

Effects of *Saccharomyces cerevisiae* on Nutrient Digestibility, Rumen Fermentation, and Plasma Biochemical Indices in Xiangzhong Black Cattle (Postprint)

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Abstract

This experiment aimed to investigate the effects of different supplementation levels of *Saccharomyces cerevisiae* on growth performance, nutrient digestibility, rumen fermentation, and plasma biochemical indices in Xiangzhong Black cattle. Eighteen Xiangzhong Black cattle [average body weight (330±\$30) kg] were selected and divided into 3 groups using a single-factor completely randomized design, with 6 cattle per group; the control group was fed a basal diet, while the experimental groups were supplemented with 120 (Group I) and 240 g/kg *Saccharomyces cerevisiae* (Group II) in the basal diet, respectively. The preliminary period was 10 d, and the formal experimental period was 30 d. The results showed that: 1) Dietary supplementation of *Saccharomyces cerevisiae* had no significant effect on average daily gain and feed-to-gain ratio in Xiangzhong Black cattle ($P>0.05$). 2) Dietary supplementation of *Saccharomyces cerevisiae* significantly reduced crude protein fecal excretion ($P<0.05$), but had no significant effect on digestibility of dry matter, neutral detergent fiber, and crude protein ($P>0.05$). 3) There were no significant differences in concentrations of ammonia nitrogen (NH₃-N) and volatile fatty acids (VFA) in rumen fluid among all groups ($P>0.05$). 4) Dietary supplementation of 240 g/kg *Saccharomyces cerevisiae* significantly reduced plasma total cholesterol concentration in Xiangzhong Black cattle ($P<0.05$). These results suggest that dietary supplementation of 240 g/kg *Saccharomyces cerevisiae* can reduce crude protein fecal excretion and plasma total cholesterol concentration in Xiangzhong Black cattle, which is of great significance for reducing nitrogen emission, improving farming environment, and promoting healthy cattle farming.

Full Text

Effects of *Saccharomyces cerevisiae* on Nutrient Digestibility, Rumen Fermentation and Plasma Biochemical Parameters of Xiangzhong Black Beef Cattle

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Abstract

This study investigated the effects of different supplementation levels of *Saccharomyces cerevisiae* on growth performance, nutrient digestibility, rumen fermentation, and plasma biochemical parameters of Xiangzhong black beef cattle. Eighteen Xiangzhong black beef cattle [average body weight (330±30)kg] were randomly allocated to three groups using a single-factor completely randomized design, with six cattle per group. The control group received a basal diet, while the experimental groups received diets supplemented with 0, 120, or 240 g/kg *S. cerevisiae*. The pre-trial period lasted 10 days, followed by a 30-day formal experimental period. The results showed that (1) dietary supplementation with *S. cerevisiae* had no significant effect on averaged daily gain or feed-to-gain ratio ($P > 0.05$); (2) *S. cerevisiae* supplementation significantly reduced fecal crude protein excretion ($P < 0.05$), but had no significant effect on digestibility of dry matter, neutral detergent fiber, or crude protein ($P > 0.05$); (3) concentrations of ammoniacal nitrogen (NH_3 -N) and volatile fatty acids (VFA) in rumen fluid were not significantly different among groups ($P > 0.05$); and (4) supplementation with 240 g/kg *S. cerevisiae* significantly decreased plasma total cholesterol concentration ($P < 0.05$). These findings suggest that dietary supplementation with 240 g/kg *S. cerevisiae* can reduce crude protein fecal excretion and plasma total cholesterol concentration in Xiangzhong black beef cattle, which is important for reducing nitrogen emissions, improving the farming environment, and promoting healthy beef cattle production.

Keywords: Xiangzhong black beef cattle; total cholesterol; volatile fatty acids; feed-to-gain ratio; growth performance

Introduction

As a novel microbial additive, probiotics are live bacterial preparations that exert beneficial effects on the host, also known as microecological preparations. These mainly include lactic acid bacteria preparations, *Bacillus* preparations, yeast preparations, and mixed bacterial preparations [1]. Yeast has been applied as a valuable animal feed additive for many years, with *Saccharomyces cerevisiae* being the most commonly used species in feed additives, comprising two major types: live yeast preparations and yeast culture preparations [2]. *S. cerevisiae* has been widely used to improve milk production performance in dairy cows, maintain cow health, and promote calf development [3]. In beef cattle production, *S. cerevisiae* has also been extensively studied. Geng [4] reported that under high-concentrate dietary conditions, *S. cerevisiae* supplementation improved the growth performance of finishing cattle. Zhang [5] found that dietary supplementation with 0.1% active dry yeast maintained the rumen anaerobic environment and positively affected rumen fermentation while promoting the growth of rumen fiber-degrading bacteria and lactate-utilizing bacteria. Ding [6] reported that yeast supplementation had no significant effect on rumen fermentation patterns in beef cattle but improved lipid and phospholipid metabolism, which was beneficial for enhancing growth performance. Jing et al. [7] demonstrated that under high-concentrate dietary conditions, active dry yeast supplementation significantly increased the rapidly degradable fraction and effective degradation rate of neutral detergent fiber (NDF) and acid detergent fiber (ADF) in the rumen of beef cattle. However, few studies have reported the effects of *S. cerevisiae* supplementation on growth performance, nutrient digestibility, rumen fermentation, and plasma biochemical parameters in Xiangzhong black beef cattle. Therefore, this experiment used Xiangzhong black beef cattle as experimental animals to investigate the effects of different supplementation levels of *S. cerevisiae* on these parameters, aiming to provide data support for healthy beef cattle production in practical farming operations.

1.1 Experimental Material

The *S. cerevisiae* used in this experiment was an active yeast product manufactured by Lesaffre Group (France), with a viable count of 1.5×10^{10} CFU/g.

1.2 Experimental Design

This experiment employed a single-factor completely randomized design. Eighteen Xiangzhong black beef cattle with an average body weight of (330 ± 30) kg were randomly divided into three groups, with six cattle per group. The

control group was fed a basal diet, while the experimental groups received the basal diet supplemented with 120 g/kg (Group I) and 240 g/kg (Group II) of *S. cerevisiae*. The pre-trial period lasted 10 days, followed by a 30-day formal experimental period.

1.3 Experimental Diets and Management

The basal diet for Xiangzhong black beef cattle was formulated according to NRC (2001) and consisted of rice straw and concentrate, with a concentrate-to-roughage ratio of 55:45. The composition and nutrient levels of the basal diet are presented in Table 1. Experimental cattle were housed individually in tie-stalls with *ad libitum* access to feed and water throughout the trial period. The ambient temperature was maintained at approximately 25 °C with good ventilation. Manure was cleaned twice daily, and pens were disinfected 2-3 times per week. Cattle were managed according to routine farm practices for immunization and deworming.

Table 1 Composition and nutrient levels of the basal diet (dry matter basis), %

Item	Content
Ingredients	
Rice straw	
Rapeseed meal	
Distiller' s grains	
Corn	
Soybean meal	
Wheat bran	
Calcium carbonate	
Premix	
Salt	
Sodium bicarbonate	
Total	
Nutrient levels	
Crude protein	
Neutral detergent fiber	
Calcium	
Metabolic energy (MJ/kg)	

Note: Each kilogram of premix contained: Mg 50 g, Fe 2.5 g, Cu 0.4 g, Mn 2 g, Zn 1.5 g, Se 10 mg, I 25 mg, Co 5 mg, VA 80,000 IU, VD 12,500 IU, VE 1,250 mg.

1.4 Measurements

1.4.1 Growth Performance Body weight was measured on an empty stomach on days 1 and 30 of the formal experimental period to record weight changes and calculate average daily gain. Feed intake was recorded from days 20–25 of the formal period, and average daily feed intake was calculated based on feed offered and refusals. The feed-to-gain ratio was then calculated from average daily gain and average daily feed intake using the following formulas:

Average daily gain = Total weight gain / (Number of animals × Experimental days)

Average daily feed intake = Total feed intake / (Number of animals × Experimental days)

Feed-to-gain ratio = Average daily feed intake / Average daily gain

1.4.2 Nutrient Digestibility Feces were collected from each group during days 25–30 of the formal period using the total collection method. Fecal samples were weighed, and 10% of the total weight was sampled. For every 100 g of sample, 20 mL of 10% H₂SO₄ was added, and samples were stored at -20 °C for subsequent analysis of dry matter (DM), neutral detergent fiber (NDF), and crude protein (CP) content. Fecal samples from each animal collected over six days were pooled, and DM, NDF, and CP contents were determined using the feed analysis methods described by Yang [8]. Nutrient digestibility was calculated as follows:

DM digestibility = $100 \times (\text{Total DM intake} - \text{Fecal DM}) / \text{Total DM intake}$

NDF digestibility = $100 \times (\text{Total NDF intake} - \text{Fecal NDF}) / \text{Total NDF intake}$

CP digestibility = $100 \times (\text{Total CP intake} - \text{Fecal CP}) / \text{Total CP intake}$

1.4.3 Plasma Biochemical Parameters Blood samples were collected from the tail vein on day 30 of the formal period at three time points: pre-feeding (0 h) and 2 h and 6 h post-feeding. Heparinized vacuum tubes were used for blood collection. Blood samples were immediately centrifuged (5,000 r/min, 4 °C, 10 min) to harvest plasma, which was stored at -20 °C. Plasma alanine aminotransferase (ALT), lactate dehydrogenase (LDH) activities, and concentrations of glucose (Glu), total cholesterol (TC), total protein (TP), triglycerides (TG), and urea nitrogen (UN) were measured using an automatic biochemical analyzer (Beckman Coulter CX4, USA) and commercial kits (Nanjing Jiancheng Bioengineering Institute).

1.4.4 Rumen Fermentation Parameters Rumen fluid was collected orally on day 30 of the formal period at two time points: 2 h and 6 h post-feeding. Collected rumen fluid was placed in 10 mL centrifuge tubes and stored at -20 °C for NH₃-N concentration determination. The remaining rumen fluid was centrifuged at 10,000 r/min for 15 min at 4 °C. The supernatant (1.5 mL) was mixed with 0.15 mL of 25% metaphosphoric acid for fixation and stored at -20 °C for volatile fatty acid (VFA) concentration determination. Concentrations

of VFA and $\text{NH}_3\text{-N}$ in rumen fluid were determined according to the methods described by Chen et al. [9].

1.5 Statistical Analysis

Data were analyzed using the MIXED procedure of SAS 9.1 software. Statistical significance was defined as $P < 0.05$.

2.1 Effects of Different Supplementation Levels of *S. cerevisiae* on Growth Performance of Xiangzhong Black Beef Cattle

As shown in Table 2, there were no significant differences in average daily gain or feed-to-gain ratio among the three groups ($P > 0.05$).

Table 2 Effects of different supplementation levels of *Saccharomyces cerevisiae* on growth performance of Xiangzhong black beef cattle

Item	Control group	Group I	Group II	P-value
Average daily gain (kg)				
Feed-to-gain ratio				

2.2 Effects of Different Supplementation Levels of *S. cerevisiae* on Nutrient Digestibility of Xiangzhong Black Beef Cattle

As shown in Table 3, compared with the control group, both experimental groups significantly reduced fecal crude protein excretion ($P < 0.05$), with Group I and Group II decreasing by 19.35% and 16.13%, respectively. Crude protein digestibility in Groups I and II was 6.92% and 6.22% higher than in the control group, respectively, but the difference among the three groups was not significant ($P > 0.05$). Different supplementation levels of *S. cerevisiae* had no significant effects on dry matter or neutral detergent fiber digestibility ($P > 0.05$).

Table 3 Effects of different supplementation levels of *Saccharomyces cerevisiae* on nutrient digestibility of Xiangzhong black beef cattle

Item	Control group	Group I	Group II	P-value
Dry matter				
Intake (kg/d)				
Fecal excretion (kg/d)				
Digestibility (%)				
Crude protein				
Intake (kg/d)				
Fecal excretion (kg/d)	0.31a	0.25b	0.26b	
Digestibility (%)				
Neutral detergent fiber				

Item	Control group	Group I	Group II	P-value
Intake (kg/d)				
Fecal excretion (kg/d)				
Digestibility (%)				

2.3 Effects of Different Supplementation Levels of *S. cerevisiae* on Rumen Fermentation Parameters of Xiangzhong Black Beef Cattle

As shown in Table 4, ruminal NH₃-N concentration increased with increasing *S. cerevisiae* supplementation level, but the difference among groups was not significant ($P>0.05$). In Group I, concentrations of acetate, propionate, valerate, butyrate, and total VFA were 3.43%, 1.45%, 8.22%, 23.10%, and 4.95% higher than in the control group, respectively. Rumen VFA concentrations in Group I were higher than in Group II, with acetate, propionate, valerate, isovalerate, butyrate, isobutyrate, and total VFA being 8.37%, 9.67%, 9.72%, 7.35%, 15.24%, 6.19%, and 9.31% higher, respectively. Different supplementation levels of *S. cerevisiae* had no significant effects on VFA concentrations ($P>0.05$).

Table 4 Effects of different supplementation levels of *Saccharomyces cerevisiae* on rumen fermentation parameters of Xiangzhong black beef cattle

Item	Control group	Group I	Group II	P-value
NH ₃ -N (mg/dL)				
Acetate (mmol/L)				
Propionate (mmol/L)				
Valerate (mmol/L)				
Isovalerate (mmol/L)				
Butyrate (mmol/L)				
Isobutyrate (mmol/L)				
Acetate/propionate				
Total VFA (mmol/L)				

2.4 Effects of Different Supplementation Levels of *S. cerevisiae* on Plasma Biochemical Parameters of Xiangzhong Black Beef Cattle

As shown in Table 5, there were no significant differences among groups in plasma ALT and LDH activities or concentrations of Glu, TG, TP, and UN ($P>0.05$). Plasma total cholesterol concentration in Group II was 15.61% and 28.11% lower than in the control group and Group I, respectively, with significant differences ($P<0.05$).

Table 5 Effects of different supplementation levels of *Saccharomyces cerevisiae* on plasma biochemical parameters of Xiangzhong black beef cattle

Item	Control group	Group I	Group II	P-value
ALT (U/L)				
Glu (mmol/L)				
LDH (U/L)				
TC (mmol/L)	4.02a	3.46b	2.92c	
TG (mmol/L)				
TP (g/L)				
UN (mmol/L)				

3.1 Effects of Different Supplementation Levels of *S. cerevisiae* on Growth Performance of Xiangzhong Black Beef Cattle

Geng [4] reported that yeast culture had no significant effect on the growth performance of finishing cattle, and active dry yeast had no significant effect on feed-to-gain ratio. Dong et al. [10] also found that probiotics had no significant effects on growth performance indices such as average daily gain, average daily feed intake, or feed-to-gain ratio in suckling calves. These findings are consistent with the results of the present study. Sanchez et al. [11] reported that dietary supplementation with probiotics [*Propionibacterium acidipropionici* P1691] had no significant effect on body weight in beef cattle.

However, other studies have reported positive effects of yeast culture on calf growth. Francia et al. [12] indicated that dietary *S. cerevisiae* supplementation had positive effects on increasing average daily gain and feed conversion ratio in calves. Fu et al. [13] reported that dietary supplementation with *Bacillus licheniformis* alone increased average daily gain and body indices in calves (0–8 weeks of age), while composite probiotics (*B. licheniformis*, *Bacillus subtilis*, and *Lactobacillus plantarum*) increased body indices at 8 weeks of age. Chou et al. [14] found that dietary supplementation with probiotics (*Lactobacillus* and *Bifidobacterium*) significantly increased average daily gain in calves. Differences among studies may be primarily attributed to variations in probiotic species, supplementation levels, and experimental animals, as well as differences in dietary composition and cattle age.

3.2 Effects of Different Supplementation Levels of *S. cerevisiae* on Nutrient Digestibility of Xiangzhong Black Beef Cattle

Vyas et al. [15] reported that probiotic supplementation (*Propionibacterium*) in beef cattle diets had no significant effects on ruminal digestibility of dry matter (DM), organic matter (OM), NDF, acid detergent fiber (ADF), or starch. Sanchez et al. [11] also found that probiotic supplementation (*Propionibacterium acidipropionici* P1691) in low-quality forage diets for beef cattle had no significant effects on OM or NDF digestibility and did not significantly alter body weight. Wang et al. [16] reported that probiotic supplementation (*S. cerevisiae*) had no significant effects on in vitro DM and NDF digestibility of corn straw.

These results are similar to those of the present study. However, other studies have reported that dietary yeast supplementation could improve fiber and DM digestibility [17,18]. These discrepancies may be due to differences in dietary composition. In the current study, *S. cerevisiae* supplementation significantly reduced fecal crude protein excretion and improved crude protein digestibility, possibly by promoting rumen microbial degradation of dietary protein. This is indirectly supported by the ruminal $\text{NH}_3\text{-N}$ concentrations observed across groups. Liu et al. [19] reported that dietary supplementation with composite probiotics significantly increased ruminal $\text{NH}_3\text{-N}$ concentration, consistent with the present results.

3.3 Effects of Different Supplementation Levels of *S. cerevisiae* on Rumen Fermentation Parameters of Xiangzhong Black Beef Cattle

$\text{NH}_3\text{-N}$ is the primary precursor for microbial protein synthesis in the rumen, mainly generated from the degradation of dietary protein by rumen microorganisms. In this study, *S. cerevisiae* supplementation promoted protein degradation in the rumen, resulting in higher ruminal $\text{NH}_3\text{-N}$ concentrations in the experimental groups compared to the control group, which aligns with the findings of Liu et al. [19]. Volatile fatty acids in the rumen primarily originate from carbohydrate fermentation and serve as an important energy source for ruminants while providing energy for microbial synthesis. The lack of significant effect of *S. cerevisiae* supplementation on ruminal VFA concentrations in this study may be due to minimal impact on carbohydrate-degrading bacterial populations. Liu et al. [19] reported that dietary supplementation with composite probiotics had no significant effects on propionate, butyrate concentrations, or acetate/propionate ratio, but significantly increased total VFA and acetate concentrations. Other studies have reported that dietary supplementation with active yeast or yeast culture could alter ruminal VFA concentrations and proportions [20–22], while some found that *Propionibacterium* supplementation significantly modified ruminal VFA proportions [11,15]. Peng et al. [23] reported that dietary supplementation with *Bacillus subtilis* natto in dairy cows significantly increased ruminal propionate concentration while decreasing acetate concentration. These discrepancies may be attributed to differences in probiotic species and supplementation levels, as well as dietary chemical composition.

3.4 Effects of Different Supplementation Levels of *S. cerevisiae* on Plasma Biochemical Parameters of Xiangzhong Black Beef Cattle

Plasma biochemical parameters can reflect animal growth performance to some extent and are important indicators in animal production research. In this study, except for plasma total cholesterol concentration, *S. cerevisiae* supplementation had no significant effects on other plasma biochemical parameters. Wang et al. [24] reported that dietary supplementation with composite yeast culture significantly increased serum Glu concentration and significantly decreased serum UN concentration. Fu et al. [13] found that different probiotic

combinations had no significant effects on serum biochemical parameters in calves. Yan [25] reported that yeast culture supplementation had no significant effect on blood TP concentration in calves. Peng et al. [23] also found that *B. subtilis* natto supplementation had no significant effects on blood biochemical parameters in dairy cows. These results suggest that the effects of probiotic supplementation on blood biochemical parameters may be related to animal species, growth stage, probiotic composition, and dietary factors. Total cholesterol is a major component of blood lipids, and its concentration can reflect lipid absorption and metabolism status [26]. Numerous studies have demonstrated that probiotics can reduce body cholesterol [27]. Interestingly, in the present study, low-level probiotic supplementation significantly increased plasma TC concentration, possibly due to enhanced lipid metabolism, as Ding [6] reported that yeast supplementation positively affected animal lipid and phospholipid metabolism. In contrast, high-level *S. cerevisiae* supplementation significantly decreased plasma TC concentration, which may be because high levels of probiotics can absorb or adsorb intestinal cholesterol, thereby reducing plasma TC concentration [28,29].

Conclusions

1. Dietary supplementation with *S. cerevisiae* significantly reduced crude protein fecal excretion in Xiangzhong black beef cattle, which is important for reducing nitrogen emissions, improving the farming environment, and decreasing environmental pollution during beef cattle production.
2. Dietary supplementation with 120 g/kg *S. cerevisiae* significantly increased plasma total cholesterol concentration, while supplementation with 240 g/kg *S. cerevisiae* significantly decreased plasma total cholesterol concentration in Xiangzhong black beef cattle, suggesting positive effects on meat quality improvement and healthy cattle production.

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