

Effects of Dietary Proline Supplementation on Body Composition of Pregnant Huanjiang Mini-Pigs (Postprint)

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Abstract

This study aimed to investigate the effects of dietary proline supplementation on the body composition of pregnant Huanjiang mini-pigs. Thirty-two Huanjiang mini-pigs at 15 days of gestation were selected and randomly divided into 2 groups, with 8 replicates per group and 2 pigs per replicate. They were fed diets supplemented with 0.77% L-alanine (isonitrogenous control group) and 1.00% L-proline (proline group) in the basal diet, respectively. On days 45 and 70 of gestation, one sow from each replicate was selected for slaughter to determine body composition. The results showed that compared with the isonitrogenous control group, the proline group exhibited significantly decreased fat percentage ($P < 0.05$) and a trend toward increased skin percentage ($0.05 \leq P < 0.10$) on day 45 of gestation; on day 70 of gestation, the proline group showed significantly increased fat percentage ($P < 0.05$), a trend toward decreased skin percentage ($0.05 \leq P < 0.10$), significantly increased proline content in the longissimus dorsi muscle ($P < 0.05$), and a trend toward increased arginine and alanine contents ($0.05 \leq P < 0.10$). Compared with day 45 of gestation, the proline group on day 70 exhibited significantly increased fat percentage ($P < 0.05$) and significantly decreased lean meat percentage and skin percentage ($P < 0.05$). These results indicate that dietary proline supplementation can increase the deposition of body fat and arginine in pregnant Huanjiang mini-pigs, with greater body fat deposition in mid-pregnancy than in early pregnancy, thereby benefiting fetal growth and development.

Full Text

Effects of Dietary Proline on Body Composition in Pregnant Huanjiang Mini-pigs

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Abstract: This study investigated the effects of dietary proline on body composition in pregnant Huanjiang mini-pigs. Thirty-two pregnant Huanjiang mini-pigs on day 15 of gestation were randomly assigned to two groups, each comprising eight replicates with two pigs per replicate. The experimental groups received basal diets supplemented with either 0.77% L-alanine (isonitrogenous control group) or 1.00% L-proline (proline group). On days 45 and 70 of gestation, one sow from each replicate was slaughtered to determine body composition. The results demonstrated that compared with the isonitrogenous control group, the proline group exhibited significantly reduced fat percentage ($P < 0.05$) and increased skin percentage ($0.05 \leq P < 0.10$) on day 45 of gestation. On day 70 of gestation, the proline group showed significantly increased fat percentage ($P < 0.05$), decreased skin percentage ($0.05 \leq P < 0.10$), elevated proline content in the longissimus dorsi muscle ($P < 0.05$), and increased arginine and alanine contents ($0.05 \leq P < 0.10$). Furthermore, when comparing day 70 to day 45 of gestation, the proline group displayed significantly higher fat percentage ($P < 0.05$) and lower lean meat and skin percentages ($P < 0.05$). These findings indicate that dietary proline supplementation can enhance the deposition of body fat and arginine in pregnant Huanjiang mini-pigs, with greater fat deposition occurring during mid-gestation than early gestation, thereby promoting fetal growth and development.

Keywords: proline; pregnancy; Huanjiang mini-pigs; body composition; amino acid composition

Introduction

Sows represent the core resource of pig production enterprises, and their reproductive performance has received considerable attention. Nutritional requirements differ across gestational stages, and both maternal undernutrition and overnutrition can reduce uterine blood supply to the fetus, diminishing nutrient delivery and impeding fetal growth and development [1]. Research has shown that high nutritional levels before mating can increase ovulation rate and oocyte quality in primiparous sows, but continued high nutritional levels after mating reduce embryo survival rate [2]. Thus, regulating maternal nutritional status through dietary manipulation is crucial for improving embryo and fetal survival.

Our previous research revealed that compared with an isonitrogenous alanine control, dietary proline supplementation improved nitrogen and lipid metabolism in pregnant Huanjiang mini-pigs and increased average litter weight

[from (102.5 ± 9.2) g to (109.8 ± 7.8) g on day 45, and from (726.2 ± 54.5) g to (961.7 ± 74.7) g on day 70] and average individual fetuses by day 70 [3]. Additionally, proline oxidase (POX), ornithine aminotransferase (OAT), and ornithine decarboxylase (ODC) are present in porcine placenta, enabling proline to synthesize polyamines that improve maternal nutrition and promote fetal development [4].

Due to hormonal regulation, sow nutritional requirements and body composition change substantially with advancing gestation. As fetuses grow rapidly during late gestation, their nutrient extraction from the mother increases correspondingly. While dietary proline promotes fetal growth, its effects on sow body composition remain unclear. Building on our previous findings, this study utilized Huanjiang mini-pigs to investigate the impact of dietary L-proline supplementation on body composition during gestation, thereby establishing a foundation for its application in sow diets.

Materials and Methods

1.1 Experimental Animals, Grouping, and Management The animal trial was conducted at the Huanjiang Mini-Pig Experimental Base of the Chinese Academy of Sciences in Huanjiang County, Guangxi. Thirty-two healthy primiparous Huanjiang mini-pigs weighing approximately 25 kg were naturally mated and randomly allocated on day 15 post-mating into two groups, each consisting of eight replicates (pens) with two pigs per replicate. The pigs were fed experimental diets supplemented with either 0.77% L-alanine (isonitrogenous control group) or 1.00% L-proline at 09:00 and 18:00 daily. The basal diet was formulated according to NRC (1998) [5] swine feeding standards combined with commercial Huanjiang mini-pig diet formulations; its composition and nutrient levels are presented in . Throughout the trial, pigs had ad libitum access to feed and water, and routine management and immunization procedures followed standard husbandry practices.

1.2 Sample Collection On days 45 and 70 of gestation, one sow per pen was selected, exsanguinated, and slaughtered according to the National Standard “Operating Procedures of Pig Slaughter” [6]. Carcass traits were measured on-site using the right half-carcass. The longissimus dorsi muscle was sampled between the 6th and 7th ribs, and a 200 g sample from the right half-carcass was collected in a sealed bag and stored at -20°C for subsequent chemical analysis.

1.3.1 Carcass Traits Slaughter percentage, backfat thickness, lean meat percentage, fat percentage, and skin percentage were determined according to *Swine Production Science* [7].

1.3.2 Longissimus Dorsi Muscle Composition Frozen, lyophilized, and pulverized longissimus dorsi muscle samples were analyzed for crude protein content using GB/T 5009.6-2003 [8], crude fat content using GB/T 5009.5-2003

[9], hydrolyzed amino acid content according to Liu et al. [10], and medium- and long-chain fatty acid content according to Liu et al. [11].

1.4 Data Processing and Analysis Data from the same dietary group across different gestational periods were analyzed using independent samples t-tests, while data between dietary groups at the same gestational stage were analyzed using one-way ANOVA. Duncan's multiple range test was employed for post-hoc comparisons, with results expressed as least squares means. Statistical significance was declared at $P < 0.05$, and trends were identified at $0.05 \leq P < 0.10$.

Results

2.1 Effects of Dietary Proline on Carcass Traits in Pregnant Huanjiang Mini-pigs As shown in , compared with the isonitrogenous control group, the proline group exhibited significantly reduced fat percentage ($P < 0.05$) and increased skin percentage ($0.05 \leq P < 0.10$) on day 45 of gestation. On day 70, the proline group showed significantly increased fat percentage ($P < 0.05$) and decreased skin percentage ($0.05 \leq P < 0.10$). When comparing day 70 to day 45 within the proline group, fat percentage was significantly higher ($P < 0.05$), while lean meat and skin percentages were significantly lower ($P < 0.05$).

2.2 Effects of Dietary Proline on Conventional Nutrient Contents in Longissimus Dorsi Muscle of Pregnant Huanjiang Mini-pigs As presented in , no significant differences were observed in dry matter, crude protein, or intramuscular fat contents in the longissimus dorsi muscle between dietary groups or gestational ages ($P > 0.05$).

2.3 Effects of Dietary Proline on Amino Acid and Fatty Acid Contents in Longissimus Dorsi Muscle of Pregnant Huanjiang Mini-pigs As shown in and , no significant differences in fatty acid contents were detected between dietary groups or gestational ages ($P > 0.05$). However, compared with the isonitrogenous control group, the proline group exhibited significantly increased proline content ($P < 0.05$) and elevated arginine and alanine contents ($0.05 \leq P < 0.10$) in the longissimus dorsi muscle on day 70 of gestation.

Discussion

During late gestation, sows must meet both their own nutritional requirements and those of the developing fetuses, making body condition critical for reproductive performance. Analysis of sow body composition provides specific insights into how dietary nutrients satisfy maternal and fetal demands. In practice, backfat thickness is commonly measured to assess sow condition, as it positively correlates with fat deposition [12]. In the animal intestine, microbial fermentation of carbohydrates and amino acids produces short-chain fatty acids that can be absorbed and utilized for lipid synthesis. Muscle and adipose tissues serve

as nutrient storage depots, accumulating nutrients when supply is adequate or excessive (e.g., postprandial) [13].

The current findings revealed that dietary proline supplementation significantly increased carcass fat percentage in sows on day 70 of gestation compared with the isonitrogenous control group, indicating that proline promotes fat deposition in pregnant Huanjiang mini-pigs and provides adequate energy for subsequent fetal growth. By day 70 of gestation, placental development ceases while fetal growth accelerates, and reduced abdominal volume limits nutrient absorption, prompting sows to mobilize body fat reserves to support fetal development [14]. The significant increase in fat percentage and decreases in lean meat and skin percentages in the proline group from day 45 to day 70 demonstrate that proline facilitates maternal fat storage during mid-gestation.

The liver stores absorbed amino acids and gradually releases them into circulation for peripheral tissue protein synthesis. Thus, the quantity, type, and ratio of amino acids reflect their utilization and deposition. Proline is a crucial component of collagen [15] and metabolizes to produce ornithine, glutamate, citrulline, arginine, and polyamines, playing important biological roles in nutritional metabolism [16]. Plasma citrulline and mitochondrial ornithine serve as precursors for arginine synthesis [17]. Arginine participates in metabolism through multiple enzymes and enhances placental nitric oxide (NO) synthesis, a key regulator of angiogenesis, embryogenesis, and placental and fetal growth [1,15,18]. This study found that dietary proline increased proline and arginine contents in the longissimus dorsi muscle of sows on day 70 of gestation. The uterus avidly absorbs and synthesizes arginine to meet embryonic developmental demands [1]. Since proline metabolism generates polyamines, it reduces maternal arginine utilization for polyamine synthesis, thereby increasing arginine deposition.

In conclusion, dietary supplementation with 1% proline increases fat and arginine deposition in pregnant Huanjiang mini-pigs, enhancing maternal nutrient reserves to support fetal growth and development.

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