A Thesis

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Effects of Different Levels of Crude Protein on Physiological Responses, Litter Performance, Blood Profiles, Milk Composition and Odor Emission in Lactating Sows

사료 내 조단백질 첨가수준이 모돈 및 자돈의 생리적 변화, 포유능력, 혈액성상, 돈유성분, 악취발생에 미치는 영향

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Summary

Dietary protein plays a very important role in maintaining healthy growth and normal immune function in animals. Excessive protein increases the cost of feed and the burden on the farm, and increases nitrogen emissions in manure and urine, which contribute to environmental pollution. Dietary protein level is an important factor influencing nitrogen utilization and lactation performance of sows. Additionally, variation in sow intake during different seasons may also affect protein intake, nitrogen utilization and reproductive performance of lactating sows. Therefore, this experiment was conducted to investigate the effects of dietary crude protein level on physiological response, reproductive performance, blood profile, milk composition and odor emission of lactating sows in summer. A total of 48 F1 multiparous sows (Yorkshire × Landrace) with average body weight (BW) of 238.0 ± 4.65 kg, average backfat thickness of 20.95± 1.38mm, and an average parity of 4.65 ± 0.44 was used in a 3wk trial at a research farm located in Eum seong, Korea. All sows were allotted to one of six treatments considering body weight (BW), backfat thickness, and parity in completely randomized design (CRD) with 8 replicates. All experimental diets for lactating sows were formulated based on corn soybean meal. Treatments are as followed: 1) CP15: corn–soybean-based diet containing 15% CP, 2) CP16: corn– soybean-based diet containing 16% CP, 3) CP17: corn–soybean-based diet containing 17% CP, 4) CP18: corn–soybean-based diet containing 18% CP, 5) CP19: corn–soybean-based diet containing 19% CP, and 6) CP20: corn–soybean-based diet containing 20% CP. All other nutrients in experimental diets were formulated to meet or exceed the NRC requirements (2012). There were no significant differences in body weight, backfat thickness and average daily feed intake of lactating sows between dietary crude protein levels throughout the trial period.

Similarly, there was no significant difference in the effect of increasing dietary crude protein level on the number of live births, average litter weight and average piglet weight of sows. Casein content in milk decreased linearly with increasing crude protein level in the diet ($P<0.05$). The increase of protein level in the diet did not have a significant effect on the blood biochemical parameters of piglets. The effect of changing the crude protein levels in the feed on triglyceride, creatinine and glucose concentrations in the blood of sows was no significantly different. However, as the crude protein level increased, there was a linear increase in urea nitrogen and total protein content levels in the blood of lactating sows (Linear, $P=0.01$). The concentrations of odor gases, including amine, ammonia and hydrogen sulfide, increased linearly when sows were fed diets with higher levels of the dietary CP (Linear, $P=0.01$). Increasing the protein level in the diet did not improve the performance of lactating sows and piglets. Consequently, this experiment demonstrated that crude protein level of 15% in sow diet help to reduce the odor gas emissions from manure as well as feed cost of sows without any detrimental effects of reproductive performance of sow.

Keywords: Dietary crude protein, Blood profiles, Litter performance, milk composition, Piglets, Lactating sow

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List of Abbreviations

ADG	:	Average daily gain
ADFI	:	Average daily feed intake
ASF	:	African swine fever
ANOVA	:	Analysis of variation
ARC	:	Agricultural Research Council
BW	:	Body weight
BUN	:	Blood urea nitrogen
FCR	:	Feed conversion ratio
HPLC	:	High performance liquid chromatography
NRC	:	National Research Council
RCB	:	Randomized complete block
SAS	:	Statistical analysis system
SBM	:	Soybean meal
SEM	:	Standard error of the mean
TP	:	Total protein

I. Introduction

The feeding management of lactating sows plays a pivotal role in the overall feeding management process of sows, and the feeding management level at this stage directly affects the production efficiency of pig farms. The reproductive performance of modern lactating sows has been greatly improved through genetic selection targeting improved reproductive performance. However, this improvement has been accompanied by a reduction in feed intake, thereby imposing a specific production burden on the sow and necessitating precise provision of nutrients. The nutritional needs of lactating sows are characterized by a lack of lactate and nursing piglets, as well as to maintain normal body metabolism and growing body tissues. The composition of sow's milk and lactation quantity is closely dependent on the nutrients supplied through feed. In contrast, the quantity and quality of lactation directly affect the weaning weight and survival rate of piglets. Therefore, the vital goal of sow nutrition during lactation is to prevent excessive weight loss in sows, improve their lactation performance, and facilitate their prompt return to estrus after weaning.

Korea is located in a mid-latitude temperate climate zone with hot and humid summers under the influence of high pressure from the North Pacific Ocean. Producers have been facing challenges with decreased production performance in sows under high-temperature environments. There are two possible reasons for the decrease in production performance: firstly, the high-temperature environment in summer causes a decrease in the sow's feed intake, which leads to the reduction of production performance; secondly, the high-temperature environment changes the blood circulation of the sow to increase heat dissipation, thus decreasing the utilization of nutrients in the mammary gland, or the high temperature directly affects the milk production and therefore the production performance (Mullan, 1992; Messias, 1998). The decrease in animal feed intake due to high temperatures may be to reduce the heat gain from feeding (Renaudeau, et al, 2005), thus reducing total heat production and alleviating the adverse effects of heat stress on animals. While the livestock industry has achieved greater development, it has also caused a more serious impact on the ecological environment. Taking growing pigs as an example only 50% of the ingested nitrogen is deposited in their bodies, while the excess nitrogen is excreted into the environment through their excrement**.** Progress has been made in the application of low-protein diets to growing and fattening pigs, and studies have shown that reducing dietary protein levels by 2-3% does not affect growth performance, but nitrogen excretion is reduced by 20-30% (Lopez et al. 1994; Tuitoek et al. 1997; Liu et al. 1999; Yang et al., 2000).

This experiment was conducted to study the effect of different protein levels in diets on the performance of lactating sows on responses, litter performance, blood profiles and milk composition in summer.

II. Review of Literature

1. Introduction

Korea is a major importer of grain, with annual imports totaling 18 million tons. This includes a variety of gains, such as soybean meal. The amount fluctuates between four and seven trillion won, contingent on the cost of international grain items. According to 2021 data from the National Statistical Office, feed costs account for between 50 and 60 percent of the total costs (Table 1). Since the beginning of 2022, the price of international cuisine has been increasing, and the instability of the world food industry continues to be unpredictable due to worldwide factors. In Korea, the expenditure on feed for pig farming comprises approximately 60% of the total pig production cost. The increasing cost of raw materials are expected to impact the production expenses associated with manufacturing animal feed. The price of manufacturing formula feed is determined by production costs, which constitute a substantial portion of the manufacturing formula feed costs, thus influencing the quality of the feed. The ratification of commercial agreements such as the Free Trade Agreement has opened new prospects for the livestock sector. The prices of imported livestock products are more competitive than domestic products, owing to superior foreign production technology and lower production costs. To guarantee the success of protecting local livestock products and rejecting foreign livestock products, it is essential to have a dependable and consistent source of feed. Having a reliable supply of feed is essential to the success of the livestock industry, as it allows livestock owners to manage their operations with certainty. Moreover, having a steady supply of feed ensures that livestock products remain of the highest quality. With a dependable source of feed, the livestock industry is able to defend itself against foreign competition adequately.

As society progresses and technology develops, people become more conscious of the need to safeguard the environment. The farming industry has come under increased scrutiny due to its potential environmental impacts. There are legitimate concerns that the industry contributes to water and air pollution. For the swine industry to remain viable in the current environment, it must demonstrate a commitment to eco-friendly practices for the long term.

The primary cause of nitrogen contamination caused by pig farms is the breakdown of crude protein in the feed consumed and the decomposition of amino acids within the animal's body. Jongbloed A W, Previous research has indicated that pigs can only use 30% of the nitrogen they consume, with the remaining 70% being expelled in their urine and feces.

Table 1. Changes in total pig production cost and feed cost by year

(Unit: won)

Year	Total pig production cost (A)	Feed cost (B)	B/A
2012	331,097	174,181	52.61%
2013	323,645	179,507	55.46%
2014	313,608	177,734	56.67%
2015	307,077	167,173	54.44%
2016	301,273	158,804	52.71%
2017	321,006	166,284	51.80%
2018	323,298	164,967	51.03%
2019	323,225	167,929	51.95%
2020	315,079	172,312	54.69%
2021	339,629	191,456	56.37%

(National Statistical Office, 2022)

Table 2. The Nutritional requirements of lactating sows

	NRC(1998)	**NRC(2012)**
ME (kcal/kg)	3,265	3,300
CP (%)	19.2	13.43
Lys (%)	0.90	0.84
Met (%)	0.23	0.22
Thr (%)	0.56	0.53
Trp (%)	0.17	0.16
Ca (%)	0.75	0.76
Total P (%)	0.60	0.65

2. Sow protein nutrition

Protein plays a vital role in the pig. One of the main physiological functions of protein is to act as a component of the body's structure. The muscles, nerves, connective tissue, skin, coat, and blood of pigs are composed of protein. In addition, proteins regulate the body's metabolism as the main component of enzymes. Some proteins have hormonal functions and regulatory functions for the d metabolism of nutrients. For example, insulin is involved in regulating blood glucose metabolism and reduces the glucose level in the blood. Protein can also be broken down directly in the body and oxidized for energy to make up for the lack of energy. The protein ingested by pigs through feeding refers to feed protein, of which the natural protein, due to its substantial molecular weight, must be transformed into small molecules of amino acids and small peptides through the decomposition of the digestive tract before it can be absorbed into the blood through the intestinal mucosa. Protein is metabolized in the pig's body through continuous synthesis and breakdown. Protein degradation involves the breakdown of somatic proteins into amino acids, further broken down into amino and carboxylic acids. These enter the tricarboxylic acid cycle to synthesize amino acids or are stored in the body as energy or oxidized for energy supply. During lactation, inadequate protein or energy intake can lead to decreased milk production and impair the subsequent reproductive performance, an adverse effect particularly pronounced during the summer months and often causes seasonal infertility (Tokach et al, 1992; Kotesu et al., 1996). An animal's need for protein is amino acids. The type, level, and proportion of limiting amino acids will directly affect the diet's nutritional value and, thus, the animal's performance. In the diet formulation, the addition of limiting amino acids creates the balance between the diet's amino acid content and the body's amino acid requirement, which can maximize feed utilization and improve production performance.

2.1 Requirement of protein

Protein is one of the most essential animal nutrients, playing a crucial role in their productive lives. It helps build and repair tissues, creates enzymes and hormones, and serves as an energy source. Protein is also essential for producing antibodies and helps maintain the body's balance of fluids and acids. Additionally, it helps animals to transport oxygen to their cells and plays a role in the metabolism of carbohydrates and fats. Without protein, the body's organs and tissues would not be able to function correctly, so animals need to consume a diet high in protein to remain healthy and productive. When the requirements for essential amino acids (EAA) and total nitrogen are met, the amount of dietary crude protein (CP) can be decreased in the production of pigs since pigs' need for dietary protein is essentially a need for amino acids (AA). Because corn only contains a small quantity of lysine, classic corn-soybean meal (CSBM) diets featured a more significant amount of soybean meal (SBM) to satisfy the pigs' need for lysine, which led to high CP levels. High-protein (HP) diets decreased nitrogen utilization efficiency because they caused excesses of other EAA and excess nitrogen to be excreted in the urine and feces.

Moreover, the gut's health might be harmed by protein fermentation in the hindgut. Crystalline amino acids (CAA) supplementation and dietary CP reduction of 2% to 4% from NRC (1998) recommendations have been shown to boost nitrogen utilization, decrease feed costs and nitrogen excretion, and promote gut health without affecting pig growth performance. Four crystalline amino acids (FCAA)—L-lysine, DL-methionine, L-threonine, and L-tryptophan—were given to low-protein (LP) diets in these trials because they are the first four restricted amino acids that must be supplied to balance for an optimal protein ratio. The 2012 National Research Council (NRC) eliminated the specific recommendations for crude protein (CP) requirements for swine. It replaced them with a total nitrogen requirement as part of their standard nutritional requirements for swine. This change in the NRC's recommendations reflects the advances in understanding the relationship between nitrogen and protein in swine diets. The CP need is 2% to 4% lower than the suggested amount of the NRC (1998) if the total nitrogen demand of NRC (2012) is multiplied by the standard CP coefficient of 6.25. Because of the development of industrial synthetic AA technology, it is now possible to supplement bovine diets with feed-grade AA, such as L-valine and L-isoleucine, which can further lower dietary CP.

Studies using high-grade amino acids show that raising digestible CP to 13.5% (or around 15.5% CP) increased sow milk protein output, boosting litter growth rate (Strathe et al.,2017). Greater digestible CP levels of 14.3% (or roughly 16.5% CP) appeared to reduce sow BW loss by delaying muscle protein mobilization for milk production (Strathe et.al.,2017). The minimal digestible CP content for lactation meals is between 13.5% and 14.3%.

2.2 Requirement of Amino Acid

As you can see in table 3, amino acids are nutritionally classified into essential or non-essential amino acids. Amino acids are traditionally grouped according to nutritionally essential or non-essential based on the animal's growth or nitrogen balance and their transport affinities. Provision of these nutrients is necessary for all animals. Also, they are recognized by the standard structural features. For example, leucine, isoleucine, and valine are called branched-chain amino acids. However, tyrosine and phenylalanine are categorized as aromatic amino acids. Current research has analyzed the amino acid needs of high-producing lactation sows in order to ensure their optimal performance. The estimations of the required amino acids generally change depending on the study's performance standards and statistical approach. Estimates of lysine requirements are the most researched since models indicate that breastfeeding sows with big, rapidly expanding litters will have much higher lysine requirements. Although the literature is divided on the impact of dietary Lys consumption on litter growth rate and subsequent reproductive success, it has an impact on reducing BW loss and body protein mobilization. (Xue et al; Shi et al.; Gourley et al). The Lys requirement estimate to minimize sow BW loss during lactation is roughly 0.72 to 0.79 g SID Lys per MJ ME, according to studies using a range of 0.50 to 0.81 g SID Lys per MJ ME. Although the estimates for primiparous and multiparous sows appeared to be within the same range, the BW loss has been observed to be significantly higher in primiparous than multiparous sows, at about 12% and 7%, respectively (Xue et al.,2012; Gourley et.al., 2017). Reduced loss of loin eye depth during lactation suggests a low mobilization of muscle protein is to blame for the decreased sow BW loss.

Table 3. Nutritional classification of amino acids (Wu et al., 2014)

Essential		**Conditionally non-essential**	**Non-essential**
Common core	**Additional Species-related requirements**		
Lysine	Arginine	Cyst(e)ine	Glutamate
Histidine	Taurine	Tyrosine	Glutamine
Leucine		Arginine	Glycine
Isoleucine		Proline	Serine
Valine			Alanine
Methionine			Aspartate
Threonine			Asparagine
Tryptophan			
Phenylalanine			

3. Ideal amino acid balance for sow

The ideal protein is one that has an optimal balance between the various essential amino acids and the nitrogen sources available for the synthesis of non-essential amino acids. A sow's need for protein is really a need for amino acids. According to the "barrel principle", a shortage of a single amino acid prevents the utilization of other amino acids in sufficient supply. The requirement of essential amino acids to maximize lactation and reproductive performance of lactating sows has been one of the hot research issues. Since the breast milk protein output of lactating sows is the most important part, because researchers mostly determine the ideal amino acid pattern based on the amino acid ratio of milk protein. For lactating sows, amino acids are required for milk production and mammary tissue growth. When amino acid intake is insufficient, maternal tissues are mobilized to break down proteins and fats to meet the sow's amino acid requirements (Trottier and Johnston, 2001; Kim and Easter, 2003). The ideal protein model simplifies the problem of protein-related parameters during formulation design. As soon as the requirement of y one amino acid is determined, the requirement of other amino acids can be obtained accordingly. In addition, the ideal protein model provides a reference protein that can be used to rate the nutritional value of other proteins. In general, biological j-values are commonly used to evaluate proteins in feed. However, the amino acid ratios of the reference protein itself required by this method differ from those required by the animal and are not additive. The ideal protein is formulated according to the pig's need for lysine and is the relationship between the amino acid requirements in the ideal state. Therefore, the smaller the difference with the feed l ideal protein c, the higher the nutritional value. The ideal protein for diet preparation can improve the utilization of amino acids and reduce the level of elemental nitrogen in the diet without reducing production performance, thus saving protein resources, reducing costs, reducing nitrogen emissions and contributing to environmental protection while maximizing the production potential of livestock and poultry.

3.1 Application of the ideal protein model

Kim et al. (2001) proposed an ideal amino acid model for lactating sows based on the NRC, considering the body weight loss of lactating sows. Kim pointed out that the top three limiting amino acids recommended by the NRC for corn-soybean meal diets for lactating sows are lysine, valine, and threonine, but the degree of body loss is not considered, which is not in line with the reality of production. The ideal model of Kim et al. (2001) has the same amino acid limiting order as the NRC when body losses are the same. The study by Kim et al. (2001) found that lysine is the first limiting amino acid and valine is the second limiting amino acid when the rate of mobilization of body tissues is less than 20%, and when the rate of mobilization of body tissues is greater than 20%, the limiting order of lysine remains unchanged, threonine is the second limiting amino acid, and valine becomes the third limiting amino acid. In addition, when Soltwedel et al. (2006) conducted a validation experiment

using blood urea nitrogen, they found that the results measured when the loss of body tissue was at 8% were similar to the results of the Kim (2001) study.

Table 4. Ideal amino acid pattern and order of limiting amino acids as tissue mobilization decreases during lactation (Kim et.al., 2001)

	Level of tissue mobilization										
Items	**100%**	**90%**	**80%**	**70%**	**60%**	**50%**	**40%**	**30%**	**20%**	**10%**	**0%**
Ideal amino acid pattern relative to lysine											
Lys	100	100	100	100	100	100	100	100	100	100	100
Thr	75	72	69	67	66	64	63	62	61	60	59
Val	78	78	78	78	78	78	78	78	77	77	77
Leu	128	125	123	122	120	119	118	117	116	115	115
Ile	60	60	59	59	59	59	59	59	59	59	59
Phe	57	57	57	57	57	56	56	56	56	56	56
Phe +Tyr	130	126	123	121	118	116	114	113	111	110	109
Arg	22	31	38	45	50	55	59	63	66	69	72
His	34	35	36	37	37	38	38	38	38	38	39
Order of limiting amino acids[a]											
First	Thr	Thr	Lys	Lys	Lys	Lys	Lys	Lys	Lys	Lys	Lys
Second	Lys	Lys	Thr	Thr	Thr	Thr	Thr	Thr	Thr	Val	Val
Third	Val	Val	Val	Val	Val	Val	Val	Val	Val	Thr	Thr
Fourth	Phe+ Tyr	Phe+ Tyr	Ile	Ile	Ile	Ile	Ile	Ile	Ile	Ile	Ile
Fifth	Ile	Ile	Phe+ Tyr	Phe+ Tyr	Phe+ Tyr	Phe+ Tyr	His	His	His	His	His

[a]Sulfur-containing amino acids and tryptophan were not considered in determining limiting order.

4. Dietary Protein Levels for Sows

4. 1 Effects of dietary protein levels on reproductive performance of sows

In lactating sows, protein is essential for milk production and the maintenance of body condition. Lactation is unquestionably the most metabolically taxing stage of production, even though it only makes up 15% to 20% of a sow's reproductive cycle. The sow's primary goal during lactation is to continue producing milk for the vast and rapidly expanding litter of piglets. However, this goal is frequently not met by voluntary meal consumption alone. Body fat and protein reserves must be mobilized in order to promote milk production in high-producing sows, although it is not apparent whether this process is required in contemporary sows (Pedersen et al.,2019). During early lactation, sows have high energy demands for milk production and rely on body fat reserves to meet this energy requirement. However, as lactation progresses, the sow's body condition can decline, and her energy demands increase further.

Adequate dietary protein intake can help sows maintain body condition by promoting muscle growth and repair, which helps to prevent excessive loss of body weight and backfat thickness during lactation. In addition, protein is essential for synthesizing milk proteins, which are necessary for milk production and the growth and development of piglets. Studies have shown that increasing the dietary protein content in lactating sow diets can increase body weight and backfat thickness. However, the effect of protein on backfat thickness depends on the level of dietary protein and the balance between protein and other nutrients in the diet. Excessive protein intake can lead to increased fat deposition in the sow, resulting in increased backfat thickness. Excess protein is converted into energy and stored as fat in the body. Therefore, it is vital to provide the optimal dietary protein level to prevent excessive fat deposition in lactating sows.

In summary, protein is essential for lactating sow nutrition and plays a crucial role in milk production, maintaining body condition, and preventing excessive fat deposition. A balanced diet with optimal protein, energy, and other essential nutrients is crucial for optimizing lactation performance and ensuring the health and welfare of lactating sows and their piglets. Clowes et al. showed that low protein intake increased somatic protein mobilization in sows. That follicle volume was significantly reduced at 9–12% somatic protein mobilization, the maximum follicle number was significantly lower, the ovulation rate was reduced, and the interest interval was prolonged in sows.

Recent research has shown that modern hyper prolific sows can produce a high milk yield and nurse up to 14 piglets per lactation cycle. With increased production has come an increased demand for nutrients. Lactating sows use up to 70% of the crude protein (CP) in their diet to produce milk protein, according to a study conducted by Pedersen et al. (2016). This demonstrates the vital importance of CP in providing lactating sows with the necessary nutrients to produce milk proteins for their offspring adequately. Strathe et al. (2017) studied hyper-prolific sows to explore the impact of dietary standardized ileal digestible (SID) CP on their performance. The results showed that when SID CP was increased from 14% to 15%, the litter's average daily gain (ADG) was improved, and the sows' body weight loss was reduced. Furthermore, when the SID CP was increased from 14% to 13%, the litter's ADG was further improved, and the sows' body weight loss was even lower. These findings suggest that increasing dietary SID CP can benefit the performance of hyper-prolific sows. Guan et al. (2004) reduced the dietary protein water sum from 23.5% to 7.8% and found that milk yield and piglet weight gain decreased linearly with decreasing protein levels, true protein concentration and amino acid levels in milk decreased with decreasing protein levels, and actual protein concentration and lactose concentration in milk decreased with increasing lactation days, with no effect on dry matter and milk fat in milk. It was found that the addition of synthetic amino acids to the diet reduced crude protein levels from 16% to 12%, except for an increase in plasma concentrations of the added essential amino acids, but a significant decrease in the concentrations of other essential amino acids. In studies on lactating sows, it has also been found that diets with high lysine levels lead to valine deficiency, in which case the performance of the sows as well as the piglet, may be affected. Therefore, when lowering crude protein to add synthetic amino acids, the dietary amino acid restriction order must be considered, and potentially restricted amino acids must be added sequentially. Richert et al. (1996) reported a significant increase in sow performance with increasing valine addition at 0.9% lysine level in a corn-soybean meal diet, suggesting valine may be the second limiting amino acid for lactating sows. NRC (1998) ranked valine as the second limiting amino acid; Kim et al. (2001), in their study of the amino acid limiting order in lactating sows on corn-soybean meal diets, found that when tissue mobilization was less than 20%, lysine remained in the limiting order, threonine was the second limiting amino acid, and valine became the third limiting amino acid. Soltwedl et al. (2006) performed a validation experiment, showing that the results measured at about 8% body weight loss were like those of Kim et al. (2001). Lactating sows fed corn-soybean meal-type diets with lysine as the first limiting amino acid, valine as the second limiting amino acid, and threonine as the third limiting amino acid within the normal weight loss range (10%).

4.1.1 Possible mechanisms of the effect of protein level on the productive performance of sows

The development of the mammary gland is dormant before the sow's first heat and starts to develop after the first heat. The mammary gland volume increases while the mammary ducts k begin to differentiate, after which the mammary gland grows naturally. In early gestation, r mammary gland development begins to change, and growth and differentiation of mammary ducts begin to occur, followed by the growth of mammary lobules and mammary vesicles. As the ratio of parenchymal to mesenchymal tissue grows, the lobules and the follicles on the lobules are spaced apart by connective tissue. The development of parenchymal tissue during 2/3 of gestation is generally thought to be only the differentiation of mammary cells, followed by enlarging mammary epithelial cells and enlarging mammary vesicles (18). However, recent studies have confirmed mammary cell differentiation occurs during gestation and lactation. As the follicles develop on the mammary lobules, the mammary epithelial cells differentiate at a morphological and organelle level and become capable of releasing milk. Milk production and secretion are regulated by prolactin (PRL) and growth hormone (STH). Growth hormone plays an equally important role in milk synthesis as a nutrient. It has been shown that PRL and STH are essential for the transition from growth to secretory mammary glands in many species. PRL plays a dominant role in lactation's production and secretion phase, but STH is integral to maintaining lactation. PRL and STH induce functional differentiation of cells in the synthesis of milk proteins and milk lipids.

PRL acts through receptors on mammary epithelial cells and activates the activity of multiple transcription factors. The key active molecule in the receptor signaling pathway system of PRL is the signaling protein and transcriptional activator 5 (Stat5). The role of Stat5 in the mammary gland is to mediate the PRL signaling system. Unlike PRL, functional receptors for STH are uncommon in the mammary gland, and their biological response is indirect through IGF-I. Protein and lysine levels in the diet affect IGF concentrations. The effect of STH on lactation function is mediated by IGF-I, which is mainly produced in the liver. Studies in mice and sheep found that low protein diets reduced IGF-ImRNA expression and circulating concentrations of IGF-I.

In addition, the quality of protein in the diet was found to affect the expression of IGF-ImRNA and the circulating concentration of IGF-I in studies with mice. In the study of piglets, Katsumata et al. found the same crude protein level and a significant decrease in circulating concentrations of IGF-I in the blood of the lysine normal group (0.58%) compared to the low lysine group (0.75%) but no decrease in IGF-ImRNA expression in the liver. The expression of IGFBP3, an IGF-I binding protein, was found to be significantly decreased by protein hybridization, and the binding of IGFBP3 to free IGF-I in the blood increased its half-life from less than 10 minutes to 5 hours. Thus, the effect of lysine on IGF-I concentrations was achieved either by inhibiting post-transcriptional processing of the IGF-I gene or by increasing the clearance of IGF-I from the circulation. The pituitary gland secretes PRL and reaches the target organ, the mammary epithelium. The secretory function of the epithelial cells is activated by binding to its receptor. PRL can be secreted in activated epithelial cells by paracrine secretion, activating the surrounding cells and thus making the mammary cells of the sow functional for milk secretion. The study of lactating sows found that the difference in IGF-I mammary artery concentration was significantly higher in the 23.5% crude protein level group than in the 18.2% crude protein level group during the 18 days of lactation. The mammary arteriolar concentration of essential amino acids significantly affected the difference in PRL mammary arteriolar concentration. The mammary artery concentration of essential amino acids was positively correlated with the difference in mammary artery concentration of PRL. Therefore, protein requirement level can indirectly affect PRL concentration and, thus, milk production. Therefore, protein requirement level can indirectly affect PRL concentration and, thus, milk production. As a result, improving the balance of amino acids in the diet can help sows produce more milk.

4.2 Effects of Protein Levels on Reproductive Performance

Numerous earlier studies have suggested that dietary protein consumption impacts reproductive success. (Mahan,1998; Yang et.al.,2009; Zhang et.al.,2011). The performance of sows in terms of productivity has significantly increased along with the ongoing advancements in modern breeding technologies. Compared to earlier generations, current swine have larger litters, lower fat content, and increased body weight. This indicates the success of selective breeding and improved nutrition and care practices. Although the typical detrimental effects of excessive lactational catabolism on sows' later reproductive success are well documented (Koketsu et al.,1996), contemporary sows appear more resistant to these effects (Patterson et al.,2011). Although sow resilience over consecutive parities has yet to be thoroughly studied, it can be linked to changes in biology and body lean composition—a distinguishing trait of the modern sow. Consequently, the primary objective of the feeding regimen for lactating sows should be to maximize feed intake to maintain milk production without overutilizing BW reserves. During early lactation, sows have high energy demands for milk production and rely on body fat reserves to meet this energy requirement. However, as lactation progresses, the sow's body condition can decline, and her energy demands increase further. Adequate dietary protein intake can help sows maintain body condition by promoting muscle growth and repair, which helps to prevent excessive loss of body weight and backfat thickness during lactation. In addition, protein is essential for synthesizing milk proteins, which is necessary for milk production and the growth and development of piglets. Studies have shown that increasing the dietary protein content in lactating sow diets can increase body weight and backfat thickness. Milk's nutrients come from diet and body reserves because lactating sows frequently go into catabolic states. While BF mobilization increased with increased protein intake, the reduction in BW loss in the current trial was primarily due to decreased body protein mobilization. Dourmad et al. showed a similar response to increased dietary protein (1998). As a result, the sows may preserve body protein by using high levels of dietary SID CP as a substrate for milk protein.

Due to the downregulation of urea cycle enzymes, the sows given high SID CP levels experienced lesser protein mobilization and lower plasma ALT values (Das and Waterlow, 1974). Protein deficient diets cause the body's protein turnover to decrease, resulting in higher consumption of ingested protein. In pigs with a protein deficit, sows fed low SID CP concentrations also showed lower plasma albumin concentrations (Wykes et al., 1996), which suggested that these sows produced less albumin. In this study, the lowest amount of body protein loss (128 SID CP g/kg) happened at a dietary SID CP concentration nearly equal to the highest amount of milk protein (136 SID CP g/kg).

4. 3 Effects of dietary protein levels on Blood Profiles of sows

The level of blood urea nitrogen concentration epitomizes the balance of protein and amino acid metabolism in lactating sows ((Patidar et al., 2018). Numerous studies have shown that when the amino acid and protein transformation reaches a balanced state, the concentration of blood urea nitrogen in sows is reduced to the lowest level, which can well meet the lactation demand of sows.

When the amino acid imbalance in the body, it will cause the concentration of blood urea nitrogen to rise, and at the same time, accelerate the decomposition of protein metabolism in sows. When the amino acid balance is restored, the blood urea nitrogen concentration will be the lowest (Chen et al. 1995, Coma et al., 1996). When lactating sows were fed amino acid balanced diets with crude protein levels of 18%, 17%, 16%, and 14%, blood urea nitrogen concentrations of lactating sows were significantly lower in all groups (Liu et al., 2009).

The level of total protein in the serum reflects the dynamic balance of protein breakdown and synthesis in the body. A high level of total protein indicates a vigorous protein metabolism in the body, which is conducive to improving the efficiency of protein utilization in lactating sows. Glucose can provide energy for a series of biochemical reactions in the body, when the animal body lacks energy or amino acid intake is insufficient, glucose will be decomposed and metabolized in large quantities in the body, and sufficient energy will be produced to maintain the body's metabolic needs. Free fatty acids in serum are important indicators of fat metabolism in the body. The concentration of free fatty acids in the blood indicates the degree of fat oxidation and decomposition. It was found that fat mobilization in the lactation stage of sows, high degree of fat catabolism, and high concentration of free fatty acids in the mother's body, while in the weaning estrus stage of sows, the mother's body fat is gradually shifted from catabolism to anabolism. The concentration of free fatty acids in the body gradually decreases (Yang et al., 2009). By feeding two (17% and 20%) amino acid balanced diets with different crude protein levels, serum urea nitrogen was significantly ($P<0.05$) lower in the low-protein group, and there was no significant difference in total protein content (Liu et al., 2019). Reducing dietary protein levels (104, 114, 121, 129, 139, and 150 g/kg) in lactating sow diets resulted in non-significant differences in serum glucose and free fatty acid content between treatment groups (Strathe et al., 2020).

4. 4 Effects of dietary protein levels on Odor Emission of sow

Nitrogen in livestock manure and urine is a major source of nitrogen emissions. The main substances produced during manure storage and transportation include volatile fatty acids, phenolic compounds, indole compounds and volatile sulfides. (Trabue et al., 2019). In conventional diets, crude protein is often overused, excess nutrients are excreted due to the utilization of feed ingredients and the economic utilization of crystalline amino acids (Hauschild et al., 2012), resulting in large amounts of nitrogen excretion, and a similar effect occurs when essential amino acids are deficient in swine diets. Reducing dietary protein levels and supplementing with crystalline amino acids is an effective way to reduce N emissions without compromising animal growth performance. It has been found that amino acid balanced diets with a crude protein level of 16% can reduce nitrogen and ammonia emissions (Hou et al., 2021), mainly because lower protein levels supplemented with crystalline amino acids can also satisfy the sows' nitrogen requirements, and a reduction in the amount of nitrogen ingested will result in a reduction in the amount of nitrogen excreted. When the crude protein level in the diet was reduced from 17.6% to 14.8%, the ammonia and hydrogen sulfide contents in the feces of pigs were reduced by 32.8% and 11.86%, respectively, and the ammonia and hydrogen sulfide contents in the air of the barn were reduced by 21.56% and 29.4%, respectively, which indicated that the low-protein diet had a contributing effect on reducing nitrogen excretion and improving the air environment. Uricase exists in feces, and nitrogen in pig urine and feces mainly exists in the form of urea, which is decomposed into carbon dioxide ammonia gas under the action of urease, which leads to the volatilization of ammonia gas. When the nitrogen used for the total reaction decreases, the volatilization of nitrogen gas decreases accordingly (Aarnink et al., 2007). On the other hand, as the level of dietary crude protein decreases, the supplementation of amino acids allows for balanced nutrition, nitrogen uptake, and an increase in nitrogen deposition, which reduces nitrogen excretion. When dietary protein was reduced from 14.63% to 12.63%

and 10.63%, total nitrogen excretion was reduced by 16.9% and 31.9%, respectively, when balancing 10 essential amino acids; when protein level was reduced from 14.63% to 10.63%, total nitrogen emission was higher than that of the group with the addition of 10 essential amino acid diets when only lysine, methionine, threonine and tryptophan were added. This suggests that both dietary protein level and amino acid balance pattern affect nitrogen excretion (Zhao et al., 2019). It has also been shown that lowering dietary protein levels (18.6%-16.2%-15.6%) results in lower urinary and fecal pH, with urinary and fecal nitrogen in the form of ammonium ions. converted to ammonia emission into the air when PH is increased (Portejoie et al., 2004). Reducing crude protein levels in the diet by adding amino acids also reduces organic phosphorus excretion, attenuates the acidic environment, and inhibits the conversion of ammonium ions to ammonia (Shriver et al., 2003).

5. Problems with low protein diets

Low protein levels in the diet can also affect animal performance. Although many experiments at home and abroad have proved that the type, proportion and quantity of amino acids in the diet can meet the needs of animals, and the dietary protein level can be reduced by 2-4%, the performance of livestock and poultry will remain unchanged while the feed conversion rate, nitrogen deposition, nitrogen emission and the ability to cope with heat stress will be improved. However, by further reducing the protein level in the feed, no matter how the amino acid requirements are ensured, the growth performance of the animals will not reach the desired level. When a 17% crude protein level was used as a control, a 6% reduction in crude protein level was used as a low protein feed experimental group. The low protein experimental group was supplemented with lysine, tryptophan, isoleucine, methionine and valine to meet the NRC (1998) standard, and the results of the experiment showed that the low protein feed experimental group was not as good as the normal protein group in terms of growth performance and feed conversion ratio. The addition of synthetic amino acids and the reduction of dietary protein levels resulted in growth performance like that of the control group only when protein levels were reduced by no more than 4%. When experiments were conducted with growing pigs weighing 30 kg, the dietary protein level was reduced from 16% to 11% with the addition of synthetic lysine, threonine, tryptophan, valine, methionine, and glutamic acid, although the weight gain levels were similar the feed intake was elevated and therefore the feed conversion ratio decreased instead (Rusesll et al. 1986, 1987) The decrease in the feed conversion ratio suggests that the increased intake may be be used primarily for fat deposition rather than protein deposition. Therefore, carcass quality may be reduced for pigs that do not require high leanness.

Supplementation with lysine, threonine, tryptophan and methionine and a decrease in protein percentage showed negative effects on pig performance and body fat content (Yu et al., 1991). (Yu et al., 1991). Guay et al. (2006) found that decreasing the dietary crude protein level from 16.1% to 7.8% resulted in a significant decrease in daily weight gain and feed conversion ratio of growing pigs, a substantial reduction in crypt depth in the jejunum and ileum, and a considerable decrease in villi width; a significant reduction in jejunal mucosal protein secretion, and a decreasing trend in ileal mucosal protein secretion. Adding too many synthetic amino acids can impact how well the intestinal mucosa absorbs amino acids. This can then influence the morphological integrity of the small intestinal mucosa, affecting the absorption area and enzyme activity, resulting in a decrease in protein concentration at the base and distal ends of the small intestine and a reduction in growth performance and feed conversion rate. In addition, mucosal protein content is related to peptide-bound amino acids and not to free amino acid concentration. A decrease in the amount of intact protein in the diet causes a reduction in intestinal mucosal protein secretion, which reduces the deposition of globulins.

III. Effects of Different Levels of Crude Protein on Physiological Responses, Litter Performance, Blood Profiles, Milk Composition and Odor Emission in Lactating Sows

ABSTRACT: This experiment was conducted to study the effects of dietary crude protein levels on physiological response, reproductive performance, blood profile and milk composition of lactating sows in summer. A total of 48 F1 multiparous sows (Yorkshire × Landrace) with average body weight (BW) of 238.0 ± 4.65 kg, average backfat thickness of 20.95± 1.38mm, and an average parity of 4.65 ± 0.44was used in a 3wk trial at a research farm located in Eum seong, Korea. All sows were allotted to one of six treatments considering BW, backfat thickness, and parity in completely randomized design (CRD) with 8 replicates. All experimental diets for lactating sows were formulated based on corn soybean meal. Treatments are as followed: 1) CP15, 2) CP16, 3) CP17, 4) CP18,5) CP19, 6) CP20. All other nutrients in experimental diets were formulated to meet or exceed the NRC requirements (2012). There were no significant differences in body weight, backfat thickness and average daily feed intake of lactating sows between dietary crude protein levels throughout the trial period. And there was no significant difference in the effect of increasing dietary crude protein level on the number of live births, average litter weight and average piglet weight of sows. Casein content in milk decreased linearly with increasing crude protein level in the diet ($P<0.05$). The increase of protein level in the diet did not have a significant effect on the blood biochemical parameters of piglets. The effect of changing the crude protein levels in the feed on triglyceride, creatinine and glucose concentrations in the blood of sows was no significantly different. However, with the increase of crude protein level, the urea nitrogen and total protein content level in the blood of the lactating sows increased linearly (Linear, $P=0.01$). The concentrations of odor gases, including amine, ammonia and hydrogen sulfide, increased linearly when sows were fed diets with higher levels of the dietary CP (Linear, P=0.01). Increasing the protein level in the diet does not improve the performance of lactating sows and piglets. Collectively, this experiment demonstrated that crude protein level of 15% in sow diet help to reduce the odor gas emissions from manure as well as feed cost of sows without any detrimental effects of reproductive performance of sow.

Keyword: Dietary crude protein, Blood profiles, Litter performance, milk composition, Piglets, Lactating sow

Introduction

Protein in the diet for maintaining healthy growth and normal immune function of livestock and poultry has a very important role, but too high protein on the one hand to increase feed costs, aggravate the burden of feeding, on the other hand, it will also cause an increase in nitrogen emissions in animal feces and urine, causing environmental pollution. Undigested and unutilized crude protein and amino acids in feeds are the main sources of nitrogenous feces. Reducing the level of crude protein in the diet can significantly reduce nitrogen emissions from feces and urine during production and save production costs. Some studies have shown that when the level of crude protein in the diet is reduced by 2-4%, it will not have a significant effect on growth performance, but it can reduce the amount of nitrogen discharged and lower the cost of feed (Yi et al., 2008; Yang et al., 2008). According to the physiological characteristics of lactating sows, dietary protein and amino acid levels directly affect sow milk yield, colostrum and standing milk nutrient content and piglet production performance (Wang et al., 2000). A 1-2% reduction in crude protein levels (CP17, CP16) did not affect sow weight loss during lactation and piglet weaning litter weight compared to diets with conventional 18% crude protein levels (Liu et al., 2010). Decreased productivity of lactating sows during the hot summer season has become a problem for producers. It has been reported that when the temperature was increased from 21.5°C to 29.5°C, the feed intake of sows decreased by 14.1% and milk production decreased by 16.3% (Silva et al., 2008). Increasing the nutrient concentration of the diet can somewhat alleviate the decline in production performance caused by inadequate nutrient intake. However, under the high temperature and hot conditions in summer, too high protein concentration in the diet will increase the heat gain and metabolic burden of sows and aggravate the negative effects of heat stress. A 3.4% reduction in crude protein content at 29°C elevated temperatures significantly increased litter weight gain in lactating piglets and reduced nitrogen excretion in sows (Renaudeau et al., 2001). According to the physiological characteristics of lactating sows, dietary protein and amino acid levels directly affect sows' milk production, colostrum and standing milk nutrient content and piglet performance. With the rapid development of large-scale intensive pig farming, the harm of heat stress to l actuating sows is getting more and more attention. Low protein diets can reduce the use of protein feed, save feed costs and reduce nitrogen emission. In terms of heat stress and protein nutrition, low-protein diets can reduce the loss of energy in the form of heat. Each 1 g reduction in protein reduces heat loss by 7.0 kJ and mitigates heat gain. Lactating sows at a dietary protein level of 18.5% outperformed the experimental group at 19.5% crude protein level under high summer temperatures (Zhang et al., 2010). The effects of low protein diets on the performance, serum indices, and apparent digestibility of nitrogen in lactating sows under continuous high-temperature conditions have yet to be reported.

Therefore, this experiment was conducted to study the effect of the addition of synthetic amino acids at different protein levels on the production performance and nitrogen excretion of lactating sows in summer to provide a basis for the rational use of the ideal amino acid model in pig production. Amino acid pattern to formulate low-protein amino acid balanced diets This experiment was conducted to study the effect of the addition of synthetic amino acids at different protein levels on the production performance and nitrogen excretion of lactating sows in summer.

Materials and Methods

Experimental animals

All experimental procedures involving animals were conducted in accordance with the Animal Experimental Guidelines provided by the Seoul National University Institutional Animal Care and Use Committee. A total of 48 F1 multiparous sows (Yorkshire × Landrace) with average body weight (BW) of 238.0 ± 4.65 kg, average backfat thickness of 20.95± 1.38mm, and an average parity of 4.65 ± 0.44was used in a 3wk trial at a research farm located in Eum seong, Korea. All sows were allotted to one of six treatments considering, BW backfat thickness, and parity in completely randomized design (CRD) with 8 replicates.

Experimental design and diet

All experimental diets for lactating sows were formulated based on corn soybean meal and selenium was supplemented by sources and levels. Treatments are as followed: 1) CP15, 2) CP16, 3) CP17, 4) CP18,5) CP19, 6) CP20. All other nutrients in experimental diets were formulated to meet or exceed the NRC requirements (2012). The formula and chemical composition of experimental diets in lactation was presented in Table 1.

Animal management

A total of 48 pregnant sows were washed and moved into farrowing crates (2.40 × 1.80 m^2) on day 110 of gestation. The gestation diet was decreased gradually 0.2 kg per day during 5 days before farrowing. Delivery inducer was not used during farrowing and all sows were taken an assistance when dystocia was happened. After farrowing, the experimental lactation diet was increased gradually from 1.0 kg/d until 5 days postpartum and then provided ad libitum during the lactation period. Each farrowing crate was equipped with a feeder and a nipple waterer for sows and a heat lamp for newborn piglets. The temperature of lactating barn was kept 28 ± 2℃ and baby house under heating lamp was kept 32 ± 2℃. Air condition of lactating barn was regulated automatically by ventilation system and air-conditioner. After farrowing, piglets were cross-fostered within treatment until 24 hrs postpartum to balance suckling intensity of sows with equalization of litter size, and thus to minimize any effect of initial litter size potentially affecting litter growth. Cutting umbilical cord and tail and castration were conducted 3 days after birth, and piglets were injected with 150 ppm Fe-dextran (Gleptosil®, Alstoe, UK) ２３ injection. All piglets were not fed creep feed during whole lactation period. Weaning was performed at approximately 21 d.

Body weight, backfat thickness, lactation feed intake

Live body weight and backfat thickness of sows were measured at 24 hrs postpartum, 7th day of lactation and 21st day of lactation, respectively. Body weight of sow was measured by electric scale (CAS Co. Ltd., Yangju-si, Gyeonggi-do, Korea) and backfat thickness was measured at P2 position (mean value from both sides of the last rib and 65 mm away from the back bone) by Ultra-sound device (Lean Meter®, Renco Corp., Minneapolis, MN, USA). Daily feed waste was recorded during lactation to identify physiological effects on sows.

Chromium oxide was added to the diet as an indigestible marker at 0.20% of the diet for 7 days prior to fecal collection to calculate nutrient digestibility. Each sample was stored in a freezer at -20℃until analyzed. The fecal samples were thawed and dried at 60℃ for 72h, after which they were finely grounded to pass through a 1 mm screen. The procedures to determine nutrient digestibility were in accordance with the methods established by the AOAC (2002). Feces and urine were kept in sealed containers and were immediately stored at -4℃ for the duration of the period. After the collection period, feces and urine samples were pooled and each mixed well for each replication. Then the slurry was prepared by mixing urine and feces in 1:1 ratio. The samples were permitted to ferment for at 25℃ for 7 days. After the fermentation period a gas sampling pump (Model GV-100; Gastec corp., Ayase, Japan) was utilized for gas detection (Gastec detector tube No.3La for ammonia). Before the measurements, slurry samples were shaken manually for approximately 30s to disrupt any crust formation on the surface of the slurry sample and to homogenize them.

Litter performance

The number and body weight of piglets was measured at 24 hrs postpartum, 7th day of lactation and 21st day of lactation for calculating litter weight, piglet weight and both weight gain by electric scale (CAS CO. Ltd., Yangju-si, Gyeonggi-do, Korea). At measuring the body weight of piglets, ear notching was practiced for experiment. ADG of piglets was calculated to identify their growth performance and lactating performance after farrowing.

Blood profiles

Blood samples (n=4 for each treatment) were collected from jugular vein of sows using 10 ml disposable syringes at 24 hrs postpartum, 7 th day of lactation and 21 st day of lactation, respectively. Also, blood samples were collected from anterior vena cava of piglets using 3 mL disposable syringes at 24 hrs postpartum and 5 mL disposable syringes at 7 th day of lactation and 21 st day of lactation. All serum from blood samples were moved in serum tube (SST TMII Advance, BD Vacutainer, Becton Dickinson, Plymouth, UK) and EDTA tube (BD Vacutainer K2E, Becton Dickinson, Plymouth, UK). Individual sample was centrifuged at 3,000 rpm, 4℃ for 15 minutes (Eppendorf centrifuge 5810R, Hamburg, Germany) and the supernatant serum was separated to a microtube (Axygen, UnionCity, CA, USA) and stored at –20℃ deep freezer until analysis. The concentration of selenium in blood was measured by fluorometric method of AOAC (2000).

Milk composition

Colostrum samples were collected from functional mammary glands at 24 hrs postpartum (n=4) and milk samples (n=4 for each treatment) were taken at 7 th day of lactation and 21 st day of lactation. One mL of oxytocin (Komi oxytocin inj., Komipharm International Co., Ltd., Siheung-si, Gyeonggi-do, Korea) was injected into the blood vessels of the sow's ear to collect colostrum and milk in a 50mL 2 5 conical tubes (SPL Life Sciences Co., Ltd., Pocheon-si, Gyeonggi-do, Korea) from the first and second teats. Collected samples were stored in a freezer (−20 °C) until further analysis. Proximate analysis for fat, protein, lactose, and solids not fat of milk as well as colostrum was determined using a Milkoscan FT 120 (FOSS, Hillerod, Denmark). Also, the concentration of selenium in milk was measured by fluorometric method of AOAC (2000).

Statistical analysis

All collected data were analyzed as a completely randomized design using the General Linear Model (GLM) procedure in SAS (SAS Institute, 2004). Individual sows and their litters were used as the experimental unit in physiological response, litter performance, blood profiles, milk composition and tissue concentration. The differences among means were declared significant at $P<0.05$ and highly significant at $P\geq0.05$ and $P<0.10$. When the significance was declared, fisher's least significance difference (LSD) method was used to separate the means.

Results and Discussion

Physiological response

The effects of crude protein levels on body weight, backfat thickness and average feed daily gain of lactating sows were shown in Table 2. Sows ' body weight, backfat thickness and lactation feed intake were not affected by crude protein levels during lactation.

Growth performance is an essential measure of the nutritional needs of lactating sows. Maintaining good growth performance can guarantee the sow's lactation demand and better recover the body function after weaning due to the decline of farrowing and lactation, which plays a vital role in maintaining the good reproductive performance of the sow. Numerous studies have shown that reducing dietary crude protein levels by 2-4% by adding synthetic amino acids does not affect the growth performance of growing pigs. And reduces the excretion of nitrogen in the feces and urine. Sows need to secrete a lot of milk to feed piglets after giving birth; thus, the nutritional needs of sows during lactation are very high. If the amount of food taken during lactation is insufficient to meet the lactation needs, the lactating sow will disintegrate the body tissues to provide nutrient needs for lactation, thus leading to weight loss of lactating sow. Weight loss and backfat loss during lactation are average for lactating sows, but excessive weight loss and backfat loss can affect the body condition of sows, leading to prolonged intervals between weaning and estrus, reduced ovulation, decreased fecundity and many other problems. According to Yu et al., there was no significant difference in ADFI between lactating sows with dietary protein levels at 14.22% and 18.20%, which is similar to the results of this experiment. The effect of dietary protein level on sow backfat loss and weight loss was found to be insignificant in this experiment, in agreement with that reported by Johnston (1999). In this experiment, sows had the highest average daily feed intake when fed a diet with a crude protein concentration of 15%. However, the effect of crude protein level on backfat loss was insignificant. This is similar to the results of previous studies. The increase in average daily feed intake at CP of 15% compared to the other groups could be because the metabolism of the organism's protein generates a higher heat gain consumption, thus heat stress. In the hot summer environment, a diet with a lower crude protein level can alleviate the heat stress of sows due to feeding to a certain extent and thus increase the feed intake of sows.

Litter performance

The effects of crude protein levels on the number of piglets, litter weight, litter weight gain, average BW of piglet, and average BW gain of piglet were shown in Table 3. The number of piglets, litter weight, litter weight gain, average BW of piglet, and average BW gain of piglet were not affected by crude protein levels during lactation.

Lactating piglets are a vital link in the profitability of large-scale pig farms because they are the source of commercial pigs on the market. One of the frequently researched topics is how to enhance the growth performance of breastfeeding piglets. One study found that the weaning litter weight and average daily gain of lactating piglets increased when lactating sows were fed a low-protein diet. The piglet weaning litter weight and survival rate at 21 days of age also increased to varying degrees, but the differences were insignificant (Dong et.al., 2014). Jang et al. (2006) investigated the impact of diets containing 13% and 15% CP on the productive performance of lactating sows. The results showed that the weaning litter weight was 0.24% higher and the average individual weaning weight of piglets was 0.02% higher in the CP 13% diet group than in the CP 15% diet. This experiment achieved the highest individual piglet weaning weight when the crude protein level was 15% compared to the other experimental groups. And as the protein level increased, the weaning weight of piglets decreased.

Blood profiles

The effects of crude protein levels on serum concentration in sows and piglets were shown in Table 4. The effect of changing the crude protein level in the feed on triglyceride, creatinine and glucose concentrations in the blood of sows was not significantly different (P>0.05). However, with the increase of crude protein level, the urea nitrogen level and total protein content in the blood of lactating sows increased linearly (linear, P=0.01).

Changes in dietary CP levels in pig diets increase blood profiles, such as total protein and serum urea nitrogen (Regmi N et.al.,2018). The blood urea nitrogen concentration level can reflect the balance of protein and amino acid metabolism in lactating sows in an apparently accurate way (Matera, G, et.al.,2009). The higher concentration of blood urea nitrogen indicates the higher degree of protein degradation in lactating sows. When the mutual transformation of amino acids and proteins in lactating sows reaches the equilibrium state, the concentration of blood urea nitrogen in sows is reduced to the lowest, and the utilization efficiency of proteins in diets is the highest, which can meet the lactation demand of sows well. In a previous study, the effect of different protein levels (CP=10.4%, 11.4%, 12.1%, 12.9%, 13.9%, and 15.0%) in lactation diets on blood urea nitrogen concentrations in lactating sows was examined. The results showed that blood urea nitrogen concentrations increased linearly with increasing dietary protein levels in sows when the dietary protein level was higher than 13.5% (Strathe, A, V. et.al, 2020). Based on a diet with a crude protein level of 18.59%, blood urea nitrogen concentration was reduced by 10.43% when the crude protein level was reduced by 1.63%, and by 26.87% when the crude protein level was reduced by 3.06% (Dong et al., 2009). When lactating sows were fed diets with 18%, 17%, 16%, and

14% crude protein levels, blood urea nitrogen levels in the sows' blood decreased with decreasing protein levels (Liu et al., 2008). In this experiment, the increase in blood urea nitrogen concentration with the increase in protein level may be due to the fact that when the protein content in the feed is too high, the excess portion of protein in the sow is metabolized to amino acids after oxidative metabolism, deamidation, and urea nitrogen produced through the ornithine cycle is retained in the sow, which raises the blood urea nitrogen concentration and decreases the amino acid utilization rate.

Milk composition

The effect of crude protein level in the diet on the milk composition of lactating sows is shown in Table 5. Dietary protein level had no significant effect on fat content, total solids and non-fat solids content in the milk of lactating sows. However, as the crude protein level of the diet increased, casein content of the milk to decreased linearly (Linear, P=0.03).

Xiong et al. found that feeding diets with 17.52% and 14.47% crude protein levels did not significantly (P>0.05) affect sows' colostrum and standing milk fat, milk protein, lactose, and non-fat solids. The mammary gland of the sow can absorb free amino acids from the blood to synthesize milk protein. Bequette et al. showed that amino acid production in milk is closely related to protein catabolism and synthesis. High protein feeds contain more amino acids, metabolized to produce urea and other substances, increasing the sow's nitrogen load. The synthesis of lactose requires consuming large amounts of glucose and nitrogen sources. If the sow's nitrogen load is too high, this can lead to a deficiency of glucose and nitrogen sources, inhibiting lactose synthesis and reducing milk lactose content. In this experiment, the linear decrease of casein concentration with the increase of crude protein level may also be due to the fact that when the crude protein level increases, the heat stress response of sows increases, and the amino acids produced by protein catabolism are preferentially used for gluconeogenesis to produce glucose for the body's energy supply, resulting in the decrease of casein concentration due to the shortage of precursors for synthesized proteins. In this experiment, the relatively low level of crude protein helped improve the quality of milk in lactating sows, which positively affected piglet growth.

Odor emissions

The effects of dietary crude protein levels on odor gas emission in sows are shown in Table 7. The concentrations of odor gases, including amine, ammonia, and hydrogen sulfide, increased linearly when sows were fed diets with increasing levels of dietary CP. Moreover, as dietary CP levels increased, quadratic responses were observed in concentrations of odor gas ($P<0.05$).

Many studies have reported that reducing dietary CP intake reduces odor gas emissions in swine diets, primarily focusing on weaning and growing-finishing diets. However, the current study was conducted to estimate the effect of dietary CP level on odor gas emission in the gestating sow. Clark et al. stated that reducing dietary protein levels from 16.8%-13.6% was associated with significantly reduced manure pH, nitrogen and sulfur concentrations in growing-finishing pigs. Similarly, the results of the present study showed that odor gas concentrations, including amines, NH3 and H2S, were linearly affected as dietary CP levels increased. Recently, Kerr et al reported that excessive CP was often fed with high excretion of nutrients in pig manure and urine, evaluated to be 40 to 60% for dietary N and 50-75% for dietary S. In the current study, increasing dietary CP levels from 11% to 16% did not exert any significant beneficial effects on the performance of sows or their progenies. Therefore, reducing the amount of CP in the gestating diet was effective in reducing emissions of odor gas.

Conclusion

There were no significant differences in body weight, backfat thickness and average daily feed intake of lactating sows between dietary crude protein levels throughout the trial period. There was no significant difference in the effect of increasing dietary crude protein level on the number of live births, average litter weight and average piglet weight of sows. In addition, the protein content, casein content and lactose content of lactating sows' milk tended to decrease as the crude protein level in their diets increased. The increase of protein level in the diet did not have a significant effect on the blood biochemical parameters of piglets. For lactating sows, dietary protein levels did not have a significant effect on creatinine, glucose triglyceride levels. However, as protein levels increased, blood urea nitrogen levels in the blood of lactating sows increased, and total blood protein levels decreased. Increasing the protein level in the diet does not improve the performance of lactating sows and piglets. And increasing the protein level increases the feed cost but does not have a positive impact on production.

In the present experiment, the quality of lactating sows' milk was improved when the dietary protein level was 15%. A crude protein concentration of 15% helped to improve the utilization of protein in the feed by the lactating sows, reduced backfat loss and had a positive effect on the sows' entry into the next estrous cycle.

Table 1. The Formulas and chemical composition of the experimental gestation diet

Ingredient(%)	Treatment					
	CP15	CP16	CP17	CP18	CP19	CP20
Corn	69.85	66.91	63.98	61.04	58.11	55.17
SBM	18.69	21.77	24.86	27.95	31.03	34.12
Wheat bran	5.00	5.00	5.00	5.00	5.00	5.00
Tallow	2.11	2.22	2.34	2.46	2.57	2.69
L-Lysine Sulfate, 55%	0.70	0.56	0.42	0.28	0.14	0.00
L-Methionine ,90%	0.08	0.06	0.05	0.03	0.02	0.00
Threonine, 98.5%	0.24	0.19	0.14	0.10	0.05	0.00
L-Tryptophan, 99%	0.08	0.06	0.05	0.03	0.02	0.00
DCP	1.90	1.84	1.77	1.71	1.64	1.58
Limestone	0.66	0.68	0.69	0.71	0.72	0.74
Vit Mix	0.10	0.10	0.10	0.10	0.10	0.10
Min Mix	0.10	0.10	0.10	0.10	0.10	0.10
Choline chloride-50	0.10	0.10	0.10	0.10	0.10	0.10
Salt	0.40	0.40	0.40	0.40	0.40	0.40
Sum	100.00	100.00	100.00	100.00	100.00	100.00
Chemical composition						
ME, kcal/kg	3300.00	3300.00	3300.00	3300.00	3300.00	3300.00
CP, %	15.00	16.00	17.00	18.00	19.00	20.00
Lysine, %	1.09	1.09	1.09	1.09	1.09	1.09
Methionine, %	0.31	0.31	0.31	0.31	0.31	0.31
Ca, %	0.76	0.76	0.76	0.76	0.76	0.76
Total P, %	0.65	0.65	0.65	0.65	0.65	0.65

Table 2. Effects of dietary crude protein levels in diet on body weight and backfat thickness of sows during lactation

Criteria	Treatment						SEM[1]	p-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Lin.	Quad.
Body weight, kg									
24 hr postpartum	241.24	231.15	246.06	236.74	244.33	237.78	3.704	0.87	0.89
21st day of lactation	237.44	230.81	235.43	226.93	242.45	232.83	3.463	0.96	0.72
Change(21-0d)	-3.80	-0.34	-10.63	-9.81	-1.88	-4.95	1.714	0.79	0.30
Backfat thickness, mm									
24 hr postpartum	21.13	18.25	20.07	22.71	22.25	21.25	0.946	0.45	0.99
21st day of lactation	20.69	17.13	17.43	19.43	20.44	20.38	0.809	0.54	0.28
Change(21-0d)	-0.44	-1.13	-2.64	-3.28	-1.81	-0.88	0.329	0.45	0.12
ADFI, kg	5.30	4.92	4.75	5.09	5.13	4.96	0.099	0.71	0.41

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

Table 3. Effects of dietary crude protein levels in diet on reproductive performance of sows and litter performance during lactation

Criteria	Treatment						SEM[1]	*p*-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Lin.	Quad.
No. of piglets									
Total born	12.13	12.50	11.29	12.86	12.50	12.00	0.361	0.90	0.91
born alive	11.13	11.00	10.14	11.43	11.75	11.25	0.325	0.53	0.70
After crossing-fostering	11.50	11.50	11.00	11.29	12.25	11.38	0.293	0.75	0.82
21st day of lactation	9.25	9.63	10.00	9.14	10.38	10.50	0.308	0.22	0.75
Litter weight, kg									
Total born	17.42	16.55	16.03	15.44	17.32	15.99	0.548	0.63	0.55
litter birth weight	16.29	14.90	14.68	14.12	16.48	15.47	0.540	0.99	0.31
After crossing-fostering	16.80	15.39	15.84	14.11	17.04	15.63	0.534	0.80	0.40
21st day of lactation	52.82	45.40	52.07	51.77	53.40	55.88	1.764	0.27	0.45
Litter weight gain	34.77	30.00	36.23	37.66	36.36	40.25	1.678	0.16	0.72
Piglets weight, kg									
Piglet birth weight	1.47	1.34	1.44	1.25	1.40	1.41	0.033	0.64	0.22
After crossing-fostering	1.46	1.34	1.42	1.26	1.38	1.41	0.033	0.67	0.22
21st day of lactation	5.72	4.74	5.13	5.69	5.17	5.39	0.11	0.91	0.29
Piglets weight gain	4.26	3.40	3.71	4.43	3.79	3.98	0.106	0.79	0.48

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

Table 4. Effects of dietary crude protein levels in diet on serum concentration of sows during lactation

Criteria	Treatment						SEM[1]	*p*-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Linear	Quadratic
Sow									
Total Cholesterol									
24 hr postpartum	---------------------------------44.00--------------------------------								
21st day of lactation	76.75	78.75	74.25	75.00	78.25	79.75	0.687	0.28	0.06
Creatinine									
24 hr postpartum	---------------------------------1.99--------------------------------								
21st day of lactation	2.40	2.29	2.28	2.47	2.39	2.21	0.052	0.68	0.59
Glucose									
24 hr postpartum	---------------------------------58.00--------------------------------								
21st day of lactation	65.75	71.00	56.75	62.67	59.75	65.75	2.126	0.53	0.32
Total protein									
24 hr postpartum	---------------------------------6.72--------------------------------								
21st day of lactation	7.35	7.65	7.63	8.06	8.13	8.40	0.099	0.01	0.93
Triglyceride									
24 hr postpartum	---------------------------------27.83--------------------------------								
21st day of lactation	44.5	34.33	29.75	26	29.75	32.75	2.590	0.17	0.10
BUN									
24 hr postpartum	---------------------------------10.80--------------------------------								
21st day of lactation	13.58	15.07	15.45	18.68	19.30	22.93	0.929	0.01	0.49

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

Table 5. Effects of dietary crude protein levels in diet on serum concentration of piglets during lactation

Criteria	Treatment						SEM[1]	*p*-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Lin	Quad
Piglet									
Total Cholesterol									
24 hr postpartum	--------------------------------64.00--------------------------------								
21st day of lactation	170.75	211.75	172.50	146.00	175.75	194.75	8.924	0.94	0.41
Creatinine									
24 hr postpartum	--------------------------------0.46--------------------------------								
21st day of lactation	0.61	0.64	0.89	0.46	0.89	0.87	0.062	0.21	0.73
Glucose									
24 hr postpartum	--------------------------------93.67--------------------------------								
21st day of lactation	104.25	102.75	106.25	112.67	108.75	106.75	3.140	0.61	0.68
Total protein									
24 hr postpartum	--------------------------------6.83--------------------------------								
21st day of lactation	4.48	4.48	4.60	4.62	4.68	5.00	0.080	0.06	0.47
Triglyceride									
24 hr postpartum	--------------------------------118.83--------------------------------								
21st day of lactation	76	74.75	69.00	98.00	112.25	82.75	7.920	0.31	0.74
BUN									
24 hr postpartum	--------------------------------13.28--------------------------------								
21st day of lactation	5.80	6.30	6.33	6.60	7.97	8.20	0.483	0.11	0.73

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

Table 6. Effects of dietary crude protein levels in diet on milk composition of sows during lactation

Criteria	Treatment						SEM[1]	*p*-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Linear	Quadratic
Fat, %									
Colostrum	--------------------------------9.73------------------------------						-	-	-
21st day of lactation	6.57	5.67	6.31	5.81	6.28	6.00	0.203	0.74	0.63
Protein, %									
Colostrum	---------------------------------6.39--------------------------------						-	-	-
21st day of lactation	5.41	5.19	5.04	4.93	4.91	4.35	0.099	0.16	0.06
Lactose, %									
Colostrum	---------------------------------4.71--------------------------------						-	-	-
21st day of lactation	5.65	5.81	5.76	5.75	5.67	5.85	0.034	0.72	0.99
Total solid, %									
Colostrum	---------------------------------21.98--------------------------------						-	-	-
21st day of lactation	17.95	16.83	17.63	16.38	17.66	17.26	0.272	0.70	0.39
Solid not fat, %									
Colostrum	---------------------------------12.34--------------------------------						-	-	-
21st day of lactation	11.65	11.62	11.57	11.06	11.55	11.48	0.087	0.38	0.33
Casein, %									
Colostrum	---------------------------------3.91--------------------------------						-	-	-
21st day of lactation	3.38	3.10	3.06	2.66	3.12	2.96	0.065	0.03	0.07

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

Table 7. Effects of dietary crude protein levels diet on Odor emissions of sows during lactation

Criteria	Treatment						SEM[1]	*p*-value[2]	
	CP15	CP16	CP17	CP18	CP19	CP20		Lin.	Quad.
Amines	28.00	37.00	46.00	47.00	54.00	75.00	3.729	0.01	0.06
Ammonia	3.30	7.50	12.50	13.40	16.50	25.00	1.672	0.01	0.02
Mercaptans	5.63	7.29	10.50	13.50	17.50	21.00	1.331	0.01	0.04
Hydrogen Sulfide	11.50	22.00	27.14	46.00	57.14	60.00	4.418	0.01	0.02

1. Standard error of the mean.
2. Abbreviation: Lin. (linear) and Quad (quadratic).

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Ⅴ. Summary in Korean

본 연구는 포유돈 사료 내 조단백질 수준이 모돈 및 자돈의 생리적인 변화, 포유능력, 혈액성상, 돈유성분 및 악취 발생에 미치는 영향을 조사하여 포유돈 사료 내 조단백질의 수준을 규명하기 위해 수행되었다. 본 실험은 평균 체중(BW)이 238.0±4.65kg, 평균 등지방 두께가 20.95±1.38mm, 평균 산차가 4.65±0.44인 인 2 원교잡종 (Yorkshire × Landrace) F1 모돈 48 두를 공시하여 6처리 8반복, 반복 당 1 두씩, 체중과 등지방 두께에 따라 완전임의배치법 (CRD; Completely randomized design)으로 구배치하여 실험을 수행하였다. 실험의 처리구는 포유돈 사료 내 조단백질 첨가수준에 따라: 1) CP15, 2) CP16, 3) CP17, 4) CP18,5) CP19, 6) CP20으로 나뉘었다. 실험결과, 사료 내 조단백질 첨가 수준에 따라 분만 24시간이내 및 포유 21일령의 체중 및 등지방 두께에서 처리구 간 유의적인 차이가 나타나지 않았다 (P>0.05). 또한, 총 산자 수, 사산두수, 생존자돈 수, 포유개시 두수, 이유두수, 복당 생시체중 및 복당 이유체중에서 처리구 간 유의적인 차이가 나타나지 않았다 (P>0.05). 돈유 중 카제인 함량은 사료 내 조단백질 수준이 증가함에 따라 선형적으로 감소하였다 (linear, P=0.03). 포유 자돈의 혈액 성상에서 유의적인 차이가 나타나지 않았으며 (P>0.05), 포유 21일령 모돈 혈액 중 총 단백질과 BUN 수치에서 조단백질 수준이 증가할수록 선형적으로 증가하였다 (linear, P=0.01; linear, P=0.01). 조단백질 첨가수준이 증가함에 따라 아민, 암모니아 및 황화수소를 포함한 악취 발생이 선형적으로 증가하였다 (linear, P=0.01).

결론적으로, 사료 내 조단백질 수준이 20%에서 15%까지 감소하여도 모돈의 체형변화, 번식성적 및 포유자돈 성장에 부정적인 영향을 미치지 않으며, 체내 단백질 이용율을 개선하여 악취 발생을 줄일 수 있는 것으로 사료된다.