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Effect of enzyme and probiotic supplementation on growth performance, nutrient digestibility, carcass traits, and meat quality of Simmental steers

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ABSTRACT - The objective of this trial was to investigate the potential benefits of enzyme and probiotic supplementation on the growth performance, nutrient digestibility, carcass traits, and meat quality of Simmental cattle. Sixty Simmental steers (367.75±4.69 kg) were randomly divided into three groups: control (fed basal diet), BC1 group (fed basal diet and 10 g/d brewer's yeast and cellulase supplementation per cattle), and BC2 group (fed basal diet and 20 g/d brewer's yeast and cellulase supplementation per cattle). A 10-d preparation period was followed by a 120-d experimental period. The results showed that the final weight of the BC2 group was greater than that of the control group. Also, feed conversion ratio of the BC2 group was better than that of the control group. Net meat weight increased by 3.86% in the BC2 group compared with the control group. The apparent digestibility of neutral detergent fiber, acid detergent fiber, and crude protein of the BC2 group was greater than that of the control group. The apparent digestibility of neutral detergent fiber, acid detergent fiber, and crude protein of the BC2 group was greater than that of the control group. Dietary supplementation with brewer's yeast and cellulase supplementation could improve the growth performance and nutrient digestibility of Simmental cattle.

Keywords: additives, beef cattle, brewer's yeast, cellulase, meat yield

1. Introduction

Growth performance and meat production are the key to the economic benefits of the beef cattle industry (Romanzini et al., 2020). Improving the absorption and utilization of feed nutrients in beef cattle is an important factor to enhance their production performance. Numerous previous studies have shown that the use of antibiotic feed additives had a significant effect on improved performance of beef cattle (Kamphues, 1999; Agga et al., 2016; Cazer et al., 2020). However, with China banning the use of antibiotics in feed, the research and development of natural feed additives have received more and more attention in cattle industry (Wierup, 2001; Millet and Maertens, 2011). Probiotics have been more widely used in beef cattle feed. Brewer's yeast, derived from Saccharomyces cerevisiae, is extensively used as a feed additive in cattle diets (Uyeno et al., 2015; Barreto et al., 2021). Seymour (1995) found that the brewer's yeast is rich in viable yeast and has positive effects on the growth performance of beef cattle. Brewer's yeast belongs to facultative anaerobe and can consume oxygen, which is beneficial for maintaining an anaerobic environment in the rumen, thus limiting the growth and reproduction of pathogenic bacteria and offering a favorable fermentation environment (Alugongo et al., 2017). Furthermore, brewer's yeast can promote the conversion of lactate into propionate so that cattle can obtain more energy from their feeds and improve the growth performance of cattle (Fomenky et al., 2018).

In the cattle industry, cellulase has been confirmed to be an effective feed additive that can improve the digestibility of crude fiber (Schroeder et al., 2014; Zhu et al., 2018). Early studies indicated that dietary cellulase supplementation was beneficial for nutrient digestibility, rumen fermentation (He et al., 2015), and meat yield (Maeda et al., 2019). Some other studies suggested that cellulase supplementation could increase the dry matter intake and average daily gain of beef cattle (So et al., 2022). All in all, the addition of either probiotics or cellulase alone in the feeds has a positive effect on the productive performance of cattle. However, the studies on the combined application of probiotics and cellulases in beef cattle feeds are less common. Whether there is a synergistic effect between probiotics and cellulases is a question worth investigating. In this study, we mixed brewer's yeast and cellulase into Simmental cattle feed to investigated the effect of the additive mixture on the growth performance, nutrient digestibility, carcass traits, and meat quality of Simmental cattle.

2. Material and Methods

2.1. Animals, experimental design, and dietary treatments

This study was conducted in Urumqi, Xinjiang, China (86°37' N, 42°45' W). The research on animals was conducted according to the Institutional Committee on Animal Use (case number 2019006).

A total of 60 Simmental steers (body weight [BW] = 367.75 ± 4.69 kg, age = 420 ± 20 d) were used in a single factor design. The selected Simmental cattle were marked with ear tags and randomly assigned to three test groups: control (fed basal diet), BC1 (fed basal diet and 10 g/d brewer's yeast and cellulase supplementation per cattle), and BC2 (fed basal diet and 20 g/d brewer's yeast and cellulase supplementation per cattle). Cattle in the three groups were assigned to identical research pens (2.50 × 4.00 m), each group had 10 replicates with two cattle in each replicate (10 pens/group, two cattle/pen). The basal diet was formulated according to the Chinese Feeding Standard of Beef Cattle (NY/T 815-2004) (Table 1). All the cattle were regularly provided rations two times a day at 08:00 and 18:00 h and had free access to water. A 10-d preparation period was followed by a 120-d experimental period, and the trial period was divided into early (1-60 d) and late (61-120 d) phases. Bacteria and

| Itom | Farly poriod | Late period |
|------------------------------|--------------|-------------|
| | Early period | Late period |
| Ingredient | | |
| Corn silage | 41.50 | 31.50 |
| Distillers grains | 12.50 | 12.00 |
| Corn | 35.80 | 45.50 |
| Wheat bran | 1.73 | 3.12 |
| Soybean meal | 3.00 | 0 |
| Cotton meal | 4.59 | 6.95 |
| NaHCO ₃ | 0.35 | 0.38 |
| Stone powder | 0.09 | 0.09 |
| NaCl | 0.22 | 0.22 |
| Premix ¹ | 0.22 | 0.24 |
| Total | 100.00 | 100.00 |
| Nutrient levels ² | | |
| NE _{mf} (MJ/kg) | 6.33 | 6.74 |
| Crude protein | 12.35 | 11.76 |
| Neutral detergent fiber | 34.03 | 28.91 |
| Acid detergent fiber | 19.53 | 15.34 |
| Са | 0.61 | 0.67 |
| Total P | 0.33 | 0.38 |

Table 1 - Composition and nutrient levels of basal diets (%; dry matter basis)

¹ The premix provided the following per kg of diets: vitamin A, 4500 IU; vitamin D3, 780 IU; vitamin E, 45 IU; Fe, 60 mg; Cu, 60 mg; Zn, 50 mg; Mn,30 mg; I, 0.5 mg; Co, 0.4 mg; Se, 0.1 mg; Ca, 10-20%.

² NE_{mf} was calculated according to Feeding Standard of Beef Cattle of China (NY/T 815-2004); while the other nutrient levels were measured values.

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enzymes supplementation containing brewer's yeast (viable yeast $\geq 5 \times 10^9$ cfu/g) and cellulase (enzyme activity ≥ 500 U/g) were provided by Henan Kangxing Pharmaceutical Co. (Henan, China).

2.2. Performance criteria

The BW of all steers was measured on days 0 and 120 before morning feeding, and average daily gain (ADG) was calculated from initial and final BW. Accurate feed intake of each steer was recorded daily (average daily feed intake, ADFI) and converted into feed conversion ratio (FCR).

2.3. Determination of apparent nutrient digestibility

Three days before the end of the experiment, 10 cattle in each group were randomly selected, and fecal samples were collected in their rectum. About 400 g of fecal samples were collected per cattle per day, and half the fecal samples were directly sealed and stored, and the other half was added to 10% sulfuric acid for nitrogen fixation (Maeda et al., 2019). Twenty milliliters of sulfuric acid were added per 100 g of fecal sample, and stored frozen. Meanwhile, the diets of the experimental cattle were collected and stored frozen. Fecal samples and diet samples were dried and crushed at the end of the experiment. The crude protein (CP), ether extract (EE), calcium (Ca), total phosphorus (TP), neutral detergent fiber (NDF), and acid detergent fiber (ADF) of the faeces and feed were analysed according to national standards GB/T 6432-2018, GB/T 6433-2006, GB/T 6436-2002, GB/T 6437-2002, GB/T 20806-2006, and NY/T 1459-2007, respectively. Apparent digestibility of nutrients was calculated according to national standard GB/T 23742-2009.

2.4. Carcass traits and meat quality

According to the average weight, 10 cattle in each group were selected for slaughtering trial and meat quality determination. The cattle were transported to commercial abattoir, where they were held in lairage overnight and slaughtered as a single group the following day. Muscle samples (thickness: 2.00 cm, weight: 200 g) were collected from each carcass in the region of the ninth and tenth rib of the *longissimus thoracis*, trimmed away visible fat. Muscle pH was determined by acidity meter (UB-7, USA), and cooking loss, water loss rate, and shear force were determined with reference to the industry standard of the Ministry of Agriculture (Refer to the state standard NY/T 1180-2006). Meat color L*, a*, and b* were determined using CR-400 Minolta colorimeter with illuminant D65, 10° observer and 2.54 cm diameter aperture (Konica Minolta Sensing Americas Inc., USA).

2.5. Statistical analysis

All data were analyzed according to a one-way experimental design. For the growth performance traits and the other parameters, data were analyzed as a complete randomized group design using the general linear model (GLM) procedure of SPSS 20.0 software (SPSS Inc.). The statistical model was as follows:

$$Y_{ij} = \mu + \beta_i + \varepsilon_{ij}$$

in which Y_{ij} = dependent variable, μ = variable mean, β_i = fixed effect of i-th cattle of the treatment, and ϵ_{ij} = experimental error associated with observation Y_{ij} . Significance was declared when P<0.05, results were presented as means and standard error of the mean (SEM).

3. Results

3.1. Feed intake and growth performance

The use of enzyme and probiotic supplementation (20 g/d) had a positive effect on the growth performance of Simmental cattle, including feed intake and daily gain (Table 2). The initial weight,

ADG, and ADFI did not show a significant difference among groups (P>0.05). The final weight of the BC2 group was greater than that of the control group (P = 0.031). The FCR in the BC2 group was better than that of the control group (P = 0.027).

| ltem | Group ² | | | CEM | Darahaa |
|---------------------|--------------------|---------|---------|------|---------|
| | Control | BC1 | BC2 | SEM | r-value |
| Initial weight (kg) | 367.23 | 367.25 | 368.77 | 2.17 | 0.357 |
| Final weight (kg) | 529.11b | 530.27b | 549.54a | 4.12 | 0.031 |
| ADG (kg/d) | 1.34 | 1.36 | 1.49 | 0.12 | 0.089 |
| ADFI (kg/d) | 10.29 | 10.34 | 10.48 | 0.54 | 0.258 |
| FCR | 7.66a | 7.63a | 7.11b | 0.31 | 0.027 |
| | | | | | |

| Table 2 - Effect of | enzyme and | probiotic supplementation | on on growth | performance of Si | mmental cattle ¹ |
|---------------------|------------|---------------------------|--------------|-------------------|-----------------------------|
| | | | | | |

ADG - average daily gain; ADFI - average daily feed intake; FCR - feed conversion ratio; SEM - standard error of the mean.

¹ Data are given as treatment means.

² Control group fed basal diets; BC1 group fed basal diet with brewer's yeast and cellulase supplementation (10 g per cattle per day); BC2 group fed basal diet with brewer's yeast and cellulase supplementation (20 g per cattle per day).

a,b - Means within a row with different letters are significantly different (P<0.05).

3.2. Carcass traits

The dressing percentage, carcass meat yield, bone weight, meat to bone ratio, backfat thickness, and eye area did not show an obvious difference among groups (P>0.05) (Table 3). The net meat weight increased by 3.86% in the BC2 group compared with the control group, but there was no significant difference (P = 0.085).

| Item | Group ² | | | CEM | D .l . |
|-----------------------------|--------------------|--------|--------|------|---------|
| | Control | BC1 | BC2 | SEM | P-value |
| Net meat weight (kg) | 255.48 | 256.98 | 265.33 | 4.10 | 0.085 |
| Dressing percentage (%) | 56.55 | 56.48 | 57.03 | 2.10 | 0.201 |
| Carcass meat yield (%) | 83.25 | 83.46 | 84.14 | 2.73 | 0.547 |
| Bone weight (kg) | 48.56 | 48.72 | 49.23 | 1.05 | 0.309 |
| Meat to bone ratio | 5.31 | 5.32 | 5.33 | 0.33 | 0.120 |
| Backfat thickness (cm) | 0.23 | 0.23 | 0.24 | 0.01 | 0.245 |
| Eye area (cm ²) | 88.15 | 88.22 | 89.87 | 2.03 | 0.204 |

Table 3 - Effect of enzyme and probiotic supplementation on carcass traits of Simmental cattle¹

SEM - standard error of the mean.

¹ Data are given as treatment means.

² Control group fed basal diets; BC1 group fed basal diet with brewer's yeast and cellulase supplementation (10 g per cattle per day); BC2 group fed basal diet with brewer's yeast and cellulase supplementation (20 g per cattle per day).

3.3. Meat quality

The pH, cooking loss rate, water loss rate, shear force, and meat color did not show an obvious difference among groups (P>0.05) (Table 4).

3.4. Apparent digestibility of nutrients

The use of enzyme and probiotic supplementation had a positive effect on the apparent digestibility of nutrients of Simmental cattle (Table 5). The apparent digestibility of NDF, ADF, and CP of the BC2 group was greater than that of the control group (P<0.05). The apparent digestibility of calcium and phosphorus did not show an obvious difference among groups (P>0.05).

| Item | Group ² | | | 0714 | |
|-----------------------|--------------------|-------|-------|------|---------|
| | Control | BC1 | BC2 | SEM | P-value |
| рН | 5.65 | 5.63 | 5.49 | 0.18 | 0.330 |
| Cooking loss rate (%) | 18.88 | 18.83 | 19.12 | 1.45 | 0.201 |
| Water loss rate (%) | 30.56 | 30.12 | 29.84 | 2.21 | 0.103 |
| Shear force (kg) | 4.98 | 5.02 | 4.88 | 0.21 | 0.651 |
| Meat color | | | | | |
| L* | 32.55 | 32.15 | 31.79 | 0.32 | 0.246 |
| a* | 14.56 | 15.21 | 14.35 | 0.16 | 0.284 |
| b* | 4.23 | 4.55 | 4.11 | 0.11 | 0.251 |

Table 4 - Effect of enzyme and probiotic supplementation on meat quality of Simmental cattle¹

SEM - standard error of the mean.

¹ Data are given as treatment means.

² Control group fed basal diets; BC1 group fed basal diet with brewer's yeast and cellulase supplementation (10 g per cattle per day); BC2 group fed basal diet with brewer's yeast and cellulase supplementation (20 g per cattle per day).

| Item | | Group ² | | | Develue |
|------|---------|--------------------|--------|------|---------|
| | Control | BC1 | BC2 | SEM | P-value |
| NDF | 55.45b | 56.59b | 62.35a | 1.21 | 0.032 |
| ADF | 48.23b | 48.38b | 53.26a | 1.52 | 0.043 |
| СР | 65.54b | 66.32b | 72.25a | 1.89 | 0.028 |
| Са | 43.56 | 44.51 | 44.21 | 2.11 | 0.268 |
| P | 48.56 | 49.28 | 49.22 | 1.77 | 0.158 |

Table 5 - Effect of enzyme and probiotic supplementation on apparent digestibility of nutrients in Simmental cattle¹

NDF - neutral detergent fiber; ADF - acid detergent fiber; CP - crude protein; SEM - standard error of the mean.

¹ Data are given as treatment means.

² Control group fed basal diets; BC1 group fed basal diet with brewer's yeast and cellulase supplementation (10 g per cattle per day); BC2 group fed basal diet with brewer's yeast and cellulase supplementation (20 g per cattle per day).

a,b - Means within a row with different letters are significantly different (P<0.05).

4. Discussion

Both probiotics and enzymes were widely used as feed additives in cattle production, but most of them were used separately, and the practice of mixing the two additives are less common. It was found that the addition of yeast cultures to beef cattle diets was effective in improving production performance (Liu et al., 2021). In addition, yeast can competitively bind to the binding sites in the gastrointestinal tract with the toxins produced by pathogenic bacteria and help to eliminate the toxins, which is advantageous to enhance the growth performance of cattle (Ma et al., 2021). It has also been found that the addition of brewer's yeast to cattle diets can improve the microecological environment of the rumen, regulate the pH in the rumen, increase feed conversion, and thus improve the performance of cattle (Suntara et al., 2021). Meanwhile, cellulase was also more commonly used in beef cattle production, and had a good promotion effect on the performance of beef cattle (Dehghan-Banadaky et al., 2013). In this experiment, the application of a mixture of yeast and cellulase supplementation (20 g/d) had a favorable effect on Simmental cattle production performance. It was mainly reflected in the improvement of Simmental cattle BWG and FCR. The effect of BC2 group was better, which indicated that the enzyme and probiotic supplementation needed to reach a certain dose to be effective in cattle production. The high dose (20 g/d) of enzyme and probiotic supplementation in this study also significantly improved the feed conversion of Simmental cattle.

The improvement in performance of Simmental cattle in this experiment by the bacteria and enzymes mixed feed additives was presumed to be due to the increased digestibility of the nutrients in the feed. Therefore, we measured the nutrient digestibility. The results showed that the application of enzyme

and probiotic supplementation could significantly improve the digestibility of NDF, ADF, and CP in diets for cattle. Numerous studies have shown that the addition of cellulase to diets can effectively improve the digestibility of fibrous substances in diets for animals (Abrão et al., 2017). In ruminants, the digestibility of fiber in the diets affects the amount of short-chain fatty acid production, which in turn affects the energy supply of the body (Schwaiger et al., 2013). Cellulase can degrade the cell walls of plant tissues, degrade large molecules that are difficult to absorb into small molecules, and improve the digestibility of plant fibers (Azzaz et al., 2020). The addition of yeast in this experiment also had the ability to aid digestibility. Early studies had shown that brewer's yeast could reduce the abundance of pathogenic microbiota in the rumen and promote digestibility in cattle, and improve the immune capacity of the body (Alugongo et al., 2017). The absorption and utilization of protein in diets by animals plays an important role in their muscle development (Tipton and Wolfe, 2001). In this experiment, the digestibility of protein in the diets was significantly greater in the cattle of the enzyme and probiotic supplementation group (20 g/d) than in the control group, which may also be an important reason for the significant increase in final weight of cattle in the test group.

Meat production performance has always been an important production index in the beef cattle industry, and better meat production performance means better economic benefits (Onono et al., 2012). In the present experiment, the net meat weight of the BC2 showed an increased trend compared with the control group, which indicated that the application of enzyme and probiotic supplementation in Simmental cattle production helped to promote their muscle development and, thus, improve the economic efficiency of cattle production. As mentioned in the previous section, the application of the enzyme and probiotic supplementation significantly improved the digestibility of the protein in the diets for cattle, which may also be an important reason for the better meat production performance of the cattle in the BC2 group.

5. Conclusions

The application of enzyme and probiotic supplementation (20 g/d) in Simmental cattle production can effectively improve growth performance and meat yield of Simmental cattle.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Conceptualization: Y. Liu and C. Xia. Data curation: L. Gao and X. Yan. Formal analysis: L. Gao. Funding acquisition: C. Xia. Methodology: X. Yan and Y. Liu. Writing-original draft: L. Gao. Writing-review & editing: X. Yan and Y. Liu.

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