Effects of High Rice Diet on Growth Performance, Nutrients Apparent Digestibility, Nitrogen Metabolism, Blood Parameters and Rumen Fermentation in Growing Goats

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Abstract
This study was conducted to evaluate the effects of high rice grain diet on apparent nutrient digestibility, growth performance, nitrogen digestion, blood parameters and ruminal fermentation in goats. Sixteen growing goats were divided into 2 groups and fed a normal-concentrate diet (NC, 55% concentrate of dry matter; n=8) or a high concentrate diet (HC, 90% concentrate of dry matter; n=8) for 5 wk.

Growth performance, nutrients digestibility, nitrogen digestion, blood parameters and ruminal fermentation were measured. Total weight gain and average daily gain increased in the HC group (P<0.01). Digestibility of Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF), nitrogen intake and digested nitrogen also increased (P<0.05) by HC feeding. Triglycerides, cholesterol, high density lipoprotein and low-density lipoprotein concentrations in the blood decreased (P<0.05). HC diet feeding decreased (P<0.01) the pH value, acetate level and ratio of acetate to propionate, but increased (P<0.05) the concentrations of propionate, valerate and total volatile fatty acids in the rumen.

These findings revealed that the HC diet could promote the growth of growing goats, change the ruminal fermentation pattern and lipid metabolism in the blood, but cause subacute ruminal acidosis, which might increase the risk of body health.

Keywords: High concentrate diet, Goats, Apparent digestibility, Metabolism, Fermentation

Büyüme Döneminindeki Keçilerde Pirinç Ağırlıklı Diyetin Büyüme Performansı, Görünür Sindirilebilirlik, Nitrojen Metabolizması, Kan Parametreleri ve Rumen Fermentasyonu Üzerine Etkileri

Öz
Bu çalışma, keçilerde pirinç ağırlıklı diyetin, görünür sindirilebilirlik, Büyüme performansını, azot sindirimi, kan parametreleri ve rumen fermentasyonu üzerindeki etkilerini değerlendirerek amacını yerine getirmiştir. Bu amaçla büyüme dönemindeki on altı keçi, 2 gruba ayrıldı ve 5 hafta boynuzca normal konsantrasyon (NC, %55 kuru madde konsantrasyonu; n=8) veya yüksek konsantrasyonlu diyet (HC, %90 kuru madde konsantrasyonu; n=8) verildi. Büyüme performansı, besin sindirilebilirliği, azot sindirimi, kan parametreleri ve rumen fermentasyonu ölçülüdür. Sonuçta, HC grubunun toplam kilo artışı ve ortalama günlük kilo artışı yüksek bulunmuş (P<0.01). Nitrojen Deterjan Lif (NDF) ve Asit Deterjan Lif (ADF)‘in sindirilebilirliği, azot alımı ve sindirilmiş azot oranının da, HC beslemesi ile arttığı belirlendi (P<0.05). Kandaki trigliseritler, kolesterol, yüksek yoğunluklu lipoprotein ve düşük yoğunluklu lipoprotein konsantrasyonları azaldı (P<0.05). HC diyet beslmesi pH değerini, asetat seviyesini ve asetan propionat oranını azaltırken (P<0.01), rumendeki propionat, valerat ve toplam uçucu yağ asitlerinin konsantrasyonlarını arttırdı (P<0.05). Bu bulgular, HC diyetinin Büyüme dönemindeki keçilerin gelişimini destekleyebileceğini, kandaki rumen fermentasyon düzeyini ve lipid metabolizmasını değiştirebileceğini, ancak subakut rumen asidoza neden olma riskini ortaya koymaktadır.

Anahtar sözcükler: Yüksek konsantrasyon diyet, Keçiler, Görünür sindirilebilirlik, Metabolizma, Fermentasyon

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INTRODUCTION

Relative high fiber feeding and stable microbial flora are the necessary conditions for the health of ruminants [1]. However, at present, feeding high concentrate diet is widely adopted to provide adequate protein and energy supply for meeting higher performance needs at finishing stage of ruminants [2]. Non-fibrous carbohydrates in the concentrate of diets can provide ruminants with energy and small intestinal absorbable glucose to meet high production and energy needs. Appropriate increase of dietary concentrate level can improve the production performance of ruminant animals [3]. It is well known for feeding excessive amounts of non-structural carbohydrates and highly fermentable roughages easily result in a series of metabolic diseases in high-yield ruminants [4,5]. For instance, previous studies have shown that feeding high concentrate diets can induce subacute ruminal acidosis (SARA) in ruminants [6]. The presence of SARA is an important concern in terms of both productivity and animal welfare. The high rumen digestibility of most grains in the concentrate improved the ruminal VFA production, and the accumulation of volatile fatty acids (VFA) causes a decrease in ruminal pH [7]. Typically, when rumen pH is lower than normal level, then gram-negative bacteria in the rumen and intestinal tracts releases a lot of lipopolysaccharide (LPS) [8], also known as endotoxin, is a part of the outer membrane of the gram-negative bacteria [9], seriously disturbing the normal physiological function of gastrointestinal tracts and body health. Additionally, ruminal fermentation and digestion processes mediated by symbiotic microorganisms can convert dietary carbohydrates into available nutrients such as VFA and sugars [9].

Black goat is common in the south of China, by a wide range of breeds, mainly located in Hunan, Jiangxi, Guizhou and surrounding provinces and cities. Liuyang black goat is a rare pure black goat breed in China. Liuyang black goat has tender meat, delicious taste, leaner meat, less fat, high nutritional value and strong disease resistance. Therefore, Liuyang black goat has good research value. Previous evidence has suggested that rapid growth can be achieved by feeding the diets exceeding 50–65% proportions of concentrate (mainly containing ground corn grain) to ruminants [10]. In fact, because corn grain is commonly scarce in the traditional rice cropping region of southern Asia, rice grain is thereby used as an alternative feed applied in the goat diets. Howbeit few studies were conducted to explore the effects of dietary high rice grain proportion on the gastrointestinal nutrient digestion and metabolism. The aim of this research is to conduct a feeding trail to investigate whether a diet with high unhusked rice grain proportion would affect the growth performance, nutrients apparent digestibility, nitrogen metabolism, blood metabolites and ruminal fermentation, and to seek for the rational and practical approaches of rice grain in a ruminant production system.

MATERIAL and METHODS

The study was approved by the Animal Care Committee, Institute of Subtropical Agriculture, the Chinese Academy of Sciences, Changsha, China and the Animal Care and Use Committee of Hunan Agricultural University (HAU201408).

Animals, Diets and Management

Sixteen Liuyang Black goats (6 months old, a local breed in the south of China), with an average BW of 15.33±1.67 kg were randomly divided into two groups and fed normal-concentrate diet (NC, the ratio of concentrate to forage was 55:45) and high-concentrate diet (HC, the ratio of concentrate to forage was 90:10), respectively. The ingredients and nutrient levels of the diets are given in Table 1. Unhusked rice was in the form of dried powder and provided by the Hunan LiFeng Bio-Technology Company Ltd. (Changsha, China). Before formulating the diets, the rice straw was chopped to approximately 2 cm in length.

The experiment duration consisted of 35 days, with 7 days for diet adaptation and 28 days for sampling. Diets were equally offered at approximately 08:00 and 18:00 h.

<table>
<thead>
<tr>
<th>Item</th>
<th>NC</th>
<th>HC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ingredients composition (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Forage</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice straw</td>
<td>45.0</td>
<td>10.0</td>
</tr>
<tr>
<td>Concentrate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice with shell</td>
<td>33.2</td>
<td>54.3</td>
</tr>
<tr>
<td>Soybean meal</td>
<td>9.60</td>
<td>15.7</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>6.00</td>
<td>9.80</td>
</tr>
<tr>
<td>Fat powder</td>
<td>3.20</td>
<td>5.20</td>
</tr>
<tr>
<td>Calcium carbonate</td>
<td>0.50</td>
<td>0.80</td>
</tr>
<tr>
<td>Calcium bicarbonate</td>
<td>1.10</td>
<td>1.80</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>0.60</td>
<td>1.00</td>
</tr>
<tr>
<td>Premix1</td>
<td>1.00</td>
<td>1.40</td>
</tr>
<tr>
<td>Nutrient levels4, % of DM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Crude protein</td>
<td>13.5</td>
<td>17.6</td>
</tr>
<tr>
<td>Crude ash</td>
<td>9.34</td>
<td>9.12</td>
</tr>
<tr>
<td>Crude fat</td>
<td>4.18</td>
<td>6.01</td>
</tr>
<tr>
<td>Neutral detergent fiber</td>
<td>49.8</td>
<td>38.4</td>
</tr>
<tr>
<td>Acid detergent fiber</td>
<td>36.5</td>
<td>9.51</td>
</tr>
</tbody>
</table>

1 NC: normal-concentrate diet; 2 HC: High-concentrate diet; 3 Premix composition per kg diet: 68 mg FeSO4·7H2O, 44 mg CuSO4·5H2O, 411 µg CoCl2·6H2O, 1.70 mg KIO3, 211 mg MnSO4·H2O, 126 mg ZnSO4·H2O, 56 µg Na2SeO3, 462 mg MgSO4·7H2O, 737 IU Vit. A, 8.29 mg Vit. E, 4.0 g NaHCO3, 5.1 g carrier zeolite powder; 4 Nutrient levels were measured values; 5 NFC: Non-fibrous carbohydrate. NFC was calculated in accordance with NFC = DM - (CP + EE + Ash + NDF)
respectively. All goats were fed in separate cages. Goats had free access to water and feed intake of each goat was recorded daily.

**Sampling and Collection**

The goats were kept in metabolic cages which enabled the separation of urine from feces. Feces and urine were collected from goats twice daily before feeding (08:00 h and 18:00 h) and lasted for 7 days. For feces samples, further subsamples (2% of total weight) were acidified with 10% H₂SO₄ and stored at -20°C for the determination of dry matter (DM), organic matter (OM), neutral detergent fiber (NDF), acid detergent fiber (ADF), and nitrogen (N). The fecal subsamples were dried at 65°C for 48 h and stored in plastic bags until laboratory analysis. For each goat, the total volume of urine was recorded daily. A 10% urine subsample was taken, in which 10% H₂SO₄ was added to keep pH less than 3.0 for the determination of N. Samples of the diets were collected after feeding in the morning each week, and urine was stored at -20°C until analysis.

Blood samples were collected aseptically in tubes with heparin sodium from the jugular vein of goats before feeding in two consecutive days (d34 and d35) of the sampling period, respectively. Plasma samples were separated by centrifugation by 1000 g for 15 min at 4°C, and stored at -20°C for further analysis. Rumen fluid samples were taken at 0h (before feeding in the morning), 3 h (3 h after feeding in the morning) and 6 h (6 h after feeding in the morning) on day 35 through the oral cavity. Meanwhile, pH values of rumen fluid were immediately determined after sampling using a pH meter (Model PHS-3C, Shanghai Precision Science Instrument Co., Ltd., China). Further subsamples were centrifuged at 15,000 g for 10 min and the supernatant fluids were acidified with 25% (w/v) metaphosphoric acid in a ratio of 10:1, vortexed and stored at -20°C for VFA determination.

**Chemical Analysis**

The samples of diets and feces were dried at 105°C overnight and ignited at 550°C for 6 h for measuring DM, OM and Ash (method 942.05; AOAC, 1995), respectively [11]. Crude protein content was calculated as 6.25×N which was determined using the Kjeldahl method [12]. The NDF and ADF were measured by using the procedures of Van Soest et al.[13]. The plasma biochemical components including lactate dehydrogenase (LDH), lactate (LACT), glucose (GLU), triglycerides (TG), cholesterol (CHOL), high density lipoprotein (HDL), low density lipoprotein (LDL), total protein (TP), and albumin (ALB)were determined using an Automatic Biochemistry analyzer (Cobas c 311, Roche). Insulin-like growth factor 1 (IGF-1) in the plasma was measured by ELISA kits (Cusabio, Wuhan, China) according to the instructions of the manufacturer. LPS and growth hormone (GH) in the plasma were also detected using corresponding ELISA kits (Jian Cheng Bioengineering Institute, Nanjing, China).

The ingested N (IN, g/d), fecal N (FN, g/d) and urinary N (UN, g/d) were used to calculate N balance as following:

\[
\text{Digested N (DN) [g/d] = IN–FN,}
\]
\[
\text{Retained N (RN) [g/d] = IN–FN–UN,}
\]
\[
\text{Availability of RN [%] = (IN–FN–UN)/IN × 100%,}
\]
\[
\text{Availability of DN [%] = (IN–FN–UN)/(IN–FN) × 100%,}
\]
\[
\text{Apparent N digestibility [%] = (IN–FN)/IN × 100%.
\]

The rumen fluid samples were thawed and centrifuged at 10,000 g and 4°C for 15 min. The supernatant solution was used for VFA determination by gas chromatography (HP5890, Agilent Technologies Co. Ltd., USA).

**Statistical Analyses**

Statistical analyses of data were evaluated through independent sample T-test, and animal were used as experimental unit. Values are expressed as the mean ± standard error of the mean (SEM). Statistical significance was set at P<0.05 and tendencies at 0.05≤P≤ 0.10. All statistical analyses were conducted with SPSS 19.0 (SPSS Inc., Chicago, IL, USA, 2009).

**RESULTS**

As shown in Table 2, the total weight gain and average daily gain were increased by HC (P<0.01). But HC diet significantly decreased the FCR (feed conversion ratio). Meanwhile, no significant differences in final weight and DMI were noted between groups (P>0.10). As shown in Table 3, the intake, fecal output and apparent digestibility of OM were not affected (P>0.05) by HC. HC decreased the intake of NDF (P<0.01), but increased the fecal output (P<0.01), and finally reduced the digestibility of NDF (P<0.01). ADF intake was not influenced by HC (P>0.05), but the fecal output was greater (P<0.01) and the digestibility was reduced (P<0.01) in the HC group.

Nitrogen intake, fecal nitrogen, urinary nitrogen excretion and DN were increased (P<0.05) by the HC diet (Table 4). However, there were no significant differences in the RN, DN and their availability, and N digestibility (P>0.05).

| Table 2. Effects of high-concentrate diet on growth performance in goats  |
|-----------------------------|-------|-------|-------|-----|
| Items                       | NC1   | HC2   | SEM  | P-value |
| Initial weight/kg           | 15.5  | 15.2  | 0.51 | 0.78   |
| Final weight/kg             | 17.4  | 18.7  | 0.64 | 0.34   |
| Total weight gain/kg        | 1.88  | 3.50  | 0.33 | <0.01  |
| Average daily gain (g/d)    | 67.1  | 125   | 12   | <0.01  |
| DMI (g/d)                   | 572   | 602   | 13   | >0.10  |
| FCR3                        | 11.46 | 7.27  | 1.09 | 0.032  |

1 NC: normal-concentrate diet, 2 HC: High-concentrate diet, 3 FCR: feed conversion ratio
HC increased the concentration of LACT (P=0.040), while LACT was not detected in the NC group (Table 5). Meanwhile, no significant differences were observed (P>0.05) between the dietary treatments in contents of LPS, LDH, Glu, TP, ALB and GH. Concentrations of TG (P<0.01), CHOL (P=0.029), HDL (P=0.035) and LDL (P=0.034) in blood were decreased in HC goats.

Variation of pH values in the rumen fluid was shown in Fig. 1. It is obvious that goats fed the NC diet had greater ruminal pH than that of the HC group (P<0.01). Ruminal pH in the HC group was affected by sampling hour (P<0.01). The pH values in rumen samples of two groups were the same before feeding in the morning (0 h), but it was declined in the HC group at 3 h or 6 h after feeding (P<0.01). During the time period from 3 h after the feeding until the sampling at 6 h, the average ruminal pH was below 5.7 in the HC group and remained significantly lower than that of the NC goats.

Data of metabolites in the ruminal fluid are displayed in Table 6. There was no difference in the LACT concentration of ruminal fluid in two groups (P=0.77). The acetate level was lower in the HC group (P<0.01), but the propionate (P<0.01), valerate (P=0.023), and total VFA (P<0.01) concentrations were higher in the HC group. There were no differences in isobutyrate, butyrate, and isovalerate between the two groups (P>0.05). Additionally, the ratio of acetate and propionate in the ruminal fluid in the HC group was lower than that of the NC group (P<0.01).

**DISCUSSION**

In the present study, the HC diet increased the total weight gain and average daily gain of goats, indicating that HC...
Table 6. Effects of high-concentrate diet on the ruminal lactate, molar proportions of VFAs in goats

<table>
<thead>
<tr>
<th>Items</th>
<th>NC(^1)</th>
<th>HC(^2)</th>
<th>SEM</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>LACT (mmol/L)</td>
<td>0.125</td>
<td>0.144</td>
<td>0.030</td>
<td>0.77</td>
</tr>
<tr>
<td>Total VFA (mmol/L)</td>
<td>97.4</td>
<td>164</td>
<td>11</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Acetate: Propionate</td>
<td>4.24</td>
<td>2.20</td>
<td>0.34</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

\(^1\)NC: normal-concentrate diet, \(^2\)HC: High-concentrate diet, \(^3\)LACT (lactate), \(^4\)VFA (volatile fatty acids)

Dietary nutrients, such as fiber, is also essential for digestion mediated by chewing nutrients, microbial fermentation in the rumen and the rate of passage in the gastrointestinal tract \(^{17}\). In this study, the decreases in NDF intake and apparent digestibility and increase in fecal output of NDF were observed in the HC group. ADF fecal output was greater and the digestibility was reduced in the HC group. These results suggest that growing goats are less able to digest and degrade NDF and ADF of diet in the HC group. Our data agree with that of Mahgoub that observed that the final weight and dry matter intake are not affected by the ratio of dietary concentrates to forage \(^{14}\). Because the NC group fed a large amount of forage to the goat in the study and a little concentrate, contrary to HC group fed a large amount of concentrate, finally, the FCR of the HC group was lower than NC group. We speculate that an increase of VFA in the rumen might partly be contributed to the overall weight and average daily gain of goats. The rumen absorbs energy material (i.e., SCFA), which accounts for about 70% of the energy required for ruminant growth, maintenance and production \(^{15}\). Additionally, we found that DMI in the HC group was not varied, which is agreed with previous research which has performed in Omani goats \(^{16}\).

In this study, HC did not influence plasma LPS concentration. Gozho et al. \(^{27}\) also did not observe increased LPS in the peripheral blood plasma of lactating dairy cows with SARA. This is partly attributable to the fact that lipoprotein can neutralize LPS in the body. There was evidence that lipoproteins can neutralize LPS in vivo \(^{28}\). Several experiments have also demonstrated that plasma lipoprotein, especially HDL, can be combined with LPS and preferentially shunt liver cells away from liver macrophages, thereby increasing LPS excretion through the bile and preventing immune responses \(^{29}\). HC diet reduced TG, CHOL, HDL and LDL, indicating that the metabolism of lipid in blood was changed. These data are very important in preventing hyperlipidemia and the heart and liver diseases \(^{30}\).

Ruminal propionate produced during ruminal fermentation is used as a substrate in the gluconeogenesis pathway to produce glucose \(^{31}\). In the current study, although propionate in the rumen feeding HC was higher than NC, there were no significant differences on the concentration of glucose between NC and HC suggesting that ALB is the main protein produced in the liver and usually accounts for more than 50% of the total blood protein content \(^{32}\). Both TP and ALB were not influenced by HC in our study. TP is more sensitive to the effects of nutrition, but the changes are often subtle and difficult to detect and explain. The level of serum protein depends on a number of factors, including the presence of inflammatory or metabolic liver processes and other organ diseases \(^{33}\).
Hormones can regulate various physiological and behavioral activities. The synthesis, storage and secretion of GH by the somatotropin cells, stimulate the growth of animals and the production of IGF-1 [34]. There is a positive correlation between the concentration of IGF-1 and the growth rate of animals [35]. Though the HC diet did not rise the plasma GH level, there is a significant increase in IGF-1, suggesting that the HC diet promotes growth development in goats, which can explain the increases in the total weight gain and average daily gain. The enzyme LDH is widely found in body tissues. As a marker of common injury and disease, it is released during tissue damage. In particular, feedback inhibition by LDH can reduce the rate of conversion of pyruvate to lactic acid at high lactate concentration [36]. Our data display the significant rise in plasma LACT content in response to the HC diet, but there was no significant influence in LDH between treatments. Further exploration is needed to explain this phenomenon.

The pH value is an extremely important indicator of the epithelial barrier and the ruminal metabolic state [10]. In this study, the rumen pH of NC group ranged from 6.07 to 6.73, and it was ideal for optimum rumen metabolism [37]. The rumen pH thresholds of 5.8 or lower were usually used to define a clinical diagnosis of SARA [38]. In this study, the data in Fig. 1 indicated that NC-fed goats have a higher rumen pH. We observed that the duration of rumen pH below 5.8 in the SARA group was approximately 4 h after the first feeding, which meant that SARA occurred in goats by feeding the HC diet. The sampling time also affected the rumen pH. Briefly, the data indicated that HC-raised induced SARA in growing goats.

LACT is one of the main products in the rapid fermentation of rumen, and a large number of LACT can cause SARA has been widely recognized [19]. As we know, LACT can cross the rumen wall and dissolve in the blood to cause an increase in plasma. Although the concentration of LACT was found to be negligible in the rumen, we found a significant difference in the blood. This may mean that LACT was produced and immediately converted to VFA as suggested by some authors, thus preventing the accumulation of LACT in the rumen [40]. Previous studies have shown that long-term HC feeding increased the production of VFA in the rumen. Accumulation of these acids reduces the long-term HC feeding increased the production of VFA in the rumen. Accumulation of VFA decreased the ruminal pH and led to SARA. Therefore, an HC diet destroyed the balance of the rumen fermentation.

This study is the first in using unhusked rice as HC diet to survey the growth performance, nutrients apparent digestibility, nitrogen digestion, blood parameters and rumen fermentation of growing goats. In summary, these findings revealed that the HC diet could promote the growth of growing goats, changed the ruminal fermentation pattern and lipid metabolism in the blood, but cause subacute ruminal acidosis, which increases the risk of body health. The exact mechanism of this relationship between the blood parameters and the rumen microbial metabolites is needed to explore.

ACKNOWLEDGMENTS

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CONFLICT OF INTEREST

There is no conflict of interest.

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