Rumen Degradability and Post-ruminal Digestion of Nitrogen and Amino acids by Cows Grazing Temperate Pasture

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Abstract This experiment was conducted to evaluate nutrient digestion in the rumen and flow to the duodenum of steers grazed on orchardgrass (*Dactylis glomerata* L., OG) or meadowfescue (*Festuca pratensis* Huds., MF.) pastures located on northern part of Japan without supplement. Fifty-two digestion trials were carried out grazing Holstein steers fitted with cannulas in the rumen, duodenum and distal ileum in the three years. The pastures were divided into several paddocks, and the steers grazed each paddock for a day and allowed double of dry requirements the grazing cattle. The pre-grazing herbage mass did not significantly differ between pastures but the crude protein (N×6.25) content of herbage ranged from 19.3% to 27.5% on OG pasture and from 20.0% to 32.8% on MF pasture. Total N and AA-N intake did not differ but the degradability of dietary nitrogen in the rumen (RDN) was higher in MF than in OG pasture. The ratio of RDN to OM truly digested in the rumen was negatively related to the apparent N absorption in the rumen, and it was suggested that the amount of apparent N absorption in the rumen would be positive when the ratio was above 25g/kg. Non-ammonia nitrogen (NAN) and AA-N flows to the duodenum of steers were lower in MF than in OG pasture. Duodenal AA flows were more linked to duodenal NAN flows and accounted for approximately 60% of duodenal NAN flows. The proportion of methionine and lysine slightly increased in the duodenal flows compared with the consumed AA, but a comparison between the essential AA composition of milk and the lean tissue indicated that duodenal digesta was most limiting in methionine, lysine, arginine, and histidine, and that deficiencies of arginine and histidine for milk production were relatively small.

Key words Grazing cows, Temperate pastures, Nitrogen, Degradability, Post-ruminal digestibility

1 Introduction

The source of dietary N and energy fed to ruminants significantly influences the utilization of N and energy in the rumen and nutrient flow to the small intestine. Well-managed temperate pasture herbage is characterized by high N content and degradability in the rumen (Aibibula *et al*. 2003; Bryant *et al*. 2013). The dietary N degraded in the rumen (RDN) is synthesized to microbial protein or absorbed from the rumen wall as ammonia when there is surplus microbial protein synthesis. Several researchers (Minson, 1990; Holden *et al*. 1994; Elizalde *et al*. 1998; Ulyatt *et al*, 1988) have reported of high protein intake in well-managed temperate pasture and high proportion of protein degradability (60 - 80%) in the rumen. Although intake and apparent absorption of dietary N from fresh herbage may be high, the quantity of dietary protein actually absorbed from the small intestine is not always high. A limitation in ruminal available energy and/or an excess of the dietary N degraded in the rumen (RDN) restrict the conversion efficiency of RDN to microbial N. The organic matter truly digested in the rumen (OMTDR) has been used as one of the parameters to estimate the ruminal available energy (Beever and Siddons, 1986; Kolver *et al*., 1998, McCarthy *et al*., 1989). Beever and Siddons (1986) suggested that the ratio of RDN to OMTDR should be approximately 25g/kg for optimal N utilization in the rumen. Consequently, metabolizable protein and AA supply to the small intestine may be a limiting factor in the performance of high yielding ruminants in a grazing system even with an increase in intake and degradability of dietary N (Leaver, 1985). Several reports (Wilkerson *et al*., 1993; Titgemeyer and Loest, 2001) have indicated that lack of some essential amino acids limits animal production. Our objective was to study the effects of fresh herbage on N and amino acids nutrition of steers grazing temperate pasture without supplementary feeds.

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2 Materials and methods

2.1 Pasture and grazing method Orchardgrass and meadowfescue pastures located at the experimental farm of Obihiro University of Agriculture and Veterinary Medicine in Hokkaido, Japan, were used in an intensive rotational grazing. Fifty-two digestion data were carried out using Holstein steers fitted with rumen, proximal duodenum and distal ileum (average body weight: 321kg) and grazed on each pasture without supplementary feed from May to October alternatively, in the pass three years. The pastures were divided into several paddocks by using electric fences and the steers grazed each paddock daily. The steers were rotated to a new paddock at 0900 of every day. Steers were able to access water and mineral block at all times, but other supplementary feed were not given to the steers during the experimental period. Chromic dioxide (Cr_2O_3) was used to estimate duodenal, ileal digesta flow and fecal output of steers. Each steer received twice daily (0900 and 1700) a gelatin bag containing 4.0 g of Cr_2O_3 inserted into the rumen during the experimental period.

2.2 Sample collection Each experimental period lasted 23 days; the first 14 days were used to allow the steers to adapt to each new pasture and the following 9 days were used for collecting samples. Herbage mass and composition of the grass were measured and herbage samples were collected before grazing daily during the first 5 days of the sample-collection period. Herbage mass was measured by cutting herbage within four quadrates (0.5m × 0.5m) to ground level using hand-held electric grass clippers in each paddock. Herbage samples, plucked by hand to adjusted grazing height from many different areas were composited. Fecal sample were collected at 0900, 1500, 2100 and 0300 during the 5 days and were composited on an equal weight basis for each steer. The composited sample of herbage and feces were dried at 60℃ for 48 hrs in an air forced oven and then ground to pass a 1-mm screen and stored for subsequent analysis. Duodenal and ileal digesta were collected every four hours on the last 2 days of the sample-collection period. Each of duodenal and ileal digesta sample was mixed on the fresh weight basis. The composited sample was divided into two portions; one half was lyophilized for subsequent general analysis and the other was acidified with 50% H2SO4 and frozen at −20℃ until measurement of ammonium N concentration. The samples of ruminal fluid were obtained every four hours on day 8 of the sample-collection period. The samples of ruminal fluid collected were immediately measured for ruminal pH and then each sample was strained through four layers of gauze and acidified with 50% H2SO4 and frozen at −20℃ until analyzed. Ruminal fluid was also obtained at 1200 on the last day in each experimental period to obtain rumen bacteria fraction. The bacterial fraction was isolated using the method of Smith and McAllan (1974) stored at −20℃ and freeze − dried until appropriate chemical analysis.

2.3 Analytical methods Herbage, duodenal and ileal digesta, and fecal samples were analyzed for dry matter (DM), ash, N (crude protein) (AOAC, 1990), neutral detergent fiber (NDF) and acid detergent fiber (ADF) (Goering and van Soest, 1970). The neutral detergent insoluble N and acid detergent insoluble N fractions were determined using N analysis of the neutral detergent and acid detergent residue. Herbage was analyzed for non-protein N and soluble true protein using the procedures of Licitra *et al.* (1996). The ammonium-N concentration in the rumen fluid and duodenal digesta was determined by colorimetric method (Okuda and Fujii, 1968) and urea N in serum was determined using a kit (Urea N Test WAKO; 278 − 04801, WAKO Chemicals, Osaka, Japan). In vitro DM digestibility of herbage was determined according to the method of Tilley and Terry (1963). Chromium concentration of duodenal, ileal and fecal samples were determined by phosphoric acid and potassium reagent method (Morimoto, 1970). Purine contents in rumen bacterial fraction and duodenum digesta were determined using the technique of Zinn and Owens (1986). Herbage, each digesta and rumen bacteria samples were hydrolyzed with HCl (6N) for 24 h at 110℃ and amino acids quantified using a JLC − 500 amino acid analyzer.

2.4 Calculation and statistical analysis DM flow to duodenum, ileum and feces were calculated by dividing the amount of Cr dosed daily by Cr concentration of duodenal, ileal and fecal samples. Herbage DM intake was estimated by dividing fecal DM output by DM indigestibility (1 − *in vitro* DM digestibility) of herbage. Duodenal N flows of microbial origin were calculated by dividing the amount of purine flowed to duodenum by the ratio of N to purine in rumen bacterial fraction. Apparent OM digestibility in rumen was corrected for microbial contribution to calculate the OM truly digested in rumen. The dietary N degradability (RDN) in rumen was calculated by the equation:

RDN (%) = N intake − (non-ammonia N flow to duodenum - microbial N flow to duodenum).

Comparison of the differences in the averages obtained from OG and MF pastures was made by t-test. The relationship between nutritional variables of herbage such as chemical composition and nutrients degradability, and N utilization in the rumen were estimated by correlation analysis.

3 Results and discussions

3.1 The chemical constituents of pasture Herbage mass and chemical compositions of herbage are summarized in Table 1. The pre-grazing average sward length of OG pasture was higher and herbage mass was lower than MF pasture. The herbage mass on OG and MF pastures ranged from 78 to 298 gDM/m^2 and from 92 to 294 gDM/m^2, respectively. The herbage allowance on OG and MF pastures ranged from 159 gDM/metabolic body size (MBS) to 436gDM/MBS and from 213 gDM/MBS to 525gDM/MBS, respectively. According to the Japanese Feeding Standard for beef cattle (MAFF, 2000), the predicted DM intake of dairy steers weighing 350 kg and growing at the rate of 0.8kg/day is 86.3 g/MBS, so it was thought that herbage intake was not restricted by the herbage allowance in this study. The OM content in the OG and MF pastures averaged 88.8% and 88.1%, respectively and there was no significant difference in OM content of herbage between pasture species. There were no significant differences in CP content in herbage between OG and MF pastures and the average content is 23.4 ± 2.5% and 25.0 ± 4.5% respectively. The CP content in herbage observed in this study was almost similar to the values obtained from OG pasture (Hoffman *et al.*, 1993; Kolver *et al.*, 1998), MF and timothy (*Phleum pratense*, L) pasture (Khalili and Sairanen, 2000; Sudo *et al.* 2001), perennial ryegrass (*Lolium perene*, L.) pasture (Beever *et al.*, 1986; Ulyatt *et al.*, 1988) and tall fescue (*Festuca arundinancea*, Schreb.) pasture (Berzaghi *et al.*, 1996; Elizalde *et al.*, 1998). The NDF content of herbage on OG and MF pastures averaged 48.7 ± 5.2% and 45.6 ± 4.0%, respectively and these values were within the range of average NDF content obtained from cool season grass pastures in the United States (Muller and Fales, 1998).

Table 1 Sward height, herbage mass and herbage allowance on OG and MF pasture and chemical composition of herbage plucked by hand to a level of post-grazing sward height

	OG pasture		MF pasture		SED[2)]	Difference[3)]
	Mean	SD[1)]	Mean	SD[1)]		
Sward height, cm	40.6	14.2	36.4	8.3	6.4	NS
Herbage mass, gDM/m^2	168.2	68.5	195.8	81.7	37.1	NS
Herbage allowance, gDM/MBS[4)]	71.8	18.5	87.3	36.6	58.4	NS
Chemical composition, % in DM						NS
Organic matter (OM)	88.8	0.8	88.1	0.8	0.4	NS
Neutral detergent fiber (NDF)	48.7	5.2	45.6	4.0	2.4	NS
Acid detergent fiber (ADF)	27.5	2.9	25.7	3.1	1.2	NS
Crude protein (CP)	23.4	2.5	25.0	4.5	1.7	NS
Non-protein fraction	3.6	0.8	4.1	1.0	0.5	NS
Soluble protein	6.7	1.8	6.8	2.0	1.0	NS
Neutral detergent insoluble protein	5.0	1.3	4.7	1.9	0.8	NS
Acid detergent insoluble protein	1.2	0.2	1.2	0.4	0.2	NS
Gross energy (MJ/kg DM)	19.1	0.5	19.6	0.4	0.2	NS

Note: 1) SD: Standard Deviation; 2) SED: Standard Error of Difference; 3) NS: $P > 0.05$; 4) MBS: Metabolic Body Size = (body weight)0.75.

3.2 OM intake and digestibility The results of DM intake and OM digestion in steers are shown in Table 2. The DM intake on OG and MF pastures averaged 91.8g/MBS/day and 87.5g/MBS/day, respectively and they exceeded the predicted DM intake of dairy steers weighing 350kg and growing at the rate of 0.8 kg/day (MAFF, 2000). There were no significant differences in OM intake between OG and MF pastures. Apparent and true OM digestibilities in the rumen were higher on MF pasture than on OG pasture ($P < 0.01$), but the amount of OM apparently or truly digested in the rumen did not differ between OG and MF pastures. The true digestibility of OM in the rumen (OMTDR) ranged from 51.4% to 66.4% in OG pasture and from 52.0% to 70.4% in MF pasture and there was a negative relationship between OMTDR and the NDF content in herbage ($r = -0.87$, $P < 0.01$). The amount of OMTDR ranged from 26.4g/MBS/day to 68.8g/MBS/day on OG pasture and from 27.6g/MBS/day to 75.5g/MBS/day on MF pasture. The OMTDR obtained on MF pasture in this study tended to be higher than the results of lactating cows grazing tall fescue pasture without supplements (Berzaghi *et al.*, 1996), and lower than the result of steers grazing tall fescue pasture without supplement (Elizalde *et al.*, 1998).

Table 2 Dry matter intake, organic matter intake and digestion of steers grazing OG and MF pastures without supplements

	OG pasture		MF pasture		SED[2)]	Difference[3)]
	Mean	SD[1)]	Mean	SD[1)]		
DM intake, g/MBS[4)]/day	91.8	16.6	87.5	16.5	4.8	NS
Organic matter (OM) intake and digestibility						
Intake, g/MBS[4)]/day	81.5	14.5	76.9	14.2	4.2	NS
Apparently digested in the rumen						
g/MBS[4)]/day	37.4	9.0	40.4	12.5	3.0	NS
% OM intake	45.6	6.1	51.7	9.5	2.1	**
Truly digested in the rumen (OMTDR)						
g/MBS[4)]/day	51.9	10.0	56.2	15.0	3.4	NS
% OM intake	56.6	5.4	63.5	7.9	1.8	**
Apparently digested in the total digestive tract, %	68.0	2.8	70.2	1.8	0.7	**

Note: 1) SD: Standard Deviation; 2) SED: Standard Error of Difference; 3) **: $P < 0.01$; NS: NO Significance ($P > 0.05$); 4) MBS: Metabolic Body Size = (body weight)0.75.

3.3 N and amino acid intake and digestibility Intake and passage of N, and ruminal microbial N synthesis in steers grazing on OG and MF pastures are shown in Table 2. There were no significant differences in N intake between OG and MF pastures, and the average N intake was more than twice as much as the N requirement for dairy steer weighing 350kg and growing at the rate of 0.8kg/day (1.56gN/MBS/day, MAFF, 2000). AA - N intake did not differ significantly between pastures and it accounted about 80% of total N intake. However, RDN was significantly higher ($P < 0.01$) in MF pasture than in OG pasture. The N degradability obtained in this study was lower than the result of steers grazing tall fescue pasture without supplement (Elizalde *et al.*, 1998). The ratio of RDN to ingested N was positively related with CP content in herbage ($r = 0.52$, $P < 0.01$) and negatively related to NDF content in herbage ($r = 0.61$, $P < 0.01$). Although the amount of OMTDR did not differ between pastures, the ratio of RDN to OMTDR on MF pasture was higher than that on OG pasture ($P < 0.05$). RDN and the ratio of RDN to OMTDR could be

estimated by a multi-regression equation as follows;

RDN (% of N intake) $= 1.25X_1 - 1.41X_2 + 91.9$, $r = 0.99$, $P < 0.01$

Ratio of RDN to OMTDR (g/kg) $= 2.81X_1 - 0.61X_2 - 0.55$, $r = 0.89$, $P < 0.01$

where X_1 = CP % in herbage, and X_2 = NDF % in herbage.

The conversion efficiency of RDN to microbial N on OG and MF pastures averaged 74.8% and 54.8%, respectively and there was a negative relationship between the conversion efficiency of RDN to microbial N and the ratio of RDN to OMTDR ($r = -0.74$, $P < 0.01$). The lower conversion efficiency of MF pasture compared with that of OG pasture was mainly due to the high ratio of RDN to OMTDR and it was thought that the high protein intake and degradability in the rumen and the low OMTDR intake were contributory factors to the low efficiency of N utilization in the rumen of cattle grazing temperate pasture. As has been the practice in the National Research Council (NRC, 2001), the conversion efficiency of RDN to microbial N of 85% is used to determine RDN requirement assuming an apparent ruminal N balance of zero. The N apparently absorbed in the rumen was greater on MF pasture than on OG pasture ($P < 0.01$) and it was positively related with the ratio of RDN to OMTDR ($r = 0.834$, $P < 0.01$, Fig. 1 – a) and ammonium N concentration in the rumen fluid ($r = 0.831$, $P < 0.01$, Fig. 1 – b). These relationships showed that the amount of apparent N absorption in the rumen would be positive when the ratio of RDN to OMTDR was above 24.7g/kg or the ammonium N concentration in the rumen fluid was above 8.7mg/100ml. As the averages of the ratio of RDN to OMTDR and the ammonium N concentration in the rumen were 37.2g/kg and 15.6 mg/100ml, it was suggested that some extent of N was usually absorbed in the rumen of cattle grazing OG or MF pasture without supplement. The ammonium N concentration in the rumen fluid on OG and MF pasture averaged 13.6mg/dL and 19.8mg/dl, respectively and there was higher on MF pasture than on OG pasture ($P < 0.01$), and it was positively related to the N concentration in the herbage ($r = 0.84$, $P < 0.01$) and the ratio of RDN to OMTDR ($r = 0.87$, $P < 0.01$). Thus, it appeared that the ammonium N concentration in the rumen fluid may be a useful parameter for evaluating the N utilization in the rumen of cattle grazing OG and MF pastures.

Table 3 N intake, flow to duodenum and digestion in different segments of the digestive tract of steers grazing OG and MF pastures without supplements

	OG pasture		MF pasture		SED[2]	Difference[3]
	Mean	SD[1]	Mean	SD		
Total N intake, g/MBS[4]/day	3.50	0.60	3.95	0.92	0.22	NS
Amino acid N intake, g/MBS[4]/day	2.83	0.31	3.17	0.91	0.13	NS
Amino acid-N, % of total N intake	80.90	5.90	79.30	8.10	1.70	NS
N degraded in the rumen (RDN), g/MBS[4]/day	1.67	0.47	2.20	0.99	0.21	**
RDN, % of N intake	46.20	13.20	59.80	12.80	3.80	*
RDN/OMTDR[5], g/kg	34.00	8.40	43.60	13.70	3.40	*
N apparently absorbed in the rumen, g/MBS[4]/day	0.50	0.65	1.11	0.97	0.22	**
Total N flow to duodenum, g/MBS[4]/day	2.87	0.59	2.42	0.43	0.16	**
Total N flow, % of N intake	86.60	18.00	72.50	19.90	5.40	**
Non-ammonium N (NAN) flow, g/MBS[4]/day	2.80	0.59	2.35	0.43	0.16	**
NAN flow, % of total N flow	97.30	1.20	96.80	1.40	0.17	NS
Ruminal undegraded N (RUN) flow, g/MBS[4]/day	1.41	0.19	1.34	0.27	0.04	NS
RUN flow, % of NAN flow	57.70	4.70	57.20	8.00	1.10	NS
RUN flow, % of N intake	46.80	8.40	40.20	13.20	1.80	*
Amino acid-N flow, g/MBS[4]/day	1.62	0.37	1.32	0.48	0.06	*
Amino acid-N flow, % of total N flow	59.50	7.90	57.40	6.10	1.70	NS
Microbial N synthesis in the rumen						
% of RDN	74.80	39.50	54.80	27.80	10.40	NS
g/kg of OMTDR[5]	22.20	6.50	21.50	7.50	2.00	NS
N digested post-ruminally						
g/MBS[4]/day	1.92	0.48	1.56	0.38	0.13	**

Note: 1) SD: Standard Deviation; 2) SED: Standard Error of Difference; 3) **: $P < 0.01$; *: $P < 0.05$; NS: NO Significance ($P > 0.05$); 4) MBS: Metabolic Body Size = (body weight)0.75; 5) OMTDR: Organic Matter Truly Digested in the Rumen; 6) Metabolizable protein was calculated using the method of AFRC (1993).

The total N and NAN flows to the duodenum significantly ($P < 0.01$) increased and the proportion of duodenum NAN flow to ingested N was higher ($P < 0.05$) in OG than in MF pasture. N flow to the duodenum increased with the increase of N intake, but did not always increase when the N intake was above 4.0g/MBS/day (Fig. 2), which was about 2.5 times the N requirement of

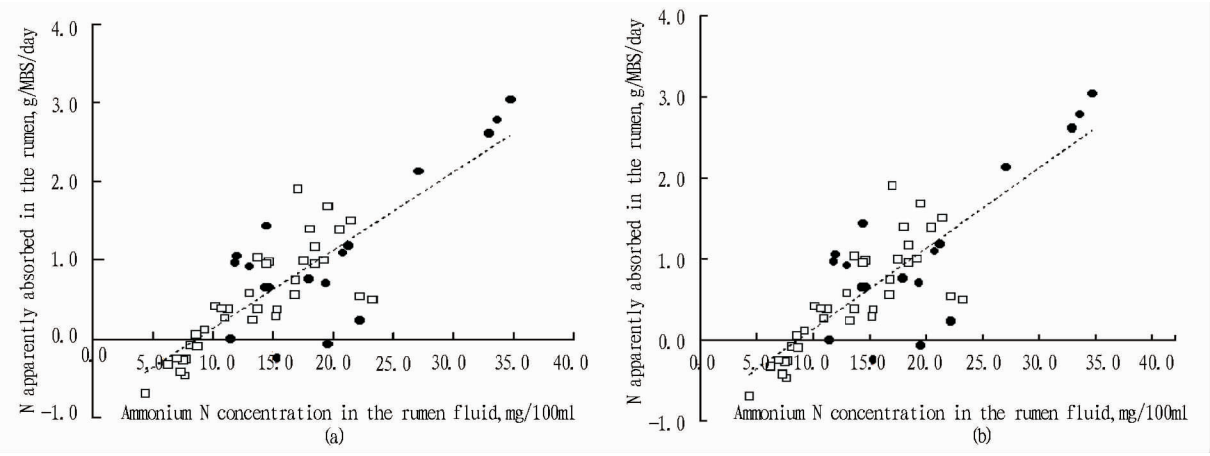

Fig. 1 Relationship between the ratio of RDN to OMTDR (a) and the ammonium N concentration in the rumen fluid (b) and the amount of N apparently absorbed in the rumen of steers grazing OG (□) and MF (●) pastures

dairy steers weighing 350kg and growing at the rate of 0.8kg/day (MAFF, 2000). Furthermore, these flows tended to decrease with the increase in the ratio of RDN to OMTDR ($r = 0.52$, $P < 0.01$). The proportion of the duodenum N flows to the ingested N averaged 86.6% on OG pasture and 72.5% on MF pasture. These recoveries was significantly higher ($P < 0.0$) on OG pasture than on MF pasture. Berzaghi *et al.* (1996) reported that the recovery of ingested N at the duodenum of lactating cows grazing tall-fescue pasture without supplement was 75% and the N recovery was improved by corn supplements. On the other hand, Elizalde *et al.* (1998) reported that the recovery of the ingested N at the duodenum of steers grazing on tall fescue pasture was about 72% and the N recovery was not improved by the energy supplementation. The low recoveries of ingested N at the duodenum on MF were probably due to the high ratio of RDN to OMTDR. There were negative relationships between the recovery of ingested N at the duodenum and the ratio of RDN to OMTDR ($r = -0.80$, $P < 0.01$) or the ammonium N concentration in the rumen fluid ($r = -0.75$, $P < 0.01$). Beever *et al.* (1986) also observed the negative relationship between the recovery of ingested N at the duodenum and the ammonium N concentration in the rumen fluid of steers grazing perennial ryegrass or white clover (*Trifolium repense*) pasture. We observed the duodenal N flow greater than the N intake in this study when the herbage CP was below 19% in DM. Ulyatt *et al.* (1988) also reported that the duodenal NAN flow was greater than the N intake when cattle grazed re-growth perennial ryegrass containing 12.6% of crude protein. The higher recovery of ingested N at the duodenum may be attributed to the recycling of N derived from saliva or rumen wall as a result of low ammonium N concentration in the rumen fluid. The high duodenal N flow and low N intake associated with low ammonium N concentration in the rumen fluid was also observed in steers fed corn gluten meal as a major protein source (Hanada *et al.*, 1993) and in lactating cows receiving high concentrate rations containing 15% of crude protein (McCarthy *et al.*, 1989).

AA-N flow to the duodenum was significantly related ($P < 0.$

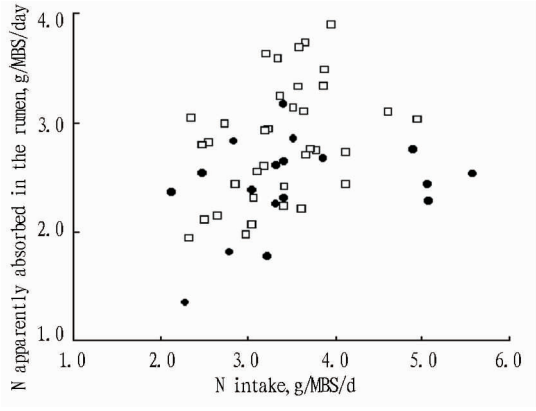

Fig. 2 Relationship between N intake and N flow to duodenum of steers grazing OG (□) and MF (●) pastures

01, $r^2 = 0.67$, Fig. 3) to duodenal NAN flows but it accounted for 60% of the NAN flows. Total AA flows to the duodenum were higher in OG than in MF but did not differ in AA composition (Table 4). There were apparent differences in essential AA composition between intake and duodenal AA flows (higher methionine and lysine), apparently enhanced by the AA profile of microbial protein outflow from the rumen. However, these essential AA seem to be insufficient for milk or growth because the AA profile of the duodenal digesta was less compared with those of the lean tissue and milk. The N digested post-ruminally was greater on OG pasture than on MF pasture ($P < 0.01$), however the apparent N digestibility through the lower gastrointestinal tract did not differ between pastures. The quantity of N digested post-ruminally did not increase with the increase of N intake (Fig. 4 – a), but tended to decrease with increase in the ratio of RDN to OMTDR (Fig. 4 – b) or the ammonium N concentration in the rumen fluid. The apparent N digestibility through the lower gastrointestinal tract obtained in this trial was higher than the result obtained from steers grazing tall fescue pasture without supplements (Elizalde *et al.*, 1998) and lower than the result obtained from lactating cows fed silage and concentrate mixed ration (McCarthy *et al.*, 1989).

Table 4 The amino acid intake and flow to duodenum and comparison of the amino acid composition of duodenal digestion with that of lean body tissue and milk

	Intake				Duodenal flow				Animal products	
	OD	SD	MF	SD	OD	SD	MF	SD	Lean tissue	Milk
Total amino acid, g/MBS/day	21.1	4.4	23.6	3.5	13.4	3.2	11.9	3.5	–	–
Amino acid composition, Mol% of total amino acids										
Essential animosity acids										
Arginine	3.73	0.14	3.52	0.08	3.17	0.10	3.17	0.09	4.85	2.55
Histidine	2.11	0.17	1.93	0.17	2.01	0.14	2.08	0.19	3.54	2.46
Isoleucine	4.68	0.06	4.62	0.14	5.03	0.16	5.07	0.29	4.83	5.49
Leucine	8.47	0.34	8.63	0.50	8.23	0.26	8.37	0.30	8.70	10.0
Lysine	4.78	0.52	4.57	0.43	5.99	0.21	6.03	0.34	8.53	7.53
Methionine	1.03	0.11	1.22	0.10	1.60	0.09	1.50	0.12	2.55	2.41
Phenylalanine	4.32	0.24	4.34	0.14	3.77	0.21	3.84	0.20	3.33	3.85
Threonine	5.58	0.28	5.60	0.20	5.98	0.14	6.01	0.24	5.32	4.62
Valine	6.78	0.12	6.73	0.14	6.24	0.16	6.28	0.37	5.59	7.41
Non – essential amino acids										
Alanine	11.64	0.36	11.01	0.42	10.16	0.23	10.35	0.44	9.01	4.99
Aspartate	11.53	0.97	10.68	0.70	10.41	0.27	10.54	0.43	9.68	7.95
Glutamate	10.99	0.41	11.54	0.33	10.73	0.23	10.88	0.46	14.2	17.3
Glycine	9.95	0.86	9.97	0.98	13.49	1.74	13.59	1.53	7.60	3.39
Proline	6.87	0.85	8.10	2.20	4.57	0.12	4.70	0.17	4.59	11.2
Serine	5.49	0.22	5.43	0.37	5.89	0.14	5.96	0.25	5.23	6.04
Tyrosine	2.05	0.11	2.12	0.17	2.71	0.16	2.62	0.14	2.45	2.80

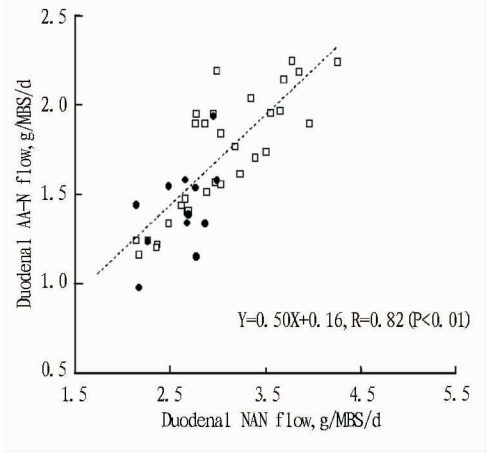

Fig. 3 Relationship between the non-ammonia nitrogen (NAN) flow to duodenum and amino acid nitrogen (AA-N) flow to duodenum of steers grazing OG (□) and MF (●) pastures

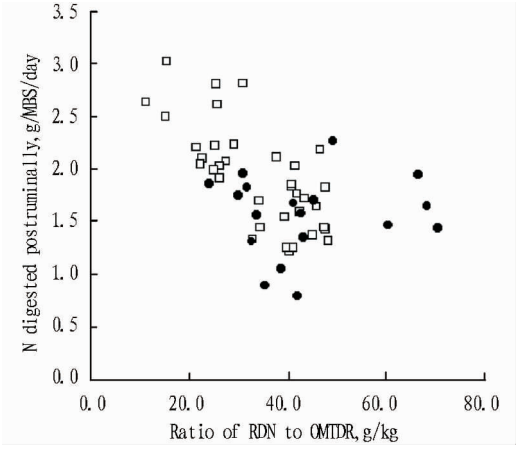

Fig. 4 Relationship between N intake (a) and the ratio of RDN to OMTDR (b) and the amount of N apparently digested in the post-rumen of steers grazing OG (□) and MF (●) pastures

The results of this study suggested that cattle grazed on these pastures may have lost some N in the rumen before reaching the duodenum. The recovery of ingested N at post-rumen depends more on the dietary N degraded in the rumen than the amount of N intake. Decrease the loss of N from the rumen and to supply more N to the small intestines, it is necessary to reduce the intake of N degraded in the rumen to OM truly digested in the rumen ratio supplement with grains feeds. Methionine and lysine were the most limiting AA for animal production from cows grazing temperate pastures.

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4.2 Transforming the agriculture development mode (i) Implementing conservation tillage. It is necessary to vigorously promote conservation tillage which can retain and conserve soil moisture, improve soil fertility and save agricultural fuels. Through the implementation of returning straw to farmland can effectively prevent and control straw burning and reduce carbon dioxide emissions. (ii) Optimizing the energy structure. It is necessary to reduce the fossil energy use in agricultural production and make full use of solar, wind and geothermal energy and other renewable energy sources; it is also necessary to vigorously promote the use of biomass, encourage the development of rural household biogas, and accelerate the construction of waste incineration power plant. (iii) Developing circular agriculture and ecological agriculture. It is necessary to promote interplanting-based three-dimensional crop planting mode, and increase the application of organic fertilizer to protect farmland ecosystems.

4.3 Promoting carbon sequestration technology (i) Improving the carbon-sequestrating agricultural varieties. China needs to develop or introduce the drought-resistant and pest-resistant crop varieties as well as new crops that have strong ability to absorb greenhouse gases. (ii) Developing agricultural carbon sequestration technologies. It is necessary to protect the existing carbon pools and use the ecosystem management techniques to strengthen the sustainable management of farming, animal husbandry, forestry and fishery, in order to maintain the long-term carbon sequestration capacity of ecosystems. (iii) Rationally applying chemical fertilizers. It is necessary to implement scientific fertilization and increase the application of organic fertilizer to reduce nitrous oxide emissions; use scientific irrigation technology to reduce methane emissions from rice paddies; carry out biogas utilization and control the growth of animal husbandry methane emissions. (iv) Updating the agricultural machinery and technology. It is necessary to eliminate the backward agricultural and fishing machinery and use advanced diesel fuel-saving technology to reduce the diesel fuel consumption.

4.4 Establishing the interests guiding mechanism (i) Setting up the carbon trading platform. It is necessary to establish the carbon trading market platform, build professional organizations, set up technical standards, and improve the supporting environmental capacity management and ecological compensation mechanisms. (ii) Achieving the sharing of interests among farmers. The formation of a reasonable interests sharing mechanism for farmers is an important way to promote the smooth implementation of agriculture carbon sequestration projects. After signing the carbon sequestration project order, the farmers' specialized cooperative economic organizations should be directly involved in the distribution of benefits from carbon trading to achieve efficiency increase.