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The Effect of Subacute Ruminal Acidosis of Dairy Cows on Productivity, Digestibility and Greenhouse Gas Emission

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Abstract

The purpose of this study was to investigate the performance, digestion of the diet and greenhouse gas emission of cows with subacute ruminal acidosis (SARA). Twelve cows were included. The blood parameters, milk yields, manure, and urine of healthy (H group) and cows with SARA (R group) were analyzed. The results showed that the plasma concentrations of total protein (TP) and globulin (GLO) of the R group were significantly lower than those of the H group. Aspartate amino transferase (AST), non-esterified fatty acids (NEFA), beta-hydroxybutyric acid (BHBA), creatinine kinase (CK) and L-lactate were significantly higher in the R group than in the H group. The levels of neutral detergent fiber (NDF) and acid detergent fiber (ADF) in feces from the R group were significantly lower than in the H group. Milk protein and milk fat were significantly lower in the R group than in the H group, and the energy corrected milk (ECM) value of the R group was significantly lower than that of the H group. The emission of ammonia and methane by the R group was slightly lower than by the H group. These results showed that the forage digestibility was significantly higher in the R group than the H group. The performance and ammonia and methane emission in the R group were slightly lower than those of the H group.

Keywords: dairy cows, digestibility, greenhouse gas emission, production, subacute rumen acidosis

1. Introduction

It is generally predicated that global warming is caused due to increases in the concentration of greenhouse gases (GHG), from anthropogentic activities including carbon dioxide, methane, nitrous oxide and chlorofluorocarbons (Patra, 2014). Currently, it was reported that agriculture plays an important role in global environment issues. Moreover the livestock sector represents a significantly source of greenhouse emissions worldwide (Gerber et al., 2013). It is all know there were climate change, sea level increased etc when happened global warming. And the climate change has direct and indirect impacts on livestock production (Chauhan & Ghosh, 2014; Taqi, Hassanein, & Khalil, 2013). This is an infinite loop, thus we should mitigation the greenhouse emission increased.

A life cycle assessment of GHG emissions indicated that livestock contributes about 18% to the global anthropogenic GHG emissions and dairy cattle sector 4% of total anthropogenic GHG emissions (Food & Organization, 2010). The main resources of dairy cow sector are feed materials, enteric fermentation, manure storage and processing. Feed materials accounts for half of total greenhouse emissions. The enteric fermentation contributing to about 40 percent of total greenhouse emissions (Gerber et al., 2013). Livestock manure management accounts for almost 10% of greenhouse gas emissions from agriculture globally (Owen & Silver, 2015). In China the total production of animal manure from large-scale centralized farms is about 837 million tons, of which 382 million tons is cow manure (Van der Weerden, Luo, Dexter, & Rutherford, 2014). In additon, ammonia (NH3) emitted from cattle manure has environmental and human health effects, including eutrophication of surface waters, acidification of ecosystems, and fine particulate matter formation in the atmosphere (Lee et al., 2012). It has aroused great concern regarding the pollution associated with livestock farming. Above all, there are also several factors affect above conditions, such as dry matter intake, feed digestion, feed component, and manure storage methods. And there is no reported whether the diseases can affect the GHG emission.

China accounts for the first place of cows numbers in the world. The number of cows reached 14.7 million until 2014. With the rapid development of the dairy industry, some farmers feed a large amount of easily fermentable carbohydrate-based feeds or acidic feeds in early and mid lactation to improve the production performance of dairy cows. This can result in high-yielding dairy cows suffering from a series of nutritional and metabolic diseases, especially subacute ruminal acidosis (SARA). SARA is defined as periods of moderately depressed ruminal pH (the minimum pH varies between 5.2 and 5.6) (Guo et al., 2013); it is one of the most common and costly digestive disorders of dairy cows, especially in well-managed dairy herds (Antanaitis, Žilaitis, Kučinskas, Juozaitienė, & Leonauskaitė, 2015). SARA is characterized by a decrease in ruminal pH, weight loss, slow growth and decreased feed conversion rate (Danscher et al., 2015). In addition, it can result in reduced dry matter intake (DMI), milk production, and milk fat content, and cause diarrhea, rumenitis, laminitis, and liver abscesses, as well as increased mortality rates (Plaizier, Krause, Gozho, & McBride, 2008). It therefore increases the cost of veterinary care. Besides, there have been no studies on the relationships among the milk yield, feces, urine output and GHG emissions of dairy cows with SARA, and no studies reported that alterations in the clinical parameters may directly or indirectly increase the emissions of GHG. Therefore, in this study, an attempt was made to determine whether changes occurred in the feed intake, productivity and feces production of dairy cows with ruminal acidosis compared with healthy cows. Moreover, we wanted to know whether changes occur of GHG emission between the cows with SARA and healthy cows.

2. Materials and Methods

2.1 Ethics Statement

The study was approved by the farm owner and all animal experiments were conducted according to the International Guiding Principles for Biomedical Research. The protocol was approved by the Committee on the Ethics of Animal Experiments of the Heilongjiang Bayi Agricultural University.

2.2 Animals and Groups

Twelve Holstein cows were selected (average parity, 2.1 ± 1.0 lactations; body weight, 618 ± 84 kg) from an intensive cattle farm with 2000 dairy cows in Heilongjiang, China. The cows were divided into two groups, according to the pH value of rumen fluid and clinical signs: a healthy group (pH > 6.2, n = 6; H group) and a SARA group (5.2 < pH < 5.6, n = 6; R group). The cows were housed in individual stalls, bedded on rubber mattresses, and had free access to drinking water throughout the trial. They were fed a total mixed ration (TMR) at 04:00 and 16:00 daily during the transition period; the composition of the feed is shown in Table 1. The cows were milked three times daily, at 04:00, 12:00 and 20:00. The amounts of food intake, and the output of milk, manure, and urine were recorded each day, and the study was conducted for 4 days.

Table 1. Composition of the diet fed to the dairy cows

Ingredients (g/kg)		Nutrient composition	
Concentrated feed	308	NE (MJ/100g)	1060
silage corn	380	EE (%)	8.09
Chinese hay	27	NDF (%)	39.08
domestic alfalfa	68	N (%)	3.31
oat grass	40	P (%)	0.25
cottonseed	9	DM (%)	58.26
wet corn	71	OM (%)	95.75
tableting	31		

Note. NE: net energy; EE: ether extract; NDF: neutral detergent fiber; ADF: acid detergent fiber; N: nitrogen; P: phosphorus; DM: dry matter; OM: organic matter.

2.3 Sample Collection

Blood samples were collected from the vena caudalis mediana using sodium heparin before feeding and milking in the morning for 4 days, and immediately centrifuged at 3000 × g for 5 min at room temperature. The supernatants were aliquoted separately into Eppendorf tubes (1 mL plasma/tube) and stored at -20 °C until

analysis. The samples were later analyzed for determination of glucose (Glc, enzymatic), non-esterified fatty acids (NEFA, colorimetric method), beta-hydroxybutyric acid (BHBA, enzyme rate method) total protein (TP, chemical method), albumin (ALB, chemical method), globulin (GLO, chemical method), aspartate aminotransferase (AST, rate method), creatinine (CREA, enzymatic kinetic method), creatine kinase (CK, rate method), and L-lactate (L-ACT, rate method). The parameters in plasma were analyzed using a clinical auto-analyzer (Cobas Integra, C701; Hoffmann-La Roche Ltd., Basel, Switzerland).

The cattle were milked three times per day, and the total milk output was recorded three times a day. At each milking, 10 mL of milk was collected, mixed with preservative and stored at -4 °C until analysis. The milk samples were later analyzed for determination of milk protein, milk fat and lactose (using a milk composition analyzer, Funke Gerber, Germany).

Fresh feces and urine samples were collected from individual cows. Feces were collected from the ground in the barn at 05:00, 13:00 and 20:00, and the total weight was recorded. Samples of 500 g fresh feces were collected from the rectum of each cow every day and stored at -20 °C until analysis. Fecal samples were later analyzed for determination of dry matter (DM, dried at 65 °C in a forced-air oven for 48 h), neutral detergent fiber (NDF, Van Soest's detergent fiber analysis), acid detergent fiber (ADF, Van Soest's detergent fiber analysis), starch (anthrone sulfuric acid method), crude protein (CP, micro-Kjeldahl method), nitrogen (N, Dumas method), and gross energy (GE).

Individual urine samples (approximately 200 mL per sampling) were collected at the same time as the fecal collections by massaging the perineum. The samples were stored frozen at -20 °C until analysis, and were later analyzed for determination of creatinine (CREA, enzymatic kinetic method), urine acid (UA, molybdenum phosphate method), and urea (urease method).

2.4 Methane and Ammonia in Fecal Emissions

Aliquots of the manure or urine samples were composited on an equal-weight basis for the healthy (H) and SARA (R) groups; the pooled fecal and urine samples were mixed on the basis of the ratio of daily manure and urine produced by the H and R groups. Gas emission from manure was defined as the rate of gas emission (mg/kg per min) from cattle feces and urine composites incubated for 100 to 122 h in simulated storage under a controlled environment (room temperature, 25 °C, and continuous air influx of 2 L/min). In this experiment, the emission potential of methane（CH4） in manure was analyzed using a steady-state flux chamber system (Wheeler, Topper, Brown, & Varga, 2007). The ammonia (NH3) emission potential in feces was analyzed using Nash reagent by spectrophotometry. Briefly, the manure and urine mixtures were placed in a chamber within glass jars (surface area 80 cm^2) equipped with a lid consisting of two inlets, which were connected to a circular diffusion Teflon tube inside the chamber, through which continuous-sweep airflow (2 L/min) was provided. The chamber outlets were attached to a multi-valve switching apparatus, which allowed automated, sequential gas measurements from each jar using an INNOVA 1412 photoacoustic gas monitor (AirTech Instruments A/S, Ballerup, Denmark). Emission data were collected approximately every 3 h; the measurements were converted to a per-minute basis and these data were used in the statistical analysis. Manure (200 g) was placed in the chambers immediately before the beginning of the incubation, thus representing manure processes occurring on the barn floor immediately following excretion, and mixing of feces and urine. CH_4 production in the rumen depends upon dietary factors, rumen function, and fermentation dynamics, and incubations were carried out at 25 °C for 122 h (Lee et al., 2012).

2.5 Statistical Analysis

SPSS17.0 software (SPSS Inc, Chicago, USA) was used for statistical analysis using single factor variance; the results were expressed as means ± standard deviation. Energy corrected milk (ECM) was calculated as: [(0.038 × g crude fat + 0.024 × g crude protein + 0.017 × g lactose) × kg milk] ÷ 3.14 (Reist et al., 2002).

3. Results

3.1 Blood Biochemistry, Milk Components and Urine Parameters

Table 2 shows that the concentrations of TP in plasma of R group was significanlty difference compare to H group ($P < 0.01$), the concentration of GLO in R group was significantly lower than H group ($P < 0.05$), the concentration of CK, AST and NEFA in R gorup was significantly higher than H group ($P < 0.01$), and the L-ACT, CREA and BHBA contents in plasma of R group were significantly higher than in the H group ($P < 0.05$). The CREA contents in urine of R group was significantly lower than H group ($P < 0.05$). The concentration of urea in urine of R group was significantly lower than H group ($P < 0.01$). In addition, the levels of fat and protein in the milk of the R group were significantly lower than those of the H group ($P < 0.01$).

Table 2. Parameters of blood, urine and milk compared between healthy cows and those with SARA

Items	H group	R group
Blood biochemistry		
TP (g/L)	79.71±5.95	71.59±4.10**
GLO (g/L)	42.39±8.38	35.18±5.49*
AST (U/L)	82.88±13.15	97.36±12.25**
Glc (mmol/L)	4.23±0.58	4.29±0.55
NEFA (mmol/L)	0.41±0.18	0.71±0.18**
BHBA (mmol/L)	0.68±0.15	1.04±0.49*
BUN (mmol/L)	5.15±0.48	5.81±2.26
CREA (umol/L)	61.50±7.38	69.45±7.85*
CK (U/L)	159.56±32.09	337.36±77.19**
L-ACT (mmol/L)	0.39±0.14	0.55±0.23*
Urine biochemistry		
CREA (umol/L)	4631.48±1780.42	2749.30±555.76*
UA (μmol/L)	756.33±222.07	755.69±208.28
Urea (mmol/L)	100.99±25.22	64.19±16.40**
Milk components		
Fat	3.28±0.43	2.55±0.47**
Protein	3.31±0.38	2.83±0.27**
Lactose	5.12±0.31	5.20±0.17

Note. TP: total protein; ALB: albumin; GLO: globulin; AST: aspartate aminotransferase; Glc: glucose; NEFA: non-esterified fatty acid; BHBA: beta-hydroxybutyric acid; BUN: blood urea nitrogen; CREA: creatinine; CK: creatinine kinase; UA: urea acid; L-ACT: L-lactate.

** Indicated highly significant difference ($P < 0.01$); * indicates significant difference ($0.01 < P < 0.05$); and without * indicates no significant difference ($P > 0.05$).

3.2 Fecal Component Analysis

As shown in Table 3, there was a significant difference in the ADF between the H group and the R group ($P < 0.01$); and the NDF of R group was significantly lower than the H group ($P < 0.05$). ADF and NDF of the R group were lower than those of the H group. Moreover, the EE of the R group was significantly higher than that of the H group ($P < 0.01$). There was no significant difference in the DM, starch, CP, nitrogen and GE between the H and R groups. However, the concentrations of CP, N and GE in the R group were slightly higher than in the H group.

Table 3. Differences in fecal components between healthy cows and those with SARA

Items	Feces	
	H Group	R group
Dry Matter (%)	16.79±1.43	16.93±4.01
NDF (%)	50.67±3.52	44.60±1.96*
ADF (%)	33.77±1.85	27.33±2.38**
Starch (%)	2.45±0.20	2.33±0.30
CP (g/100g)	2.41±0.46	2.77±0.17
EE (g/100g)	0.53±0.18	2.41±0.38**
N (%)	2.18±0.14	2.30±0.13
GE(KJ/100g)	256.04±34.91	291.33±22.19

Note. NDF: neutral detergent fiber; ADF: acid detergent fiber; CP: crude protein; GE: gross energy; N: nitrogen; EE: ether extract.

** Indicates highly significant difference ($P < 0.01$); * indicates significant difference ($0.01 < P < 0.05$); and without * indicates no significant difference ($P > 0.05$).

3.3 Differences in Performance, Manure and Urine Output between Healthy Cows and Those with SARA

As shown in Table 4, there was a significant difference in ECM and urine/ECM between the H group and the R group ($P < 0.05$), but there was no significant difference in intake, fecal dry matter excretion (FE), urine output, FE/ECM, ECM, ECM/DMI and FE/DMI.

Table 4. Comparison of performance between healthy cows and those with SARA

Items	H group	R group
DMI(kg)	13.38±2.28	13.30±2.71
MY(kg)	27.57±5.20	26.34±5.15
FE(kg)	4.88±1.59	4.64±2.03
urine(L)	23.15±6.85	15.53±4.40
ECM	25.48±4.46	21.44±2.99*
FE/DMI	0.37±0.10	0.34±0.11
FE/ECM	0.19±0.04	0.22±0.11
ECM/DMI	1.93±0.34	1.69±0.47
Urine/ECM	1.00±0.20	0.72±0.15*

Note. ECM: energy corrected milk; FE: fecal dry matter excretion; MY: milk yield.

** Indicates highly significant difference ($P < 0.01$); * indicates significant difference ($0.01 < P < 0.05$); and without * indicates no significant difference ($P > 0.05$).

3.4 Comparison of Emission of CH_4 and NH_3 between Healthy Cows and Those with SARA

The composited fecal and urine samples were thawed and mixed in a ratio of 1.17:1 and 1.58:1 for the H group and R group, respectively. There was no significant difference between the two groups; and emission values peaked at 40 h for both groups. The trend in emissions was the same (Figure 1). Figure 2 shows that, the R group achieved peak emission earlier than the H group, but the emission of NH_3 was lower than that of the H group after 12 h. Moreover, the total emissions of NH_3 by the R group were lower than those for the H group.

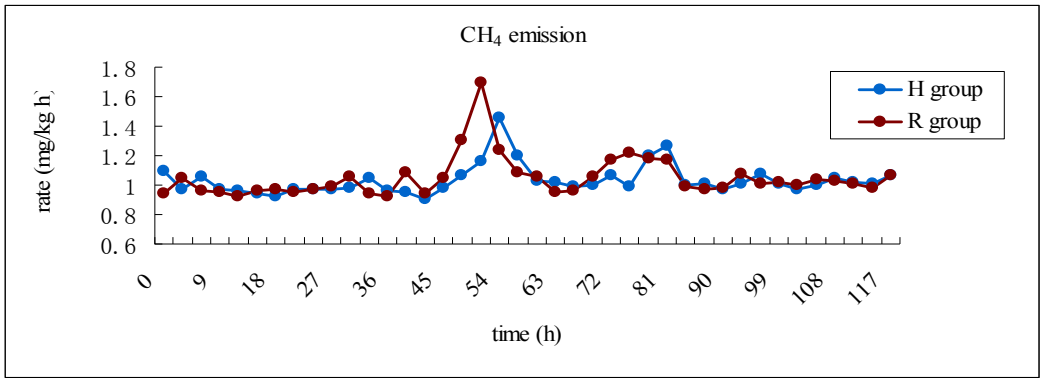

Figure 1. CH_4 emission curves for healthy cows and those with SARA

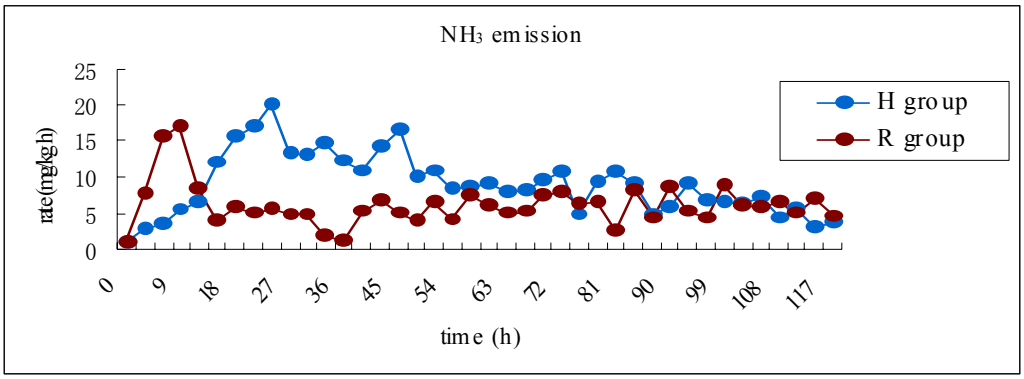

Figure 2. NH_3 emission curves for healthy cows and those with SARA

4. Discussion

4.1 Differences in Plasma Parameters between Healthy Cows and Those with SARA

SARA is characterized by reduced DMI, milk production, and milk fat content, and it causes diarrhea, rumenitis, laminitis, and liver abscesses, as well as increased mortality rates (Plaizier et al., 2008). However, it has also been reported that the DMI is not decreased in cows with SARA (Guo et al., 2013). In our study there is no significant difference between healthy cows and those cows with SARA, which is consistent with the research of Guo et al. (2013). It maybe associate with the severity of SARA or the individual difference of the cows. And no studies stated clearly the causes of DMI decrease. NEFA is mobilized as the energy of the body becomes insufficient (Bobe, Young, & Beitz, 2004; Li et al., 2012). NEFA and BHBA are indictors of negative energy balance (NEB): cows are in NEB when the contents of BHBA and NEFA are increased (van Knegsel, Van Den Brand, Dijkstra, Tamminga, & Kemp, 2005). In our study the BHBA and NEFA of cows with SARA was increased when compared with healthy cows.

AST is produced predominantly in the liver and its plasma levels become elevated whenever a disease process affects liver cells (Kunutsor, Apekey, & Walley, 2013; Vozarova et al., 2002). In our study, the concentration of AST was increased, which suggested that the liver may be damaged when cows develop SARA. The TP content also changes when the liver is damaged; it consists of globulin and albumin (Volynets et al., 2012). When the liver is damaged or chronic diseases such as diarrhea cause dehydration, the concentration of TP will be increased; cows with SARA usually have diarrhea. In this study we found the concentrations of TP and GLO were decreased, which was inconsistent with previous research.

In addition, CREA and CK are increased when muscle is damaged, and in our study these parameters were increased, which suggests that cows with SARA have muscle damage (Ehlers, Ball, & Liston, 2002). It has been reported that lactic acid increases in the rumen when the ruminal pH is lower than 5.5 (Morgante, Stelletta, Berzaghi, Gianesella, & Andrighetto, 2007); in our study, the lactate level was significantly higher in the R than in the H group.

4.2 Comparison of Performance Parameters between Healthy Cows and Those with SARA

Paradoxically, reduced DMI is seen as a clinical sign of SARA, and several researchers have shown that DMI decreases during experiments involving induced SARA. However, the effects of induced SARA on DMI are inconsistent; some researchers reported that the induction of SARA did not reduce feed intake (Guo et al., 2013). In this study, we found no difference in DMI between healthy cows and those with SARA, as in some previous studies. A decrease in milk yield is another sign of SARA, but in our study there was no significant difference in milk yield between healthy cows and those with SARA, while the ECM of the R group was significantly lower than that of the H group. The ECM is calculated from the amounts of milk fat, milk protein and lactose (Reist et al., 2002). Reduced milk fat content is frequently used on farms as an indicator of SARA, and is described as “low milk-fat syndrome” or “milk fat depression”. Milk fat depression has been used as a basis for systems predicting the effectiveness of different diet structures in encouraging chewing (Enjalbert, 2006). Rumen pH is positively associated with milk fat concentration (Kolver & De Veth, 2002). The milk fat and milk protein of the R group in this study were significantly lower than those of the H group, which indicates an effect of SARA on milk production.

4.3 Characteristics of Rumen Digestion in Healthy Cows and Those with SARA

The feed digestibility and DMI are decreased when cows develop SARA (Antanaitis et al., 2015). In our study, the digestion of NDF, ADF and starch was increased in cows with SARA when compared with healthy cows. The ADF and NDF are often used to estimate feed intake, energy values and performance. It has been reported that the digestibility of NDF and ADF has a tendency to decrease as the energy level increases (Li, He, Aziz-ur-Rahman, & Cao, 2014) and when the ruminal pH is lower than 6.3, the digestion of ADF and NDF is decreased (Anantasook et al., 2013). These observations are not in agreement with our results. Fat is the most variable component in milk and is highly dependent on dietary composition and ruminal fermentation characteristics (Moraes, Strathe, Fadel, Casper, & Kebreab, 2014). In our study, the milk fat of the R group was significantly lower than that of the H group, which suggests that the digestive ability of the rumen was decreased. This is consistent with the clinical signs of SARA. Otherwise, the digestion of CP, nitrogen, and starch showed no significant differences between the groups, but the digestion of CP and nitrogen was slightly lower in cows with SARA than in healthy cows.

4.4 Comparison of Ammonia and Methane Emissions between Healthy Cows and Those with SARA

NH_3 and CH_4 are emissions from the dairy industry, and which to be associated with severe environment pollution. Dairy cows are one of the largest livestock sources of NH_3 emissions (Hünerberg et al., 2013). CH_4 production in the rumen depends upon dietary factors, rumen function, and fermentation dynamics (Budak & Yılmaz, 2013; Patra & Lalhriatpuii, 2016). In dairy cows with SARA, the ruminal pH is decreased, which affects rumen function, and results in a decrease in CH_4 production. There is a strong relationship between CH_4 production and DMI or energy intake. A number of studies have also reported that feed intake (DM or energy) is a key explanatory variable in equations for predicting CH_4 emissions in cattle (Patra & Lalhriatpuii, 2016). In our study, there was no significant difference in DMI and energy intake between the H and R groups. Kasuya and Takahashi (2010) found a positive correlation between NDF intake and daily CH_4 emission (Aguerre, Wattiaux, Powell, Broderick, & Arndt, 2011). In our research, the NDF digestibility in the R group was higher than in the H group, while the emission of CH_4 showed no significant difference. NH_3 emission from manure, which is a serious concern, originates primarily from urine and especially urinary N. The changes in fecal N and urinary N excretion, and urinary N composition suggest that manure N may be less vulnerable to NH_3 volatilization in cows with high compared with low feed conversion efficiency (Arndt, Powell, Aguerre, Crump, & Wattiaux, 2015). Moreover, many studies have shown strong links between urinary N concentration and NH_3 emissions. The majority of urinary N is in the form of urea which, when mixed with urease enzymes found in soil and feces, is rapidly converted to ammonium and NH_3 gas. In our study, the amount of N in the feces and urine of the R group was significantly lower than in the H group, and the amount of NH_3 emitted by the R group was lower than that of the H group (Figure 2). A reduction in manure pH during the first days of storage and formed the crust at the surface, lead to the NH_3-oxidizing bacteria in the organic crust may reduce the emission of NH_3 (Aguerre, Wattiaux, & Powell, 2012). CP is mainly converted to NH_3 in the rumen, but there was no significant difference in CP between the H and R groups in our study.

5. Conclusion

In summary, this study on the changes in production performance, digestion and GHG emission of cows with SARA and healthy cows showed that the performance and digestion of cows with SARA was decreased when compared with healthy cows. There was no significant difference in GHG between the two groups of dairy cows, but the emission of NH_3 by the R group was lower than that by the H group.

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