Effect of Glutamine on Intestinal Development in Weaned Meat Rabbits (Postprint)

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Abstract

This experiment was conducted to investigate the effects of glutamine on intestinal development in meat rabbits. A total of 360 28-day-old weaned New Zealand white rabbits were randomly assigned to 4 groups with 9 replicates per group and 10 rabbits per replicate. The four groups were fed experimental diets supplemented with 0 (control), 0.4%, 0.8%, and 1.2% glutamine on the basis of the basal diet for a period of 4 weeks. The results showed that dietary glutamine supplementation had no significant effect on stomach weight, cecum weight, and small intestine length at 35, 42, and 56 days of age, or on small intestine weight at 56 days of age ($P > 0.05$). However, dietary supplementation with 0.8% glutamine significantly increased small intestine weight at 35 and 42 days of age ($P < 0.05$). Dietary supplementation with 0.8% and 1.2% glutamine significantly increased duodenum and jejunum villus height at 35 and 42 days of age ($P < 0.05$), while dietary supplementation with 0.8% glutamine also significantly increased ileum villus height at 35 and 42 days of age and duodenum and jejunum villus height at 56 days of age ($P < 0.05$). Dietary supplementation with 0.8% glutamine significantly decreased duodenum and jejunum crypt depth at 35 and 42 days of age and ileum crypt depth at 35 days of age ($P < 0.05$). These results indicate that dietary glutamine supplementation can promote intestinal development in meat rabbits during the first 1-2 weeks post-weaning. Under the conditions of this experiment, the optimal dietary supplementation level of glutamine for meat rabbits is 0.8%.

Full Text

Effects of Glutamine on Intestinal Development of Weaned Meat Rabbits

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Abstract

This experiment was conducted to evaluate the effects of glutamine on intestinal development of meat rabbits. A total of 360 28-day-old weaned New Zealand White rabbits were randomly divided into 4 groups with 9 replicates per group and 10 rabbits per replicate. The four groups were fed experimental diets supplemented with 0 (control), 0.4%, 0.8%, and 1.2% glutamine based on a basal diet for a 4-week period. The results showed that dietary glutamine supplementation had no significant effects on stomach weight, cecum weight, and small intestine length at 35, 42, and 56 days of age, nor on small intestine weight at 56 days of age ($P > 0.05$). However, supplementation with 0.8% glutamine significantly increased small intestine weight at 35 and 42 days of age ($P < 0.05$). Dietary supplementation with 0.8% and 1.2% glutamine significantly elevated duodenum and jejunum villus height at 35 and 42 days of age ($P < 0.05$), while 0.8% glutamine also significantly increased ileum villus height at 35 and 42 days of age and duodenum and jejunum villus height at 56 days of age ($P < 0.05$). Additionally, 0.8% glutamine supplementation significantly reduced duodenum and jejunum crypt depth at 35 and 42 days of age and ileum crypt depth at 35 days of age ($P < 0.05$). These findings indicate that dietary glutamine supplementation can promote intestinal development in meat rabbits during the first two weeks post-weaning. Under the conditions of this experiment, the optimal supplemental level of glutamine in meat rabbit diets is 0.8%.

Key words: glutamine; meat rabbits; intestinal development

Introduction

In modern intensive meat rabbit production, weaning stress causes intestinal mucosal damage, decreased intestinal immune function, and reduced digestive enzyme activity, leading to indigestion, diarrhea, growth retardation, and even death [1]. Statistics indicate that mortality rates in meat rabbit production reach approximately 20% in China, with over 70% of deaths attributed to diarrhea resulting from intestinal mucosal damage and compromised immune function caused by weaning stress, representing substantial economic losses [2]. Research has demonstrated that nutritional interventions can effectively alleviate weaning-induced intestinal mucosal damage and immune dysfunction [3]. Historically, antibiotics were the primary measure to prevent weaning stress, but antibiotic residues pose safety concerns for rabbit meat products [4]. Consequently, using nutritional additives to protect the intestinal mucosal barrier and enhance immune function has become a research focus for preventing weaning diarrhea at its source.

Glutamine (Gln) is a conditionally essential amino acid with unique nutritional functions that provides energy and nitrogen sources for small intestinal mucosal cell differentiation, thereby promoting repair of damaged intestinal mucosa and maintaining normal intestinal immune function. However, weaning interrupts the supply of maternal glutamine, and the limited synthetic capacity of young animals often leads to glutamine deficiency. Therefore, exogenous glutamine supplementation is crucial for alleviating weaning stress, promoting intestinal mucosal immunity, and improving immune function in weaned animals [3-10]. While numerous studies have investigated glutamine in piglets and poultry, demonstrating its ability to maintain normal intestinal morphology, promote nutrient absorption, enhance mucosal immune function, and support intestinal health to effectively mitigate weaning stress [3,9-11], research on glutamine application in rabbits remains limited and has primarily focused on Rex rabbits [12-13]. Studies examining the effects of glutamine on meat rabbit intestinal development and determining optimal dietary inclusion levels are scarce. This experiment was designed to investigate the effects of dietary glutamine supplementation on intestinal development in weaned meat rabbits and provide a reference for scientific diet formulation.

1. Materials and Methods

1.1 Experimental Design A total of 360 28-day-old weaned New Zealand White rabbits were randomly allocated to 4 groups with 9 replicates per group and 10 rabbits per replicate (equal numbers of males and females). The four groups received experimental diets supplemented with 0 (control), 0.4%, 0.8%, and 1.2% glutamine based on a basal diet for a 4-week experimental period. Dietary glutamine levels were established based on previous research from our laboratory [2] and published studies [12-13], with alanine added for isonitrogenous regulation across treatments. At 35, 42, and 56 days of age, six rabbits per group (equal numbers of males and females) with body weights close to the group mean were selected and slaughtered 1 hour after feeding at 07:00 to collect intestinal samples for developmental measurements.

1.2 Basal Diet and Management The basal diet was formulated according to nutritional levels recommended by Gu Zilin et al. [1] and local feed resources, processed into pellet feed (3 mm diameter, 6-8 mm length). The composition and nutrient levels of the basal diet are presented in Table 1 . Prior to the experiment, rabbit housing was thoroughly cleaned and disinfected. Conventional management and immunization procedures were employed with ad libitum access to feed and water.

1.3 Sample Collection and Measurements

1.3.1 Sample Collection Following slaughter by jugular exsanguination, the abdominal cavity was immediately opened. The pyloric and ileocecal valves were

ligated, and the digestive tract was removed. The small intestine was carefully separated according to anatomical characteristics, with the duodenum, jejunum, and ileum individually ligated. Two-centimeter segments from the middle portion of each intestinal section were excised, rinsed with physiological saline, blotted with filter paper to remove excess moisture, weighed, and immediately fixed in 10% formalin solution.

1.3.2 Determination of Digestive Organ Weight and Length After carefully separating intestinal loops from mesentery, small intestine length was measured using a flexible ruler at its natural state. The stomach, small intestine, and cecum were cleaned of contents, blotted with filter paper to remove excess moisture, and weighed to obtain organ weights.

1.3.3 Measurement of Small Intestinal Mucosal Morphology Paraffin sections were prepared using conventional methods and stained with hematoxylin-eosin. Villus height and crypt depth were measured according to the method of Sun et al. [14]. Villus height was measured from the villus tip to the crypt opening, while crypt depth was measured from the invagination between adjacent villi.

1.4 Statistical Analysis Experimental data were processed using Excel 2007 software and analyzed by one-way ANOVA using SPSS 17.0 statistical software. Duncan' s multiple range test was used for post-hoc comparisons, with $P < 0.05$ as the significance threshold. Results are expressed as "mean ± standard deviation."

2. Results

2.1 Effects of Glutamine on Digestive Organ Weight and Small Intestine Length As shown in Table 2 , dietary supplementation with different glutamine levels had no significant effects on stomach weight, cecum weight, and small intestine length at 35, 42, and 56 days of age, nor on small intestine weight at 56 days of age ($P > 0.05$). However, compared with the control group, supplementation with 0.8% glutamine significantly increased small intestine weight at 35 and 42 days of age ($P < 0.05$).

2.2 Effects of Glutamine on Small Intestinal Villus Height Table 3 shows that the 0.8% and 1.2% groups exhibited significantly higher duodenum and jejunum villus height at 35 and 42 days of age compared with the control group ($P < 0.05$). The 0.8% group also showed significantly higher duodenum and jejunum villus height at 56 days of age ($P < 0.05$), with no significant differences among other groups for these parameters ($P > 0.05$). The 0.8% group demonstrated significantly higher ileum villus height at 35 and 42 days of age ($P < 0.05$), while no significant differences were observed among groups for ileum villus height at 56 days of age ($P > 0.05$).

2.3 Effects of Glutamine on Small Intestinal Crypt Depth As presented in Table 4 , the 0.8% group showed significantly lower duodenum and jejunum crypt depth at 35 and 42 days of age compared with the control group ($P < 0.05$), with no significant differences from the 0.4% and 1.2% groups ($P > 0.05$). No significant differences were observed among groups for duodenum and jejunum crypt depth at 56 days of age ($P > 0.05$). The 0.8% group exhibited significantly lower ileum crypt depth at 35 days of age ($P < 0.05$) compared with the control group, with no significant differences from the 0.4% and 1.2% groups ($P > 0.05$). No significant differences were found among groups for ileum crypt depth at 42 and 56 days of age ($P > 0.05$).

3. Discussion

3.1 Effects of Glutamine Supplementation on Digestive Organ Weight and Small Intestine Length The weight of the stomach, small intestine, cecum, and small intestine length reflect the digestive capacity of meat rabbits, particularly during the first 1-2 weeks post-weaning. Glutamine and glucose serve as primary energy sources for intestinal mucosal cell metabolism and play crucial roles in small intestinal development [15]. In this study, dietary supplementation with 0.8% glutamine significantly increased small intestine weight during the first and second weeks post-weaning but had no significant effect during the third week. Zhou Liangao et al. [16] reported that glutamine significantly increased small intestine weight and length in broilers at 2-3 weeks of age but not at 4 weeks of age, with similar findings reported by Lu Jing et al. [2] in broilers. Soltan [17] noted that glutamine promotes intestinal development more effectively during early growth stages in poultry with minimal effects during later stages. These findings align with our results, indicating that glutamine effects are development stage-dependent, primarily acting during early development. Adult animals can synthesize adequate glutamine, and suckling rabbits obtain glutamine from maternal milk. However, weaned rabbits lose this maternal source while experiencing increased glutamine requirements due to weaning stress, yet cannot synthesize sufficient glutamine. This may explain why glutamine exerted significant effects during the first two weeks post-weaning but minimal effects during the third week.

3.2 Effects of Glutamine on Small Intestinal Villus Height and Crypt Depth Research has shown that weaning stress significantly reduces small intestinal villus height and increases crypt depth in meat rabbits, typically requiring 2-3 weeks for mucosal structure recovery [2]. Glutamine is a non-essential amino acid that is the most abundant free amino acid in mammalian plasma and milk [15]. However, under stress conditions (such as weaning, injury, or burns) or pathological states, endogenous glutamine often cannot meet physiological demands, potentially leading to glutamine depletion that necessitates exogenous supplementation [18]. Glutamine oxidation provides energy for mucosal cell proliferation and can be converted into precursors required for cell proliferation [2]. Under weaning stress conditions in this study, dietary glutamine supplementa-

tion increased small intestinal villus height and reduced crypt depth during the first two weeks post-weaning, alleviating weaning-induced mucosal damage and promoting intestinal development. These findings are consistent with results reported by Liu Yujie et al. [19] in goats, Fu Zhaohui et al. [12] in Rex rabbits, and Cabrera et al. [8] in piglets. In this experiment, the optimal effects on increasing villus height and reducing crypt depth were achieved at the 0.8% supplementation level, indicating this as the optimal dietary inclusion level for meat rabbits. Fu Zhaohui et al. [12] reported that average daily gain in growing Rex rabbits increased then decreased with increasing dietary glutamine levels, peaking at 0.9% supplementation, similar to our findings. Gao Yuqi et al. [13] found that the optimal glutamine level was approximately 1.6% for young Rex rabbits during the first month post-weaning, decreasing to 0.8% during the second month. These discrepancies may be related to differences in basal diet composition, particularly acid detergent lignin and amino acid levels. Additionally, the lack of significant effects of glutamine on small intestinal mucosal development at 56 days of age in this study may be attributed to the rabbits' ability to synthesize adequate glutamine endogenously by this age.

Conclusion

Dietary glutamine supplementation can promote intestinal development in meat rabbits during the first two weeks post-weaning. Under the conditions of this experiment, the optimal supplemental level of glutamine in meat rabbit diets is 0.8%.

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Note: Figure translations are in progress. See original paper for figures.