisher Scientific, Waltham, MA, USA) equipped with a 0.55x wifi camera eyepiece (MC500-W 3rd Gen., Meiji Techno Co. LTD., Saitama, Japan) and Micro-Capture software (Meiji Techno Co. LTD., Saitama, Japan) in 20 well-oriented villi and crypt columns. The villous height-to-crypt depth (**VH:CD**) ratio was calculated.

One mL of each individual sample of milk and colostrum was centrifuged at 15,000xg for 20 min at 4°C. After samples were spun, the layer of fat was discarded, and the remaining liquid was diluted to 1:500,000 (colostrum) and 1:4,000 (milk) with the dilution buffer solution provided. The concentration IgG was determined by using a Pig IgG ELISA kit (Bethyl Laboratories Inc., TX, USA).

3.4 Results

3.4.1 *Biological Markers and the Gut Microbiome*

Serum immunocrit was lower ($P = 0.01$) in SUP piglets compared to UNSUP piglets and no difference in colostrum immunocrit. Colostrum fat content was not impacted by maternal dietary treatment. Colostrum protein content increased ($P = 0.04$) in SUP sows compared to UNSUP. Colostrum lactose content in UNSUP sows tended to be greater ($P =$

0.06) than to SUP sows. SUP sows tended ($P = 0.07$) to have increased colostrum total solids and increased ($P = 0.04$) colostrum solids not fat content. Milk fat, protein, lactose, total solids, solids not fat was not affected by dietary treatment. Colostrum IgG as well as milk IgG was not impacted by supplementation of MCFA in the maternal diet. Similarly, jejunal morphology of weaned piglets (villus height, crypt depth, and their ratio) was also not impacted by maternal dietary treatment.

The *Lactobacillus* content in feces from the suckling period (d 10 of age) showed that SUP piglets tended to have a greater proportion ($P = 0.08$) compared to UNSUP piglets. In week 2 of the nursery period, SUP pigs had maintained a 39% greater proportion of *Lactobacillus* in feces, compared to UNSUP pigs. In phase 3 of the nursery (week 6), the SUP pig still had 19% greater proportion of *Lactobacillus* than UNSUP pigs.

3.5 Discussion

The objective of this study was to evaluate the impact of diet supplementation of MCFA throughout gestation, lactation, and the nursery phases on biological markers of piglet quality and gut health. Piglet quality was evaluated based on piglet/colostrum immunocrit, colostrum and milk composition, as well as IgG content in colostrum while milk and piglet gut health assessment was based on small intestinal morphology and the relative abundance of *Lactobacillus*, *Salmonella*, and *E.coli* in the feces.

An improvement in colostrum quality based on protein, total solids, and solids not fat content with MCFA supplementation in sow gestation and lactation diets was observed indicating that SUP piglets were receiving a more nutrient dense colostrum. The main roles of colostrum are to provide the neonate with energy and passive/active defense mechanisms (Salmon et al., 2009). Modifications to the colostrum in the current

study could be expected to be beneficial to the progeny. Total solids were increased by MCFA and previous research suggests that supplemental dietary energy can affect total solids content in colostrum (Farmer 2015). Total solids are the entire residue (fat, lactose, protein, minerals) that is left over after the water has been evaporated from the sample. The increase of total solids in SUP sow colostrum is likely correlated to the increase in colostrum protein observed. To understand how colostrum protein is manipulated, first one needs to know when colostrumogenesis takes place. The transfer of IgG from sow plasma to lacteal secretions begins approximately 10 d before parturition (Farmer and Quesnel 2009). During this time period, tight junctions between mammary epithelial cells are known to be 'leaky', which allows the paracellular transfer of constituents from maternal plasma (Farmer and Quesnel 2009). Medium chain fatty acids may be linked to the uptake of IgG by mammary glands leading to an increase in colostrum protein (Świątkiewicz et al., 2020). Lactose content in colostrum from UNSUP sows was greater and Farmer (2015) discussed that lactose is the major carbohydrate in sow milk, but it is also the major osmole in milk. The energy requirement of piglets is very high due to the physical activity and demand from thermoregulation (Theil et al., 2014). An increase of lactose which is a form of energy for the piglet, can lead to an increase in more colostrum consumption as well as better control of thermoregulation. The improvement of colostrum lactose that was observed in the UNSUP samples may explain the numerically higher weights on d 7 of age for those piglets. Colostrum protein was increased for SUP sows; however, serum immunocrit values were decreased. Immunocrit ratio is an inexpensive method to measure passive transfer of IgG from dam to piglet (Vallet et al., 2015). Vallet et al. (2015) reported that an immunocrit ratio of 0.125 coincided with high piglet

survivability. The values reported in this current study are well above this value, suggesting that colostrum was not a limiting factor for piglet growth or survival in this study. This suggests that SUP increase in colostrum protein may not translate to an increase in IgG. Another implication of MCFA inclusion in sow diet may be allocation of dietary nutrients to the production of colostrum proteins. Supplementation of MCFA in the gestation diet serves as a good tool to help aid piglet immunity; increasing colostrum protein/immunoglobulins sets the piglet up for a better chance of survival within those first few days of life.

Supplementation of MCFA provide the opportunity to increase the quality of colostrum and may have the potential to increase colostrum yield. Hasan et al., (2019) investigated the factors affecting sow colostrum yield and quality and discovered that one percentage unit increase of colostrum protein resulted in 5.7 g decrease of piglet colostrum. Historically, colostrum consumption determines piglet survival (Quesnel et al., 2012), well what if altering colostrum quality or more particularly increasing the protein content of colostrum could make up for those piglets that do not consume the amount needed. This study suggests MCFA allow for an increase in colostrum protein. Although serum immunocrit was decreased in SUP sows, it could suggest that the colostrum protein impacted was not IgG, and may be a different immunoglobulin or other protein components. The immunoglobulins that could have been affected by the nutritional changes are IgA, IgG, and/or IgM. The precursors for sows to generate immunoglobulins is to have antigens circulating in the blood stream. The major whey proteins include β -lactoglobulin, α -lactalbumin, whey acidic lactoferrin, serum albumin, and immunoglobulins (Theil and Hurley, 2016). Quesnel et al., 2012 noted that the two

components in colostrum that are the most sensitive to nutritional changes are immunoglobulins and lipids. The use of *Lactobacilli* as a probiotic in swine has been gaining attention due to their ability to improve growth performance as well as carcass quality (Valeriano et al., 2017). The gut microbiome affects various nutritional, immunological and physiological functions in mammalian hosts (Valeriano et al., 2017). Profiling of microbial community structure after probiotic, prebiotic and symbiotic administration has led to identification of specific bacterial groups associated with a healthy gut (Chae et al., 2016). *Lactobacillus* has been identified as one of the core genera in the GIT of pigs (Valeriano et al., 2017). Certain members of the genus influence intestinal physiology, regulate the immune system and balance the intestinal ecology of the host (Valeriano et al., 2017). This research study suggests that supplementing MCFA either in the maternal diet and the nursery diet can alter the bacterial populations in the piglet GIT. In the suckling phase, piglets from SUP sows had a greater proportion of *Lactobacillus* than UNSUP sows. However, in the nursery phases *Lactobacillus* content was still relatively higher in SUP than UNSUP piglets suggesting a slower decline in *Lactobacillus* may be beneficial to the piglet to help aid in the stress from weaning. Gut health is also associated with villus height and crypt depth, however, in this study, those variables were not impacted.

3.6 Conclusion

Although the mechanism by which colostrum protein was enhanced is unclear, the apparent increase in colostrum protein in lactating females as a result of supplementing MCFA has the potential to improve piglet survival via colostrum quality. Maintaining higher levels of *Lactobacillus* within the gut from the suckling period to two weeks into

the nursery is an indicator of improved gut health and may provide some protection during the stressful period immediately after weaning.

Table 3.1 Biological markers of pig health

Item	Dietary treatment		SEM	P-value
	Control	MCFA		
Piglet Immunocrit ¹	0.16	0.14	0.005	0.01
Colostrum Immunocrit	0.49	0.48	0.03	0.75
Colostrum Composition, %				
Fat	5.20	5.33	0.23	0.70
Protein	16.47	17.67	0.39	0.04
Lactose	2.44	2.23	0.08	0.06
Total Solids	28.64	29.94	0.48	0.07
Solids not Fat	22.25	23.36	0.37	0.04
Colostrum IgG	195.98	183.90	13.53	0.59
Milk IgG	1.63	1.67	0.17	0.18
Milk Composition, %				
Fat	7.18	6.74	0.33	0.35
Protein	4.54	4.63	0.09	0.49
Lactose	5.53	5.65	0.06	0.18
Total Solids	18.26	17.99	0.30	0.54
Solids not Fat	10.46	10.65	0.13	0.33
Jejunal morphology ² , μm				
Villus height	471.75	472.16	12.60	0.98
Crypt depth	119.76	121.26	3.78	0.79
Villus height: crypt depth	3.97	3.93	0.14	0.86

¹Analysis conducted on serum from 5 piglets each from 16 litters/dietary treatment.

²Analysis conducted on mid jejunal tissue from one average piglet in each of the 16 litters/treatment at weaning.

Table 3.2 Relative abundance of *Lactobacillus* genus in the feces of the offspring before or after weaning.

Item	Dietary Trt		SEM	P-Value
	UNSUP	SUP		
<i>Lactobacillus</i> , % ¹				
Suckling, d10 of age	0.20	0.46	0.07	0.08
Week 2	0.23	0.32	0.09	0.64
Week 6	0.21	0.25	0.06	0.66

¹*Ecoli* and *Salmonella* were deemed undetectable by qRT PCR.

4.0 General Discussion

This research work assessed inclusion of MCFA in gestation and lactation diets as well as in the first 3 phases of the nursery diet program on reproductive characteristics, offspring biological health markers, and sow and offspring growth performance. It was hypothesized that the inclusion of MCFA in the maternal diet would provide her offspring with additional benefits in the suckling and nursery phases of life. Overall MCFA supplementation did provide additional biological benefits to the suckling piglet and nursery pig.

The supplementation of MCFA throughout gestation and lactation led to an increase in lactation feed intake. Feed intake is a major factor for milk output, increasing feed intake could mean that sows do not have to mobilize as much body tissue to make up for where nutrient intake from feed is lacking. Although lactation only represents 15 to 20% of the sow's reproductive cycle, it is the most metabolically demanding stage of production (Tokach et al., 2019). During lactation, the priority of the sow is to sustain milk production for the large and fast-growing litter of piglets, but nutrients required to meet milk production are not often attained by voluntary feed intake (Tokach et al., 2019). Supplementation of MCFA in the maternal diet increased colostrum "value" with an increase in colostrum proteins and solids not fat. This improvement can contribute to achieving a high-quality piglet from birth to weaning and decreasing piglet prewean mortality. An increase in colostrum protein and solids not fat could be contributing to replenishing hepatic and muscle glycogen stores that are utilized for thermoregulation (Theil et al., 2011). Increasing colostrum protein when sows are exposed to a virus or a pathogen such as the flu, may provide the piglet with protection from various pathogens.

Lactobacillus abundance tended to be higher in suckling piglets when supplemented through the maternal diet. *Lactobacillus* is known as a beneficial bacterium (Wang et al., 2018), and MCFA inclusion in the maternal diet as well as supplementation in the nursery phases impacted the relative proportion of it within the offspring's gut. A push the swine industry is facing currently, is the growing pressure to reduce the usage of antibiotics (De Greeff et al., 2020). A potential alternative to antibiotic use is altering the microbial populations within the gut (De Greeff et al., 2020). Understanding how to manipulate gut microbial succession in pigs may provide benefit during stressful events such as weaning.

Supplementation of MCFA in gestation provides the opportunity to impact the development of the offspring gut (Chowdhury et al., 2007; Hooper, 2004; Farmer, 2015).

At weaning, the piglet undergoes severe stress, which can impact growth performance. Feed intake is the biggest concern once piglets are weaned because pigs are shifting from a liquid diet to a dry meal diet, so driving the pigs to eat is a large focus in the nursery period. Feed intake and growth performance was not affected by maternal or nursery dietary supplementation. It's not known if the lack of difference in growth performance of piglets in the suckling and nursery phases was due to the characteristics of the blend of this MCFA or whether the inclusion level was insufficient because previous research has suggested that inclusion can improve piglet growth performance. The sow feed intake information is intriguing and needs to be investigated more with different blends of MCFA to understand what individual/combinations of MCFA influences sow feed intake. For example, could including MCFA in late gestation instead of early, result in the same improvements in colostrum composition? The gut microbiome

and the bacteria it contains needs to be investigated further to understand the mechanisms of how MCFA impacts these populations. An additional item to be investigated to fully understand MCFA supplementation on the sow's energy status and how she utilizes it, would be the farrowing duration. Is farrowing duration impacted by supplementation of MCFA when viewing MCFA as an energy source?

In conclusion MCFA supplementation provided benefits beyond growth performance. Feeding MCFA in gestation and lactation provides the producer with an opportunity to try and improve performance in lactation and also at weaning. Medium chain fatty acids can be a supplement used for providing additional protection against common pathogens and helps provide beneficial bacteria for the piglet before and after weaning.

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