ChinaRxiv

AI translation · View original & related papers at
chinarxiv.org/items/chinaxiv-201711.00735

Effects of Dietary -Ketoglutarate Supplementation on Nitrogen, Calcium and Phosphorus Metabolism in Growing Pigs: Postprint

Authors: Jiashun Chen, Wu Fei, Duan Yehui, Li Jianjun, Jiang Qian, Li Huan, yellow cattle, Tian Junquan, Yin Yulong, Yao Kang

Date: 2017-10-10T00:00:00+00:00

Abstract

This experiment aimed to investigate the effects of dietary -ketoglutarate (AKG) supplementation in low-protein diets on nitrogen, calcium, and phosphorus metabolism in growing pigs. Eighteen healthy crossbred (Duroc × Landrace × Yorkshire) pigs with an initial body weight of (34.98±2.18) kg were randomly divided into 3 groups according to the principle of similar body weight and identical sex ratio, with 6 replicates per group and 1 pig per replicate. The AKG supplementation levels for each group were 0 (control group), 1%, and 2%, respectively. A 14-day feeding trial was first conducted to observe growth performance; this was followed by a digestion-metabolism trial with a duration of 7 days, including a 5-day preliminary period and a 2-day feces and urine collection period (total collection method). The results showed that: 1) The 1% AKG supplementation group exhibited a trend toward improved growth performance in 35-45 kg growing pigs, specifically manifested by increased average daily gain (P=0.1942) and average daily feed intake (P=0.2583), and decreased feed-to-gain ratio (P=0.4197). 2) With increasing AKG supplementation levels, urinary nitrogen content showed a decreasing trend (P=0.1432), while fecal nitrogen, total nitrogen content, and total nitrogen excretion rate were highly significantly decreased (P<0.01), and nitrogen apparent digestibility and net protein utilization were highly significantly increased (P<0.01). Compared with the control group, the 1% and 2% AKG supplementation groups reduced urinary nitrogen content by 13.31% and 41.88%, fecal nitrogen content by 18.73% and 54.69%, total nitrogen excretion rate by 20.57% and 50.00%, increased nitrogen apparent digestibility by 2.60% and 6.32%, and increased net protein utilization by 2.68% and 6.51%, respectively. 3) The 2% AKG supplementation group had significantly lower calcium intake, fecal calcium, and fecal phosphorus content than the control group (P<0.05), while calcium apparent digestibility and phosphorus apparent

digestibility were significantly higher than those of the control group (P<0.05); urinary calcium and urinary phosphorus content showed a trend toward being lower than those of the other groups, but the differences among groups were not significant (P>0.05). These results indicate that supplementation with 1%-2% AKG in diets for 35-45 kg growing pigs can effectively reduce nitrogen, calcium, and phosphorus excretion, and improve the utilization efficiency of nitrogen, calcium, and phosphorus as well as daily gain.

Full Text

Effects of Dietary -Ketoglutarate Supplementation on Nitrogen and Calcium-Phosphorus Metabolism in Growing Pigs

CHEN Jiashun[1],[2], WU Fei[1], *DUAN Yehui[1], LI Jianjun[1], JIANG Qian[1], LI Huan[2], HUANG Niu[2], TIAN Junquan[1], YIN Yulong[1],[2], YAO Kang[1],[2]*

[1]Key Laboratory of Agro-Ecological Processes in Subtropical Region, Hunan Provincial Engineering Research Center for Healthy Breeding of Livestock and Poultry, Scientific Observing and Experimental Station of Animal Nutrition and Feed Science in South-Central China, Ministry of Agriculture, Institute of Subtropical Agriculture, Chinese Academy of Sciences, Changsha 410125, China
[2]College of Animal Science and Technology, Hunan Agricultural University, Changsha 410128, China

Abstract

This experiment investigated the effects of -ketoglutarate (AKG) supplementation in low-protein diets on nitrogen, calcium, and phosphorus metabolism in growing pigs. Eighteen healthy crossbred (Duroc × Landrace × Large White) pigs with an initial body weight of (34.98 ± 2.18) kg were randomly allocated to three groups based on similar body weight and equal gender ratio, with six replicates per group and one pig per replicate. Dietary AKG supplementation levels were 0% (control), 1%, and 2%. A 14-day feeding trial was conducted first to evaluate growth performance, followed by a 7-day digestion and metabolism trial (5-day adaptation period and 2-day total feces and urine collection period).

The results showed: (1) The 1% AKG group exhibited a tendency to improve growth performance in 35-45 kg growing pigs, as evidenced by increased average daily gain (P = 0.1942) and average daily feed intake (P = 0.2583), along with a decreased feed-to-gain ratio (P = 0.4197). (2) With increasing AKG supplementation levels, urinary nitrogen content tended to decrease (P = 0.1432), while fecal nitrogen, total nitrogen content, and total nitrogen excretion rate decreased significantly (P < 0.01), and nitrogen apparent digestibility and net protein utilization increased significantly (P < 0.01). Compared with the control group, the 1% and 2% AKG groups reduced urinary nitrogen by 13.31%

and 41.88%, fecal nitrogen by 18.73% and 54.69%, and total nitrogen excretion rate by 20.57% and 50.00%, respectively. Nitrogen apparent digestibility increased by 2.60% and 6.32%, and net protein utilization increased by 2.68% and 6.51%, respectively. (3) The 2% AKG group significantly reduced calcium intake, fecal calcium, and fecal phosphorus content compared to the control group ($P < 0.05$), while significantly increasing calcium and phosphorus apparent digestibility ($P < 0.05$). Urinary calcium and phosphorus levels showed a decreasing trend, though differences among groups were not significant ($P > 0.05$). These findings indicate that dietary supplementation with 1–2% AKG effectively reduces nitrogen, calcium, and phosphorus excretion, improves their utilization efficiency, and enhances daily weight gain in 35–45 kg growing pigs.

Keywords: -ketoglutarate; growing pigs; nitrogen metabolism; calcium-phosphorus metabolism

Introduction

In recent years, research and application of synthetic amino acid supplementation in low-protein diets have gained increasing attention as a strategy to conserve protein feed resources, reduce feed costs, alleviate animal stress, and minimize environmental pollution from animal waste. Rotz [1] reported that reducing dietary protein levels decreases nitrogen excretion to varying degrees, and that modifying diet composition and structure can reduce nitrogen emission rates by 32–62% [2]. -Ketoglutarate (AKG), a key precursor of glutamine, offers superior stability and solubility in solution compared to glutamine without imposing additional nitrogen burdens on the body, and it is non-toxic [3]. Through the action of glutamate dehydrogenase or transaminases, AKG generates glutamate, which can then be converted to glutamine via glutamine synthetase. AKG shares similar physiological functions with glutamine, such as promoting intestinal development, leading to growing interest in its use as a glutamine alternative in production practices [4].

Numerous studies have demonstrated AKG' s important role in maintaining nitrogen balance, reducing nitrogen losses, and promoting protein synthesis. In rats, Jeevanandam et al. [5] found that dietary supplementation with 215 mol/L AKG significantly reduced nitrogen losses and increased nitrogen retention. This finding was confirmed by Piva et al. [6], who observed that AKG supplementation (3 and 6 g/kg) in nitrogen-free diets reduced urinary nitrogen by 18% regardless of dosage. Using growing rats, Prandini et al. [7] reported that dietary AKG (3–6 g/kg) significantly reduced endogenous urinary nitrogen losses and showed a tendency to decrease endogenous fecal nitrogen. Additionally, AKG (2 g/kg) effectively improved negative nitrogen balance and promoted muscle protein synthesis in post-surgical and burn patients [8]. AKG also plays a significant role in bone development and mineral deposition. Calcium and phosphorus are essential minerals that constitute the primary components of bone

and teeth while participating in metabolic regulation [9]. Harrison et al. [10] demonstrated that dietary AKG supplementation (0.1 g/kg) significantly increased trabecular and cortical bone density in lambs, while Kowalik et al. [11] found that AKG supplementation (0.4 g/kg) significantly increased bone mineral density in piglets.

However, most research on AKG' s regulation of nitrogen metabolism has focused on rodents and humans, and studies on its effects on calcium-phosphorus metabolism are scarce. Our research team previously reported that AKG (2 mmol/L) promotes protein synthesis and inhibits protein degradation in porcine intestinal epithelial cells [12]. Given AKG' s regulatory effects on protein metabolism in intestinal cells, we hypothesized that AKG could modulate nitrogen metabolism in pigs. Furthermore, current feeding standards indicate that reducing dietary crude protein by 2–4 percentage points is feasible in growing-finishing pigs [13,14]. Therefore, this study investigated the effects of AKG supplementation in low-protein diets (with 4 percentage points reduced crude protein and supplemented limiting amino acids) on nitrogen, calcium, and phosphorus metabolism in growing pigs to provide a theoretical basis for the rational application of AKG in sustainable pig production.

1.1 Experimental Material

AKG (purity 99.0%) was provided by Hubei Yuancheng Saichuang Technology Co., Ltd.

1.2 Experimental Diets

Corn and soybean meal served as the primary starch and protein sources, respectively. Diets were formulated according to NRC (2012) [15] nutrient requirements for 30–60 kg pigs. Dietary AKG supplementation levels were 0% (control), 1%, and 2%. Feed ingredients were ground, mixed progressively, and prepared as powder for storage in a ventilated, dry location. The composition and nutrient levels of experimental diets are presented in Table 1 .

1.3 Experimental Animals and Design

Eighteen healthy crossbred (Duroc × Landrace × Large White) pigs with an initial body weight of (34.98 ± 2.18) kg were randomly allocated to three groups based on similar body weight and equal gender ratio, with six replicates per group and one pig per replicate. Dietary AKG supplementation levels were 0% (control), 1%, and 2%. A 14-day feeding trial was conducted first to evaluate growth performance, followed by a digestion and metabolism trial in stainless steel metabolism crates. After a 5-day adaptation period, feces and urine were collected for 2 days using the total collection method.

1.4 Animal Management

The experiment was conducted in the metabolism room of the Animal Experimental Building at the Institute of Subtropical Agriculture, Chinese Academy of Sciences. Pigs were housed individually in stainless steel metabolism crates and fed twice daily at 08:30 and 16:30 with ad libitum access to feed and water. Daily feed intake was recorded accurately, and animal health was monitored throughout the trial. Routine cleaning and disinfection were performed, and the facility was maintained ventilated and clean.

1.5 Feces and Urine Collection

During the digestion and metabolism trial, feces and urine were collected daily at 08:00 and 16:00. Fresh feces were weighed, and 10 mL of 10% dilute sulfuric acid was added per 15 g of fresh feces to prevent ammonia nitrogen loss before storage at -80°C. Urine volume was measured, and after thorough mixing, one-fifth of the total volume was collected. Ten milliliters of 10% dilute sulfuric acid was added per 100 mL of urine to prevent ammonia volatilization and sample spoilage, and samples were stored at -80°C.

1.6 Measurements and Methods

Pigs were weighed after overnight fasting on day 1 and day 14 of the feeding trial to determine initial and final body weights for calculating average daily gain (ADG). Daily feed allowance and refusals were recorded to calculate average daily feed intake (ADFI) and feed-to-gain ratio (F/G). Nitrogen content in diets, feces, and urine was determined using the Kjeldahl method, while calcium and phosphorus contents were measured by inductively coupled plasma mass spectrometry. Calculations were performed as follows:

- Absorbed nitrogen = nitrogen intake - fecal nitrogen
- Retained nitrogen = nitrogen intake - fecal nitrogen - urinary nitrogen
- Nitrogen apparent digestibility = 100 × absorbed nitrogen / nitrogen intake
- Nitrogen apparent biological value = 100 × retained nitrogen / absorbed nitrogen
- Net protein utilization = 100 × retained nitrogen / nitrogen intake
- Total nitrogen excretion rate = 100 × (fecal nitrogen + urinary nitrogen) / nitrogen intake
- Calcium apparent digestibility = 100 × (calcium intake - fecal calcium) / calcium intake
- Phosphorus apparent digestibility = 100 × (phosphorus intake - fecal phosphorus) / phosphorus intake

1.7 Statistical Analysis

Data were initially processed using Excel 2013. Covariance analysis was performed using SPSS 20.0, and one-way ANOVA was used for significance testing.

Duncan' s multiple comparison test was applied for significant differences, with $P < 0.05$ considered statistically significant. Results are expressed as means ± standard error.

Results

2.1 Effects of Dietary AKG Supplementation on Growth Performance

As shown in Table 2 , the 1% AKG group exhibited a tendency to improve growth performance in growing pigs, manifested as increased average daily gain ($P = 0.1942$) and average daily feed intake ($P = 0.2583$), along with a decreased feed-to-gain ratio ($P = 0.4197$).

2.2 Effects of Dietary AKG Supplementation on Nitrogen Metabolism

Table 3 shows that with increasing AKG supplementation levels, urinary nitrogen content tended to decrease ($P = 0.1432$), while fecal nitrogen, total nitrogen content, and total nitrogen excretion rate decreased significantly ($P < 0.01$). Nitrogen apparent digestibility and net protein utilization increased significantly ($P < 0.01$). Compared with the control group, the 1% and 2% AKG groups reduced urinary nitrogen by 13.31% and 41.88%, fecal nitrogen by 18.73% and 54.69%, and total nitrogen excretion rate by 20.57% and 50.00%, respectively. Nitrogen apparent digestibility increased by 2.60% and 6.32%, and net protein utilization increased by 2.68% and 6.51%, respectively.

2.3 Effects of Dietary AKG Supplementation on Calcium and Phosphorus Metabolism

As shown in Table 4 , the 2% AKG group exhibited a tendency for lower urinary calcium and phosphorus compared to other groups, though differences were not significant ($P > 0.05$). However, calcium intake, fecal calcium, and fecal phosphorus were significantly lower than the control group ($P < 0.05$), while calcium and phosphorus apparent digestibility were significantly higher ($P < 0.05$).

Discussion

AKG has demonstrated effects on growth performance in livestock and poultry, with most research focusing on its growth-promoting effects in broilers and piglets. Yu et al. [16] reported that dietary supplementation with 0.7% AKG significantly increased body weight and average daily gain in 2-week-old broilers. Hu [17] found that 1% AKG supplementation improved piglet growth performance, increasing average daily gain by 9%. Additionally, 1% AKG alleviated growth inhibition induced by lipopolysaccharide stress in weaned piglets [18]. Our results align with these findings, showing that 1% AKG supplementation improved growth performance in growing pigs.

Low-protein diets reduce nitrogen excretion and alleviate environmental pollution pressure [14,19], with improved nutrient digestibility—particularly increased nitrogen retention—being the primary mechanism for reduced nitrogen excretion [20]. AKG serves as a common carbon skeleton for glutamate family amino acids and can rapidly generate glutamate and subsequently glutamine. It also contributes to the synthesis of other amino acids from glutamate, playing a crucial physiological role in amino acid metabolism [21]. Jeevanandam et al. [5] demonstrated that AKG supplementation (215 mol/L) significantly reduced nitrogen losses and increased nitrogen retention in rats. Piva et al. [6] confirmed these results, showing that AKG (3 and 6 g/kg) in nitrogen-free diets reduced urinary nitrogen by 18% regardless of dosage. Our study, conducted in low-protein diets, showed that 1% and 2% AKG reduced urinary nitrogen by 13.31% and 41.88%, respectively, with greater reductions at higher supplementation levels. These findings collectively demonstrate that dietary AKG regulates nitrogen metabolism and reduces nitrogen emissions, with fecal nitrogen reduction playing a decisive role in decreasing total nitrogen excretion. Piva et al. [6] also reported that 3 and 6 g/kg AKG increased small intestinal epithelial cell length by 25% and 49% ($P < 0.01$), respectively, and that 6 g/kg AKG decreased plasma essential amino acid content by 22.2%. Wei et al. [22] found that AKG supplementation (7.5 and 15.0 g/kg) in low-protein diets promoted amino acid metabolism in the liver and pancreas of Songpu mirror carp, improved protein utilization, and enhanced protein synthesis. These results suggest that AKG' s ability to reduce nitrogen losses and improve nitrogen utilization may be attributed to its promotion of intestinal cell growth, enhanced intestinal absorption function, and regulation of protein synthesis and degradation, though specific mechanisms require further investigation. In our study, AKG supplementation in low-protein diets (15.99% crude protein) significantly reduced total nitrogen excretion rate (primarily through fecal nitrogen reduction), increased net protein utilization and nitrogen apparent biological value, and showed tendencies to improve nitrogen retention and apparent digestibility, with 2% supplementation being optimal. This indicates that dietary AKG supplementation reduces protein catabolism and enhances protein synthesis, thereby promoting nitrogen deposition [8,23]. The underlying mechanism involves AKG serving as a glutamine precursor that provides energy and nitrogen sources for intestinal epithelial and immune cells while reducing intestinal glutamine catabolism, thereby supporting gastrointestinal cell metabolism and maintaining intestinal barrier integrity and absorptive function [24,25].

Calcium and phosphorus play vital roles in bone growth and metabolism and are indispensable minerals for skeletal development and maintenance [26]. Previous research indicates that AKG regulates bone metabolism through: (1) synthesis of glutamate, which acts as a signaling molecule in the nervous system to regulate bone metabolism [27]; and (2) metabolism to proline, which is hydroxylated to hydroxyproline—an essential amino acid for connective tissue and bone collagen synthesis, with collagen being a major component of bone matrix [4]. Tatara et al. [28] observed increased plasma proline concentration and bone

mineral density in newborn lambs after 14 days of AKG injection (0.1 g/kg). Andersen et al. [29] reported significantly increased femur mineral density in piglets fed AKG (0.1 g/kg) from days 21–24. These studies validate AKG' s regulatory role in bone metabolism and its positive effects on skeletal mineral deposition [10]. However, the mechanisms underlying AKG' s regulation of skeletal mineral deposition remain poorly understood. Hu et al. [30] found that low-protein diets (12.59%) did not significantly affect calcium-phosphorus apparent digestibility or excretion in finishing pigs, and Yin et al. [31] reported similar findings in growing pigs fed low-protein diets (14.13%). In contrast, our study demonstrated that 2% AKG supplementation in low-protein diets (15.99%) significantly reduced fecal calcium and phosphorus content while increasing their apparent digestibility. These discrepancies may be attributed to incomplete separation of feces and urine in previous digestion trials, potentially compromising results. Additionally, earlier studies examined only low-protein diets without AKG supplementation, which may be insufficient to improve calcium-phosphorus metabolism. Our findings suggest that AKG can increase calcium and phosphorus deposition [32,33], thereby reducing their excretion in feces and urine. These results demonstrate that AKG effectively improves calcium-phosphorus metabolism, though its effects on bone development require further investigation.

In conclusion, based on growth performance and nitrogen, calcium, and phosphorus metabolism trials, dietary supplementation with 1–2% AKG effectively reduces nitrogen, calcium, and phosphorus emissions, improves their utilization efficiency, and enhances daily weight gain in growing pigs weighing 35–45 kg.

References

[1] ROTZ C A. Management to reduce nitrogen losses in animal production[J]. Journal of Animal Science, 2004, 82(13_Suppl): E119-E137.

[2] SUTTON A L, KEPHART K B, VERSTEGEN M W, et al. Potential for reduction of odorous compounds in swine manure through diet modification[J]. Journal of Animal Science, 1999, 77(2): 430–439.

[3] WU G, MEIER S A, KNABE D A. Dietary glutamine supplementation prevents jejunal atrophy in weaned pigs[J]. The Journal of Nutrition, 1996, 126(10): 2578–2584.

[4] KRISTENSEN N B, JUNGVID H, FERNÁNDEZ J A, et al. Absorption and metabolism of -ketoglutarate in growing pigs[J]. Journal of Animal Physiology and Animal Nutrition, 2002, 86(7/8): 239–245.

[5] JEEVANANDAM M, ALI M R, RAMIAS L, et al. Efficacy of ornithine-alpha-ketoglutarate (OKGA) as a dietary supplement in growing rats[J]. Clinical Nutrition, 1991, 10(3): 155–161.

[6] PIVA A, MORLACCHINI M, PRANDINI A, et al. -Ketoglutaric acid reduces nitrogen losses in rats fed nitrogen-free diet[M]//LINDBERG J E, OGLE

B. Digestive physiology in pigs. Lewiston, NY, U.S.A.: CABI Publishing, 2001: 101–103.

[7] PRANDINI A, MORLACCHINI M, SIGOLO S, et al. Anticatabolic activity of alpha-ketoglutaric acid in growing rats[J]. Italian Journal of Animal Science, 2012, 11(3): 279–284.

[8] BLOMQVIST B I, HAMMARQVIST F, VON DER DECKEN A, et al. Glutamine and -ketoglutarate prevent the decrease in muscle free glutamine concentration and influence protein synthesis after total hip replacement[J]. Metabolism Clinical and Experimental, 1995, 44(9): 1215–1222.

[9] ZHANG T Y, ZHANG Y L, YAN S M, et al. Study on determination of endogenous calcium and phosphorus excretion and true digestibility of calcium and phosphorus in soybean meal for growing pigs using linear regression method[J]. Acta Veterinaria et Zootechnica Sinica, 2008, 39(12): 1684–1691.

[10] HARRISON A P, TYGESEN M P, SAWA-WOJTANOWICZ B, et al. -Ketoglutarate treatment early in postnatal life improves bone density in lambs at slaughter[J]. Bone, 2004, 35(1): 204–209.

[11] KOWALIK S, ŚLIWA E, TATARA M R, et al. Influence of alpha-ketoglutarate on mineral density and geometrical and mechanical parameters of femora during postnatal life in piglets[J]. Bulletin of the Veterinary Institute in Pulawy, 2005, 49(1): 107–111.

[12] YAO K, YIN Y L, LI X L, et al. Alpha-ketoglutarate inhibits glutamine degradation and enhances protein synthesis in intestinal porcine epithelial cells[J]. Amino Acids, 2012, 42(6): 2491–2500.

[13] LE BELLEGO L, VAN MILGEN J, NOBLET J. Effect of high temperature and low-protein diets on the performance of growing-finishing pigs[J]. Journal of Animal Science, 2002, 80(3): 691–701.

[14] KERR B J, SOUTHERN L L, BIDNER T D, et al. Influence of dietary protein level, amino acid supplementation, and dietary energy levels on growing-finishing pig performance and carcass composition[J]. Journal of Animal Science, 2003, 81(12): 3075–3087.

[15] National Research Council. Nutrient requirements of swine[S]. 11th ed. Washington, D.C.: National Academy Press, 2012.

[16] YU Q P, CHEN Y Q, XIE J C, et al. Effects of dietary -ketoglutarate supplementation on growth performance and tissue organ development in broilers[J]. China Animal Husbandry & Veterinary Medicine, 2010, 37(10): 10–14.

[17] HU Q Z. Effects of -ketoglutarate on growth performance and intestinal function in weaned piglets[D]. Master' s thesis. Wuhan: Wuhan Polytechnic University, 2008: 15–16.

[18] LIU J, HOU Y Q, DING B Y, et al. Alleviating effect of -ketoglutarate on growth inhibition induced by lipopolysaccharide stress in weaned piglets[J].

Chinese Journal of Animal Nutrition, 2009, 21(4): 519-524.

[19] LE BELLEGO L, NOBLET J. Performance and utilization of dietary energy and amino acids in piglets fed low protein diets[J]. Livestock Production Science, 2002, 76(1/2): 45-58.

[20] LIANG F G. Study on the balance pattern of digestible lysine, methionine + cysteine, threonine, and tryptophan in low-protein diets for growing pigs[D]. PhD thesis. Beijing: China Agricultural University, 2005: 4-8.

[21] LAMBERT B D, FILIP R, STOLL B, et al. First-pass metabolism limits the intestinal absorption of enteral -ketoglutarate in young pigs[J]. The Journal of Nutrition, 2006, 136(11): 2779-2784.

[22] WEI Y Y, XU Q Y, LI J N, et al. Effects of -ketoglutarate supplementation in diets with different protein levels on growth performance, body composition, and serum biochemical indices of Songpu mirror carp[J]. Chinese Journal of Animal Nutrition, 2013, 25(12): 2958-2965.

[23] HUANG G Q, YU Q P, CHEN Y Q, et al. Effects of -ketoglutarate on dietary metabolic energy and protein metabolism in yellow-feathered broilers[J]. China Feed, 2012(18): 22-24.

[24] CHEN L X, LI P, WANG J J, et al. Catabolism of nutritionally essential amino acids in developing porcine enterocytes[J]. Amino Acids, 2009, 37(1): 143-152.

[25] JONES C, PALMER T E A, GRIFFITHS R. Randomized clinical outcome study of critically ill patients given glutamine-supplemented enteral nutrition[J]. Nutrition, 1999, 15(2): 108-115.

[26] WANG J, WANG D, HE J P, et al. Effects of oral calcium supplementation on calcium and phosphorus metabolism in Gansu zokors[J]. Chinese Journal of Zoology, 2010, 45(4): 46-51.

[27] STOLL B, HENRY J, REEDS P J, et al. Catabolism dominates the first-pass intestinal metabolism of dietary essential amino acids in milk protein-fed piglets[J]. The Journal of Nutrition, 1998, 128(3): 606-614.

[28] TATARA M R, TYGESEN M P, SAWA-WOJTANOWICZ B, et al. Bone development: the effect of short-term alpha-ketoglutarate administration on long-term mechanical properties of ribs in ram lambs[J]. Small Ruminant Research, 2007, 67(2/3): 179-183.

[29] ANDERSEN N K, TATARA M R, KRUPSKI W, et al. The long-term effect of -ketoglutarate, given early in postnatal life, on both growth and various bone parameters in pigs[J]. Journal of Animal Physiology and Animal Nutrition, 2008, 92(5): 519-528.

[30] HU Q, ZHU J P, LIU C X, et al. Effects of low-protein diets on nutrient digestibility and excretion in finishing pigs[J]. Journal of Domestic Animal Ecology, 2014, 35(3): 74-77.

[31] YIN H H, ZHANG S R, SUN J G, et al. Effects of low-protein diets with different net energy levels on growth performance and nutrient digestibility in pigs[J]. Chinese Journal of Animal Science, 2008, 44(13): 25-28.

[32] TATARA M R, MAJCHER P, KRUPSKI W, et al. Influence of alpha-ketoglutarate on cortical bone density, geometrical properties and mechanical endurance of the humerus in turkeys[J]. Bulletin of the Veterinary Institute in Pulawy, 2004, 48(4): 461-465.

[33] TATARA M R, BRODZKI A, KRUPSKI W, et al. Effects of alpha-ketoglutarate on bone homeostasis and plasma amino acids in turkeys[J]. Poultry Science, 2005, 84(10): 1604-1609.

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

Source: ChinaXiv —Machine translation. Verify with original.