Effects of Dietary Phosphorus Levels on Lactation Performance and Serum Biochemical Indices in Laoshan Dairy Goats (Postprint)

Authors: Wang Huimin, Zhu Fenghua, Ge Wei, Cao Yufang, Cheng Ming, Lin Yingting

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Abstract

This experiment was conducted to investigate the effects of dietary phosphorus level on lactation performance and serum biochemical indices of Laoshan dairy goats. Thirty healthy second-parity Laoshan dairy goats in mid-lactation with body weight of (56.55$±1.17)*kgandmilkyieldof*(2.20±$0.07) kg/d were selected and randomly allocated to 3 groups using a single-factor completely randomized design, with 10 goats per group and each goat serving as one replicate. All groups were fed diets with consistent energy and protein levels, but with phosphorus levels of 0.26%, 0.36%, and 0.46%, respectively. The preliminary period lasted 15 d, and the formal experimental period lasted 60 d. The results showed that: 1) Dietary phosphorus level had no significant effect on dry matter intake (P>0.05). 2) The 0.36% group had a higher average milk yield over the entire period, which was extremely significantly higher than that of the 0.26% and 0.46% groups (P<0.01), while the difference between the 0.26% and 0.46% groups was not significant (P>0.05); the dry matter intake/milk yield ratio and dry matter intake/4% fat-corrected milk yield ratio were both lower in the 0.36% group, being extremely significantly lower than those of the 0.46% group (P<0.01). 3) Dietary phosphorus level had no significant effects on milk fat percentage, milk protein percentage, lactose percentage, milk solids-not-fat percentage, milk phosphorus content, or milk calcium content (P>0.05). 4) Dietary phosphorus level had no significant effects on serum calcium, phosphorus, urea nitrogen contents, or alkaline phosphatase activity (P>0.05). Therefore, a dietary phosphorus level of 0.36% is appropriate for lactating Laoshan dairy goats.

Full Text

Effects of Dietary Phosphorus Level on Lactation Performance and Serum Biochemical Indices of Laoshan Dairy Goats

WANG Huimin[1], ZHU Fenghua[1], GE Wei[1], CAO Yufang[1], CHENG Ming[2], LIN Yingting[1]*

[1]College of Animal Science and Technology, Qingdao Agricultural University, Qingdao 266109, China
[2]Institute of Husbandry and Veterinary of Qingdao, Qingdao 266109, China

Abstract

This experiment was conducted to investigate the effects of dietary phosphorus level on lactation performance and serum biochemical indices of Laoshan dairy goats. Thirty healthy second-parity Laoshan dairy goats in mid-lactation, with an average body weight of $(56.55\$\pm1.17)kgandmilkyieldof(2.20\pm\$0.07)$ kg/d, were randomly allocated into three groups using a single-factor randomized design, with 10 goats per group and each goat serving as one replicate. The three groups were fed diets with consistent energy and protein levels but different phosphorus levels of 0.26%, 0.36%, and 0.46%, respectively. The experiment consisted of a 15-day preliminary period followed by a 60-day formal experimental period. The results showed that: (1) dietary phosphorus level had no significant effect on dry matter intake (P>0.05); (2) the 0.36% phosphorus group exhibited higher average milk yield throughout the experimental period, which was extremely significantly higher than that of the 0.26% and 0.46% groups (P<0.01), while no significant difference was observed between the 0.26% and 0.46% groups (P>0.05); the ratios of dry matter intake to milk yield and dry matter intake to 4% fat-corrected milk yield were lower in the 0.36% group, being extremely significantly lower than those in the 0.46% group (P<0.01); (3) dietary phosphorus level had no significant effects on milk fat percentage, milk protein percentage, lactose percentage, milk non-fat solids percentage, milk phosphorus content, or milk calcium content (P>0.05); (4) dietary phosphorus level had no significant effects on serum calcium, phosphorus, or urea nitrogen contents, nor on alkaline phosphatase activity (P>0.05). These findings indicate that under the conditions of this experiment, a dietary phosphorus level of 0.36% is appropriate for lactating Laoshan dairy goats.

Keywords: Laoshan dairy goats; phosphorus level; lactation performance; serum biochemical indices

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Laoshan dairy goats are an excellent dairy goat breed in China, characterized by strong adaptability, roughage tolerance, robust physique, high milk yield, and stable genetic performance [1-2]. Scientific feeding and management according

to their nutritional requirements are essential for fully exploiting their productive potential. Phosphorus is an essential macro-mineral element in animals with important physiological functions. Dietary phosphorus deficiency or inadequate supply, as well as improper calcium-to-phosphorus ratios, can cause phosphorus metabolism disorders leading to adverse reactions or diseases in animals. Conversely, excessive dietary phosphorus levels are detrimental to calcium and other divalent mineral absorption in dairy goats and result in excessive phosphorus excretion into the environment, causing environmental pollution. In recent years, numerous studies have reported on phosphorus requirements in goat diets [3-4] and dairy cow diets [5-6], but research on phosphorus levels in dairy goat diets remains limited. This study investigated the effects of dietary phosphorus level on lactation performance and serum biochemical indices of lactating Laoshan dairy goats, aiming to determine the appropriate phosphorus level for lactating Laoshan dairy goats and provide reference for healthy dairy goat farming.

1.1 Experimental Animals and Design

Thirty second-parity Laoshan dairy goats from Qingdao Aote Laoshan Dairy Goat Original Breed Farm were selected, with lactation days of $(120\$\pm1.52)d, body weight of (56.55\pm1.17)kg, and milk yield of (2.20\pm\$0.07)$ kg/d. Using a single-factor experimental design, the goats were divided into three groups (A, B, and C) with 10 goats per group, each goat serving as one replicate. The experimental period lasted 75 days, including a 15-day preliminary period and a 60-day formal experimental period. The milk yield and milk composition of the dairy goats at the beginning of the experiment are shown in Table 1 .

1.2 Experimental Diets and Nutrient Levels

The experimental diets were formulated based on the NRC (2007) goat feeding standards. According to the milk yield, lactation days, and body weight of Laoshan dairy goats, the required dietary phosphorus level was calculated to be 0.36%, and three experimental diets with phosphorus levels of 0.26%, 0.36%, and 0.46% were designed with essentially consistent energy and protein levels. The composition and nutrient levels of the experimental diets are shown in Table 2 , and the diets were fed in the form of total mixed ration (TMR).

1.3 Feeding Management

The experimental goats were housed in individual pens and fed at 06:00, 10:30, 15:30, and 20:00 daily, with ad libitum access to feed and clean drinking water. Mechanical milking was performed at 06:00 and 18:00 daily, with feed intake and herd health status recorded simultaneously. The feeding and management conditions were identical across all groups.

1.4 Sample Collection

1.4.1 Diet Samples Concentrate, corn silage, and peanut vine samples were collected using the quartering method, dried in a 65°C oven to produce air-dried samples, and then ground for analysis.

1.4.2 Milk Samples Milk samples were collected continuously for 2 days on days 0, 30, and 60 of the formal experimental period. Sampling times were at 18:00 on the first day and 06:00 on the second day of each sampling period. Samples collected on the first day were stored at 4°C, then mixed uniformly with the second day' s samples after the second collection and stored at -20°C.

1.4.3 Serum Samples Serum samples were collected on days 0, 30, and 60 of the formal experimental period. Five milliliters of jugular blood were collected from each experimental goat using heparin sodium anticoagulant tubes, allowed to stand for 2 h, then centrifuged at 1,369×g for 10 min. The supernatant was transferred to 2 mL centrifuge tubes and stored at -20°C.

1.5 Determination Indicators and Methods

1.5.1 Diet Nutrient Analysis Dietary crude protein content was determined using the Kjeldahl method according to "Feed Analysis and Feed Quality Detection Technology" [9]. Calcium content was determined using the potassium permanganate titration method, and phosphorus content was determined using the molybdenum yellow colorimetric method. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) contents were determined according to the method proposed by Van Soest et al. [10].

1.5.2 Dry Matter Intake (DMI) Feed offered and refusals were accurately recorded daily before and after feeding. Dry matter intake was calculated according to the method described in "Feed Analysis and Feed Quality Detection Technology" [9].

1.5.3 Milk Yield Milk yield was recorded at each milking and summed to obtain daily milk production.

2.1 Effects of Dietary Phosphorus Level on Dry Matter Intake

As shown in Table 3 , dietary phosphorus level had no significant effect on dry matter intake of lactating Laoshan dairy goats ($P>0.05$). However, all groups showed a decreasing trend in dry matter intake from days 31-60 compared to days 1-30.

Table 3 Effects of dietary phosphorus level on DMI of Laoshan dairy goats (kg/(d · goat))

Items	Group A	Group B	Group C	P-value
1-30 d	2.15$\pm$0.02	2.14$\pm$0.03	2.10$\pm$0.03	
31−60*d*	2.10$\pm$0.02	2.12$\pm$0.04	2.06$\pm$0.02	
Average	2.13$\pm$0.02	2.13$\pm$0.02	2.08$\pm$$0.02	

Values in the same row with different small letter superscripts indicate significant difference (P<0.05), different capital letter superscripts indicate extremely significant difference (P<0.01), and same or no letter superscripts indicate no significant difference (P>0.05). The same applies below.

2.2 Effects of Dietary Phosphorus Level on Milk Yield of Laoshan Dairy Goats

As shown in Table 4 , there were no significant differences in milk yield among groups during days 1-30 (P>0.05). During days 31-60, Group B had the highest milk yield, which was extremely significantly higher than Groups A and C (P<0.01). For the overall average, Group B had the highest milk yield, extremely significantly higher than Groups A and C (P<0.01). During days 1-30, 4% fat-corrected milk (FCM) yield in Groups A and B was extremely significantly higher than in Group C (P<0.01). During days 31-60, Group B had the highest 4% FCM yield, extremely significantly higher than Groups A and C (P<0.01), while Group A was extremely significantly higher than Group C (P<0.01). For the overall average, 4% FCM yield in Groups A and B was extremely significantly higher than in Group C (P<0.01), and Group B was significantly higher than Group A (P<0.05).

Table 4 Effects of dietary phosphorus level on MY of Laoshan dairy goats (kg/(d · goat))

Items	Group A	Group B	Group C	P-value
Milk yield				
1-30 d	2.22$\pm$0.02	2.25$\pm$0.04	2.15$\pm$0.04	
31−60*d*	1.92$\pm$0.03*Bb*	2.06$\pm$0.04*Bb*	1.90$\pm$0.03*Bb*	
Average	2.07$\pm$0.03	2.15$\pm$0.04*Aa*	2.02$\pm$0.0	
∗4	2.21$\pm$0.03*Aa*	1.97$\pm$0.03*Aa*		
31−60*d*	1.81$\pm$0.03*Bb*	1.98$\pm$0.04*Aa*	1.69$\pm$0.02*Cc*	
Average	2.00$\pm$0.02*Aa*	2.09$\pm$0.02*Aa*	1.83$\pm$	

2.3 Effects of Dietary Phosphorus Level on Feed-to-Milk Ratio of Laoshan Dairy Goats

As shown in Table 5 , for the ratio of dry matter intake to milk yield, there were no significant differences among groups during days 1-30 (P>0.05). During days 31-60, Groups A and C were both extremely significantly higher than Group B (P<0.01). For the overall average, Groups A and C were both extremely significantly higher than Group B (P<0.01). For the ratio of dry matter intake to 4% FCM yield, there was an increasing trend with increasing dietary phosphorus

levels during days 1-30, with Group C being extremely significantly higher than Group A (P<0.01) and significantly higher than Group B (P<0.05), while no significant difference was observed between Groups A and B (P>0.05). During days 31-60, Group C was extremely significantly higher than Group B (P<0.01) and significantly higher than Group A (P<0.05), while Group A was extremely significantly higher than Group B (P<0.01). For the overall average, Group C was extremely significantly higher than Groups A and B (P<0.01), with no significant difference between Groups A and B (P>0.05).

Table 5 Effects of dietary phosphorus level on feed to milk ratio of Laoshan dairy goats

Items	Group A	Group B	Group C	P-value
DMI/MY				
1-30 d	0.97$±0.01|0.96±0.02|0.98±0.02|||31−60*d*|1.10±0.02*Aa*|0.99±0.02*Bb*|1.09±0.01*Aa*|||*Average*|1.03±0.02*Aa*|0.97±0.01*Bb*|1.04±*DMI*/4±0.01*Bb*|1.09±0.02*ABb*|1.15±0.02*Aa*|||31−60*d*|1.17±0.02*Ab*|1.07±0.02*Bc*|1.22±0.02*Aa*|||*Average*|1.12±0.01*Bb*|1.08±0.01*Bb*|1.19±			

2.4 Effects of Dietary Phosphorus Level on Milk Composition of Laoshan Dairy Goats

As shown in Table 6 , milk fat percentage showed a decreasing trend with increasing phosphorus levels, but the differences among groups were not significant (P>0.05). Milk phosphorus content showed an increasing trend with increasing phosphorus levels, but the differences among groups were not significant (P>0.05). No significant differences were observed among groups in milk protein percentage, lactose percentage, milk non-fat solids percentage, or milk calcium content (P>0.05).

Table 6 Effects of dietary phosphorus level on milk composition of Laoshan dairy goats

Items	Group A	Group B	Group C	P-value
Milk fat percentage/%	3.61$±0.26\|3.48$±0.08|3.27±0.09|||*Milk protein percentage*/±0.03|2.79±0.05|2}$ mg/g)			

2.5 Effects of Dietary Phosphorus Level on Serum Biochemical Indices of Laoshan Dairy Goats

As shown in Table 7 , dietary phosphorus level had no significant effects on serum phosphorus, calcium, or urea nitrogen contents, nor on alkaline phosphatase activity (P>0.05).

Table 7 Effects of dietary phosphorus level on serum biochemical indices of Laoshan dairy goats

Items	Group A	Group B	Group C	P-value
P/(mmol/L)	1.82$±0.20	2.17±0.04	2.13±0.19	|||$Ca/(mmol/L)$ | 2.16±0.06 | 2.00±0.20 | 2.14±0.06 |||A

3.1 Effects of Dietary Phosphorus Level on Dry Matter Intake of Laoshan Dairy Goats

Dry matter intake affects the production performance of lactating ruminants. Song Fancheng [11] fed dairy cows diets with phosphorus levels of 0.27%, 0.35%, 0.43%, 0.51%, and 0.59% and found that dietary phosphorus level had no significant effect on feed intake. Zhao Hengju et al. [6] reported that diets with phosphorus levels of 0.32%, 0.44%, and 0.56% had no significant effect on dairy cow feed intake. Ge Xin et al. [5] fed dairy cows of different parities and milk yields diets with phosphorus levels of 0.38% and 0.55% and similarly found that phosphorus level had no significant effect on feed intake. In this experiment, the average dry matter intake across groups showed that diets with phosphorus levels of 0.26%, 0.36%, and 0.46% had no significant effect on dry matter intake of Laoshan dairy goats, consistent with the aforementioned studies. This can be attributed to the fact that, except for mineral phosphorus levels, the other nutrient levels were basically identical across the three experimental diets, and the feeding management was also the same. In summary, adding different levels of mineral phosphorus did not affect the dry matter intake of Laoshan dairy goats.

3.2 Effects of Dietary Phosphorus Level on Milk Yield and Feed-to-Milk Ratio of Laoshan Dairy Goats

Feeding management and nutrient level are two important factors determining ruminant milk yield. Phosphorus, as an essential mineral element for dairy goats, is closely related to milk production performance. Reports on the effects of phosphorus on dairy cow milk production performance have been inconsistent. Song Fancheng [11] found that when dietary phosphorus ranged from 0.27% to 0.59% (DM basis), increasing or decreasing phosphorus levels did not cause significant changes in milk yield. However, more studies have reported different results. Huang Wenming et al. [12] fed high-yielding dairy cows diets with different phosphorus levels (0.34%, 0.52%, 0.60%) and found that the 0.34% group had extremely significantly higher milk yield than the 0.52% group. Liu Zhen [13] fed Chinese Holstein dairy cows diets with different phosphorus levels (0.37%, 0.47%, 0.57%) and found that during mid-to-late lactation, the 0.37% group had significantly higher milk yield than the 0.47% and 0.57% groups. Carstairs et al. [14] reported that dry matter intake and milk yield of early-lactation dairy cows reached maximum values at dietary phosphorus levels of 0.40%-0.42% and could not be improved by higher phosphorus levels. Wu et

al. [15] found that during weeks 5-12 of lactation, cows fed 0.40% phosphorus had higher milk yield than those fed 0.52% phosphorus, and that 0.40% phosphorus was sufficient to maximize milk yield during the first 78 weeks of lactation, although cows experienced negative phosphorus balance while maintaining normal serum phosphorus levels. The present study showed that the 0.36% group had extremely significantly higher milk yield than the 0.26% and 0.46% groups, indicating that the 0.36% phosphorus level was optimal. This result is consistent with the NRC (2007) goat feeding standards. No significant difference in milk yield was observed between the 0.26% and 0.46% groups. Feeding diets with phosphorus levels recommended by NRC (2007) resulted in higher milk yield, which is importantly related to phosphorus homeostasis regulation in the body. When animals consume low-phosphorus diets, plasma phosphorus stimulates the synthesis of 1,25-dihydroxyvitamin D_3 to enhance phosphorus absorption; when animals consume excess phosphorus, the additional phosphorus is absorbed with a lower absorption coefficient. Changes in milk yield may also be related to the calcium-to-phosphorus ratio. Previous studies have not specifically investigated the effects of dietary phosphorus levels on milk yield in dairy goats, and the specific mechanisms require further investigation.

The feed-to-milk ratio is an important indicator for measuring production costs and economic benefits of Laoshan dairy goats, with lower ratios indicating higher feed efficiency. In this experiment, the 0.36% group had a lower feed-to-milk ratio than the 0.26% and 0.46% groups. This occurred because dietary phosphorus level had no significant effect on dry matter intake but significantly affected milk yield, with the 0.36% group having significantly higher milk yield than the other two groups. No significant difference in dry matter intake to milk yield ratio was observed between the 0.26% and 0.46% groups, likely because both low and high phosphorus levels were unfavorable for phosphorus absorption, resulting in no significant difference in milk yield between these two groups. When comparing 4% FCM yield (corrected for different milk fat percentages across groups), the 0.36% group was extremely significantly higher than the 0.46% group and significantly higher than the 0.26% group, while the 0.26% group was extremely significantly higher than the 0.46% group. This trend was basically consistent with the changes in milk yield among the three groups. Differences in dry matter intake to 4% FCM yield between days 1-30 and days 31-60 were mainly due to the significantly higher milk yield of the 0.36% group during days 31-60 compared to the 0.26% and 0.46% groups, while no significant differences in dry matter intake or milk fat percentage were observed among the three groups throughout the experimental period.

3.3 Effects of Dietary Phosphorus Level on Milk Composition of Laoshan Dairy Goats

Milk composition is an important indicator of dairy product quality and a key factor affecting the economic benefits of Laoshan dairy goats. Many factors influence milk composition in dairy cows, including breed, genetics, feeding

management, nutrient level, physiological stage, and health status. In this experiment, feeding Laoshan dairy goats diets with phosphorus levels of 0.26%, 0.36%, and 0.46% had no significant effects on milk fat percentage, milk protein percentage, lactose percentage, milk non-fat solids percentage, or milk calcium content. However, the 0.26% group showed a trend of lower milk calcium content compared to the 0.36% and 0.46% groups, likely due to the lower phosphorus level in this group, where calcium was consumed through endocrine regulation to maintain phosphorus balance. Milk phosphorus content showed an increasing trend with increasing dietary phosphorus levels, though differences among groups were not significant. These results indicate that dietary phosphorus levels in this experiment did not affect milk composition, possibly because nutrient levels and feeding management were basically identical across the three groups. Puggaard et al. [16] reported that diets with phosphorus levels of 0.24% and 0.34% had no significant effects on milk protein and milk fat percentages in dairy cows. Ge Xin et al. [5] found that feeding lactating dairy cows diets with phosphorus levels of 0.38% and 0.55% had no significant effects on milk fat and milk protein percentages. Similarly, Song Fancheng [11] found that feeding dairy cows diets with phosphorus levels of 0.27%-0.59% had no significant effects on milk fat percentage, milk protein percentage, lactose percentage, milk non-fat solids percentage, milk phosphorus content, or milk calcium content. These studies all yielded results consistent with our experiment.

3.4 Effects of Dietary Phosphorus Level on Serum Biochemical Indices of Laoshan Dairy Goats

Serum calcium and phosphorus contents and alkaline phosphatase activity are important indicators reflecting calcium and phosphorus metabolism in the body. Malmolf [17] suggested that serum urea nitrogen content can accurately reflect protein metabolism and amino acid balance status and is importantly related to production performance in dairy goats. Inadequate calcium and phosphorus supply or improper calcium-to-phosphorus ratios in dairy goat diets often cause abnormal bone structure, osteomalacia, reduced appetite, slow growth, and decreased production performance. Studies have also shown that when dairy cows consume low-phosphorus diets for extended periods, serum phosphorus levels decrease or even fall below normal values [18], which can cause increased ionized calcium in blood, thereby reducing parathyroid hormone secretion and decreasing renal phosphorus excretion. Additionally, it can lead to increased 1,25-dihydroxyvitamin D_3 levels, promoting renal phosphorus reabsorption and intestinal phosphorus absorption from digesta [19]. Therefore, calcium and phosphorus metabolism in ruminants is closely related to their health and production.

Puggaard et al. [20] reported that when dietary phosphorus levels met the requirements of dairy cows, serum biochemical indices were not affected by changes in dietary phosphorus levels; conversely, if requirements were not met, serum biochemical indices would show significant differences and phosphorus

deficiency symptoms would appear. Lu Dexun [21] found that normal values for serum calcium, phosphorus, alkaline phosphatase activity, and urea nitrogen content in sheep were 2.12-2.87 mmol/L, 1.13-2.58 mmol/L, 12-120 U/L, and 2.9-7.1 mmol/L, respectively. In this experiment, serum calcium contents in the 0.26%, 0.36%, and 0.46% groups were 2.17, 2.16, and 2.14 mmol/L, respectively; serum phosphorus contents were 1.82, 2.13, and 2.00 mmol/L; alkaline phosphatase activities were 58.20, 62.60, and 59.80 U/L; and urea nitrogen contents were 6.16, 6.27, and 6.01 mmol/L. All values were within the normal ranges for sheep, with no significant differences among groups. Wang Jianhua et al. [22] fed Laoshan dairy goats diets with phosphorus levels of 0.29%, 0.41%, and 0.59% and found no significant effects on serum phosphorus content, a conclusion similar to that of Lopez et al. [18] and consistent with our results. Call et al. [23] fed dairy cows diets with phosphorus levels of 0.24%, 0.32%, and 0.42% and found no significant differences in serum calcium content among groups. Wu et al. [24] found that feeding dairy cows diets with phosphorus levels of 0.38% and 0.48% did not significantly affect serum calcium content, consistent with our findings. Sun Guoqiang et al. [25] reported that dietary phosphorus level had no significant effect on alkaline phosphatase activity in dairy cows, consistent with our results. This experiment also showed that dietary phosphorus level had no significant effect on serum urea nitrogen content in Laoshan dairy goats. Ge Xin et al. [26] studied the effects of dietary phosphorus level on nitrogen digestion and metabolism in Laoshan dairy goats and found that different phosphorus levels had no significant effect on plasma urea nitrogen content, consistent with our results.

Conclusion

Dietary phosphorus level can significantly affect milk yield and feed-to-milk ratio in Laoshan dairy goats, but has no significant effects on dry matter intake, milk composition, or serum biochemical indices. Based on comprehensive analysis, under the conditions of this experiment, a dietary phosphorus level of 0.36% is recommended for lactating Laoshan dairy goats.

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