FATE AND METABOLISM OF NITROGEN IN GOATS FED ^{15}N-LABELLED RICE STRAW

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Summary

Three goats were fed with ^{15}N-labelled rice straw to study the characteristics of digestion, assimilation, metabolism and excretion of C and N compounds from rice straw. It was shown that the amount of ^{15}N assimilated into the bodies of the two slaughtered goats accounted for 38.5 and 23.6 % of the total amount of ^{15}N ingestion in the experimental diet by each goat. The ^{15}N excreted via the feces and urine for the three goats accounted for 34.8, 33.2 and 33.9% of the total amount of ^{15}N ingested in the feed of the 3 goats. The recoveries of total ^{15}N from the two slaughtered goats were 73.3 and 57.5%, with the corresponding rates of ^{15}N loss 26.7 and 42.5% respectively. The amino acids digestibilities were 68.7 and 54.0%, while the digestibilities of carbohydrates were 74.8 and 67.7% respectively for goats 1 and 3.

(**Key Words**: Goats, Nitrogen Metabolism, ^{15}N-Labelled Rice Straw)

Introduction

In 1989 the overall cropping area of rice in China reached 32.7 million hectares with 180.13 million tons of rice grain produced (State Statistical Bureau, 1990). The ratio of grain to straw is normally about 1:1.1 and therefore, roughly 200 million tons of rice straw or 33.4 percent of the world production, are produced in China. The utilization rice straw was as follows: 66% as fuel, 20% as organic manure to be returned to the soils, and 12% as feed for domestic herbivous animals (FAO, 1983-1987). Numerous studies dealing with the effects of incorporation of crop straw to farmland on nutrient transformation and soil fertility have been reported (Mo, 1983; He et al., 1984; He et al., 1986; Cai et al., 1989; Wu and Ni, 1990), but there is little on the fate of nitrogen in the body of ruminants fed with ^{15}N-labelled rice straw.

In this experiment, ^{15}N-fertilizer was applied to rice plant, and the ^{15}N-labelled rice straw harvested was used as a feed stuff for goats. The extent of digestion, assimilation, metabolism and excretion of nitrogen by the goat fed rice straw were studied to provide scientific basis for developing a reliable model of nutrient cycling in the soil-crop-domestic animal ecosystem of subtropical regions where rice is the major crop.

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Materials and Methods

^{15}N-Labelled Rice straw

^{15}N-labelled urea (^{15}N 56.7%) was incorporated into the soil layer near rice plant roots at different growth stages. The rice vaviety is late-maturing rice so called V. You 64 (V_{64}). ^{15}N-labelled rice straw (3777.7 g) was derived from the rice plants that were harvested at the mature stage on October, 1990.

Experimental Animals

Three mature male Matou goats weighing 20.4, 14.2 and 12.8 kg were chosen as experiment animals.

Diet

Three types of mixed feed were compounded basal diet, pre-experimental diet and experimental diet. The components are listed in table 1. All types of diets were made from powdered material and extruded into particles.

Experimental design

TABLE 1. COMPOSITION OF DIETS FED TO GOAT (%)

Components	Basal diet	Pre-experimental diet	Experimental diet
Powdered maize	70.0		4.7
Maize starch		38.3	37.0
Soybean cake	7.0	3.8	4.2
Wheat bran	5.0	2.7	3.0
Powdered shell	0.8	0.4	1.2
Rice straw[a]	17.2	54.8	49.9

[a] Rice straw used in experimental diet was from ^{15}N-labelled (^{15}N 41.76%) rice (V_{84}). Unlabelled rice of the same variety were used for other diets.

The experimental goats were put into Dracher cages in the laboratory under the conditions of natural lighting and temperature. The feeding experiment consisted of three parts: (1) the pre-experimental period (6-7 days), (2) the experimental period (2-3 days), and (3) the post-experimental period (6-7 days), the total experiment lasted 14-17 days. Goat 1 was provided with basal diet 2800 g and experimental diet 1200 g; Goat 2 was provided with pre-experimental diet 3035 g and experimental diet 1292 g and Goat 3 was provided with pre-experimental diet 3036 g and experimental diet 1765 g. Each goat was fed 2-3 times every day, 100-200 grams once and in the period 3 supplemented with 100 grams fresh ryegrass each day. The goats feed intake and water *ad libitum*.

The excrements of Goat 1 were collected whenever excreted. The feces of Goat 2 and 3 were sampled once every four hours for eight days after the experiment began. They were weighed and stored in a refrigerator for use. Goat 1 and 3 were slaughtered, dissected and weighed, and the samples from different tissues and organs were collected in the morning after the feeding experiment ended. Each sample was minced prior to analysis.

Analytical method

^{15}N abundence was determined by Bremner's method (ZHT-1301 mass spectrometer). This procedure is as follows: (a) ^{15}N in samples (0.5 g) was changed into NH_4 via nitrification using the mixture of K_2SO_4-$CuSO_4$-Se catalyser; (b) Liquid nitrified was distilled using 10 N NaOH; (c) Distillate was titrated using 0.01 N H_2SO_4 and was also concentrated; (d) Distillate concentrated was changed into N_2 ($2NH_3 + 3NaOBr \rightarrow NaBr + 3H_2O + N_2$); (e) ^{15}N ratio value was determined by ZHT-1301 mass spectrometer (intensity of pressure: 1-3 10^{-7} mmHg; background value: 28 peak, 60-90 mV).

The composition of amino acid was determined using a Shimidzu LC-4A liquid chromatograph after hydrolysis with 6 M HCl. The contents of 15 amino acids in the three diets and in the feces were measured. The calculation of digestibility was based on a common method used in animal feed science, i.e. D = (A − B) A × 100% (D is the digestibility of certain component, A the amount of certain component in feed, and B the amount of certain component in feces).

Goering and Van Soest's method was used for determining neutral detergent fiber (NDF), acid detergent fiber (ADF) and lignin (LIG). The contents of cell soluble substances and hemicellulose (HC) were obtained from 100 NDF, and NDF-ADF respectively.

The potassium bichromate-sulphuric acid oxidizing method (Institute of soil science, Academia Ainica, 1987) was used for determining carbon in feed, excrements and animal samples.

Results and Discussion

Distribution of ^{15}N in goat's body

Of the three experimental goats, Goats 1 and 3 were slaughtered and dissected. Goats 1 and 3 took 1200 and 1292.5 grams of experimental feed containing 2501 and 2624 mg ^{15}N respectively. The distribution of ^{15}N in these goats were different due to the differences in age and weight of goat and components of feed. The results showed that: (1) The order of ^{15}N abundance in different tissues and organs of Goat 1 was liver (0.937%) > kidney (0.835%) > rumen (0.794 %) > small intestine (0.766%) > gall bladder (0.745%) > testicle (0.716%) > pancreas (0.707%) > reticulum-omasum-abomasum (0.689%), while the order for Goat 3 was small intestine (1.23%) > pancreas (1.16%) > liver (1.14%) > testicle (0.99%) > kidney (0.971%) > rumen (0.965%) > reticulum-abomasum (0.890%) > large intestine (0.875%) > spleen (0.855%); (2) The sequence of ^{15}N deposition for Goat 1 was muscle > bone structure > skin > rumen > liver > blood > small intestine; while the sequence for Goat 3 was muscle > bone structure > liver > small intestine > reticulum-omasum-abomasum > rumen > large intestines.

Table 2 showed the rates of ^{15}N recovered by different tissues and organs of the goats. The total ^{15}N recovered by Goat 1 was 964 mg, accounting for 38.5% of total ^{15}N in the feed. Of which 19.30% was recovered in the edible parts, including muscle and blood, and 3.4% in the head and hoof. Goat 3 contained 620 mg ^{15}N accounting for 23.6% of the total ^{15}N in the feed. Of which 15.5% was found in the edible parts, and 2.1% in the head and hoof. The ^{15}N abundance and the recovery of ^{15}N in different parts of Goat 1 were gound to be higher than those of Goat 3, because Goat 1 was older and had a heavier body and better quality of basal feed.

TABLE 2. DISTRIBUTION AND RETENTION OF ^{15}N IN VARIOUS OF GOATS FED ^{15}N LABELLED RICE STRAW

Sample	Organ weight (g)		^{15}N abundance (%, atoms excess)		^{15}N body deposition (mg)		^{15}N recovery (%, ingested)	
	Goat 1	Goat 3	Goat 1	Goat 3	Goat 1	Goat 3	Goat 1	Goat 3
Muscle	5,450	3,100	0.507	0.542	235	154	9.4	5.9
Blood	574	468	0.599	0.673	35	28	1.4	1.1
Viscera	2,574	1,965	0.756	0.972	221	237	8.8	9.0
Bone	3,200	1,050	0.608	0.575	223	66	8.9	2.5
Skin and wool	1,712	1,163	0.506	0.429	109	25	4.4	0.9
Head and hoof	1,560	1,220			85	56	3.4	2.1
Digesta in the intestine	2,783	2,216	0.823	0.819	56	54	2.2	2.1
Total					964	620	38.5	23.6

Dynamic changes of ^{15}N in goat's feces

^{15}N abundance in the fresh feces of Goat 1 was still equal to the background value (0.365%) 11 hours after the start of ^{15}N feeding. Thirteen and a half hours later, ^{15}N abundance in fresh feces reached 0.461%. Thus, ^{15}N began to be excreted 12-13 hours after the experiment started. ^{15}N abundance in fresh feces increased with time, and reached the peak value (7.73%) 60 67 hours later. The time covered by the ^{15}N abundance in the feces that was proximately equivalent to 2/3 of its peak value lasted about 46 hours, then came a gradual decrease of ^{15}N abundance. It still remained 1.2% abundance 6 days after ^{15}N feeding had stopped. Statistical analysis showed that the changes of ^{15}N abundance (Y_{1-1}) with the time (X) coincided with the following normal parabola distribution (figure 1):

$$Y_{1-1} = -0.00190\ X^2 + 0.250\ X - 1.65$$
$(r = 0.9085^{**},\ n = 20)$ ··························(1)

From figure 1, it can be found that ^{15}N abundance of fresh feces was roughly coincident with the regressive curve. According to Equation (1), it was predicted that ^{15}N abundance would have decreased to the backgound value 122.9 hours later. In fact, it was still 0.785% much

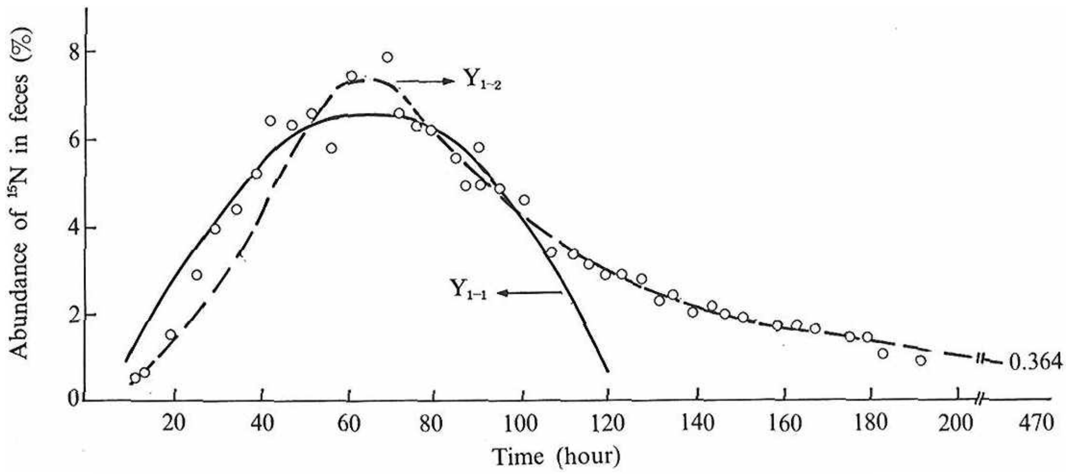

Figure 1. Dynamic changes of ^{15}N in feces of goat after feeding ^{15}N-labelled rice straw.

greater than the background value even 191 hours later. Moreover, another statistical analysis was done for the 42 samples of fresh feces excreted during the time of 11-191 hours after ^{15}N was initially fed, which showed that the correlation of ^{15}N abundance (Y_{1-2}) to the time (X) was:

$Y_{1-2} = X/(0.00747\ X^2 - 0.830\ X + 31.2)$ ($r = 0.9954^{**}$, $n = 42$)··························(2)

According to the characteristics of absorption and excretion of ^{15}N from Goat 1, it was decided that the fresh feces of Goat 2 and Goat 3 were collected once every four hours beginning from the time when ^{15}N was fed. The results showed the initial time for excretion of ^{15}N in Goat 2 and Goat 3 and the time when ^{15}N abundance in their feces reached the peak value were both different from those of Goat 1 (figure 2). During the time of 0-227.5 hours, the correlation of ^{15}N abundance (Y_2) in fresh feces of Goat 2 with the time (X) was:

$Y_2 = X/(41.7 - 1.08\ X + 0.00854\ X^2)$ ($r = 0.9809^{**}$, $n = 51$)··························(3)

and the correlation equation for Goat 3 during the time of 0-227.5 hours was:

$Y_3 = X/(35.1\quad 1.09\ X + 0.00954\ X^2)$ ($r = 0.9903^{**}$, $n = 55$)··························(4)

The rate of ^{15}N recovery of goat's feces (Y) increased with the time (X), which coincided with the following exponential regression equation:

Goat 1. $Y_1 = 52.0e^{-108.9/X}$ ($r = 0.9976^{**}$, $n=46$)

Goat 3. $Y_3 = 41.0e^{-83.4/X}$ ($r = 0.9916^{**}$, $n = 55$)

The total amount of experimental diet and the total amount of fresh feces which was taken or excreted by each goat were listed in table 3. The results showed that the total amount of ^{15}N deposition in goat's feces ranged from 677 to 769 mg, averaging 716 ± 47 mg. The recovery of ^{15}N in goat's feces ranged from 26.8 % to 29.5%, with an average of 27.8 ± 1.5%; i.e. twenty eight percent of ^{15}N in rice straw can be retrieved by goat's feces.

Dynamic changes of ^{15}N in goat's urine

^{15}N accumulation (^{15}N 0.391%) was found in the urine of Goat 1 only three hours after ^{15}N was initially fed. ^{15}N abundance in Goat 1 urine reached the peak value 45-55 hours later, which was 12 hours earlier than the time when the peak value of ^{15}N abundance occurred in the feces. However, the maximum value of ^{15}N in urine was only equivalent to 38.4 percent of the peak value of the feces, this demonstrated that the ratio of ^{15}N recovery was much lower in urine than in feces. ^{15}N abundace kept a value of approximately 2/3 of the peak value in the urine for about 46 hours, then gradually decreased. Until Goat 1 was slaughtered, ^{15}N abundance in the urine still remained 0.844%, which was equivalent to 28% of the peak value. This indicated that ^{15}N abundance decreased much more slowly in urine than in feces. Statistical analysis showed that during the time of 22-98 hours after ^{15}N was fed, the changes of ^{15}N abundance in the urine of Goat 1 (Y_{1-3}) with time coincided with a

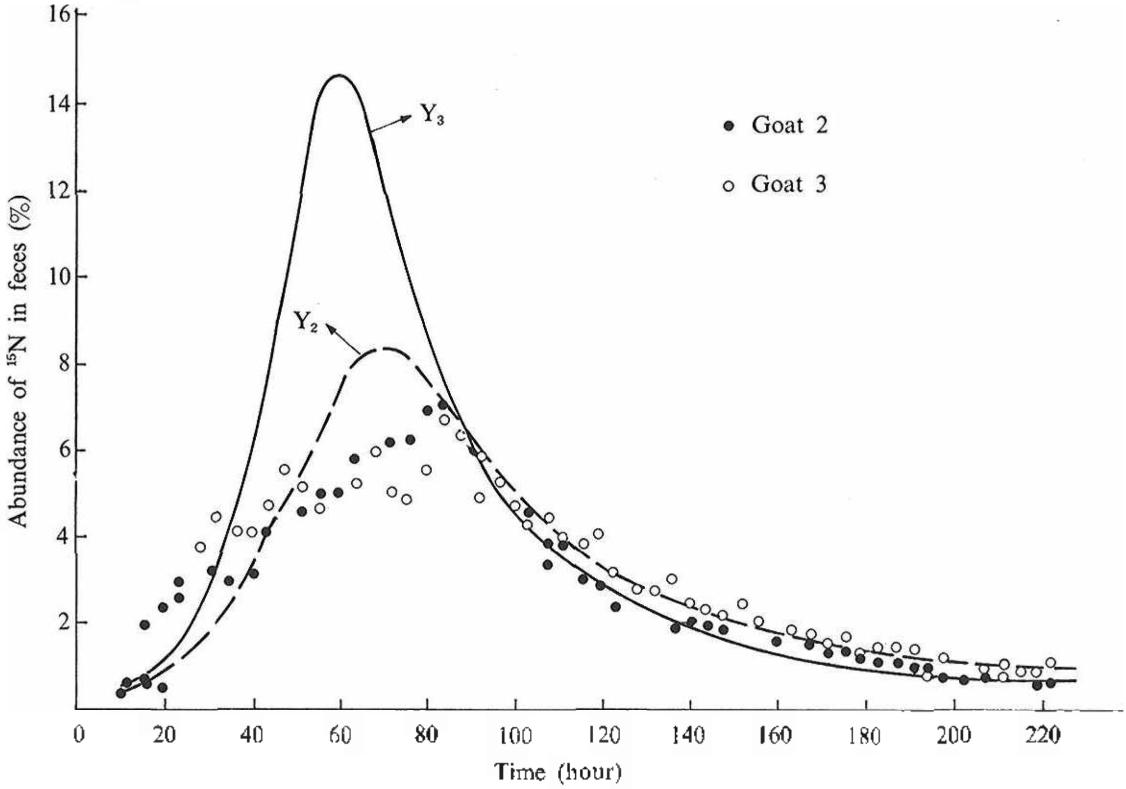

Figure 2. Dynamic changes of ^{15}N in feces of goat after feeding ^{15}N-labelled rice straw.

TABLE 3. AMOUNTS OF INGESTED APPARENTLY ABSORBED, RETAINED AND EXCRETED IN THE FECES AND URINE OF GOAT FED ^{15}N-LABELLED RICE STRAW

Goat No.	Experimental diet (g)	^{15}N in feed (mg)	^{15}N in feces (mg)	^{15}N apparently absorbed in feces (mg)	^{15}N recovery in feces (%)	^{15}N urine (mg)	^{15}N apparently ratain in urine (mg)	^{15}N recovery in urine (%)
1	1,200	2,502	677	1,825	27.1	193	1,632	7.7
2	1,285	2,609	769	1,840	29.5	97	1,743	3.7
3	1,292	2,624	703	1,921	26.8	186	1,735	7.1
Mean	1,259	2,578	715	1,862	27.8	159	1,703	6.2

gentle parabola (figure 3), the regression equation was:

$Y_{1-1} = 0.000553X^2 - 0.0648X + 0.472$
($r = 0.6763^*$, $n = 12$) ·· (5)

If the whole time of the experiment was taken into account, the dynamic changes of ^{15}N abundance in the urine with time for each goat were coincident with following regression equation:

Goat 1. $Y_{1-2} = X/(0.00667X^2 - 0.133X + 11.4)$ ($r = 0.9985^{**}$, $n = 22$) ························ (6)

Goat 2. $Y_{2-2} = X/(23.6 - 0.640X + 0.00764X^2)$
($r = 0.9948^{**}$, $n = 37$) ··································(7)

Goat 3. $Y_{3-3} = X/(17.8 - 0.253X + 0.00600X^2)$
($r = 0.9395^{**}$, $n = 40$) ··································(8)

The recovery of ^{15}N in urine changed with time (figure 4), which was also coincident with exponential regression equation. For instance, the equation for Goat 1 was: $Y_1 - 5.28e^{-26/X}$, r =

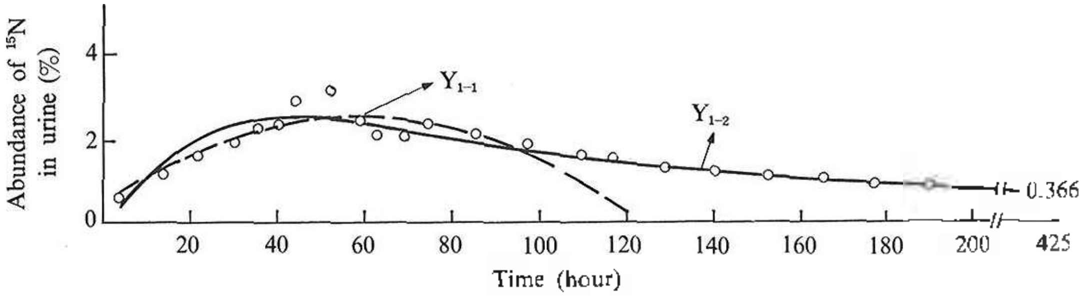

Figure 3. Dynamic changes of ^{15}N in urine of goat after feeding ^{15}N-labelled rice straw.

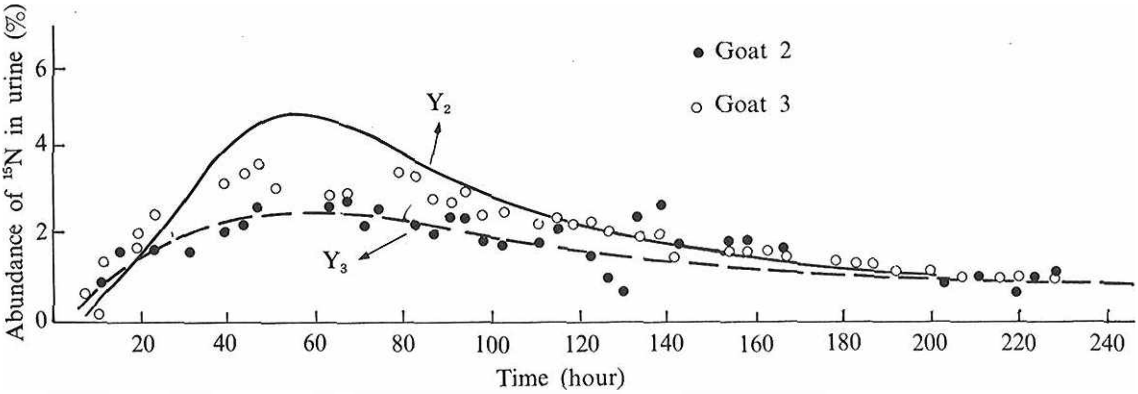

Figure 4. Dynamic changes of ^{15}N in urine of goat after feeding ^{15}N-labelled rice straw.

0.8892**, n = 22. The Total ^{15}N deposition in urine for each goat is listed in table 3. ^{15}N abundance was lower in urine than in feces. The total amounts of ^{15}N deposition in goat's urine ranged from 97 to 193 mg, averaging 159 ± 54 mg. Total ^{15}N recovery in urine ranged from 3.7-7.7%, with an average of 6.2 ± 2.2%.

The digestibilities of amino acids in the feed

In order to clarify the digestibilities of amino acids in the experimental diet in which rice straw was a dominant component, the contents of 15 amino acids in the three diets and in the feces were measured. The digestibilities of different amino acids for each type of feed are given in table 4.

It showed that the digestibilities of total amino acids in basal diet, pre experimental diet and experimental diet were 80.5%, 57.1% and 61.3% respectively. Those results were coincident with the composition of each type of diet.

The digestibilities of cellulose and other carbohydrates in the feed

Rice straw contains not only a certain amount of N-compounds but also large amounts of cellulose and other carbohydrates. In order to know the digestibilities of carbohydrates not acceptable in the goat's feed, NDF, ADF, lignin and the contents of N and C in each diet, feces and contents of rumen were determined (table 5). It was observed that: (1) The contents of cellulose and lignin were lower in basal diet than in both pre-experimental diet and experimental diet, but the content of N in basal diet was higher than that in the other two diets, so the C/N ratio of basal diet was much lower than that of the other two diets. However C/N of the feces of Goat 1 fed with basal diet was similar to that of the feces of Goats 2 and 3 fed with the other two diets. (2) Although Goat 1 was fed with basal diet and experimental diet, whereas Goat 2 and 3 were fed with pre-experimental diet and experimental diet, and the C/N ratio and composition

TABLE 4. THE DIGESTIBILITIES OF VARIOUS AMINO ACIDS IN FEEDS FOR GOATS

Amino acid	Digestibility (%)			
	basal diet	pre-experimental diet	Experimental diet	
			Goat 1	Goat 2
Asp	81.60	47.75	70.78	58.43
Thr	74.59	31.12	55.37	41.38
Ser	82.14	44.59	64.91	53.42
Glu	89.59	57.23	75.20	59.60
Pro	60.70	62.02	78.68	67.55
Gly	76.06	39.11	62.46	46.91
Ala	81.98	26.05	61.01	37.44
Val	79.64	63.70	72.91	54.83
Met	81.67	80.62	71.92	57.57
Ile	73.12	18.43	63.52	48.57
Leu	81.79	51.13	68.06	51.17
Phe	77.69	62.39	67.69	53.54
His	75.21	59.90	71.35	61.96
Lys	79.73	55.63	57.76	40.25
Arg	80.14	72.47	57.01	52.10
Total[a]	80.5	57.1	68.7	54.0

[a] The digestibility of total amino acids in the feed.

of the feed varied greataly between basal diet and experimental diet, the contents of cellulose and lignin and the C/N ratio of feces excreted by each goat were very close to each other after different diets were digested by the goats. This suggested different abilities of the goats to digest C and N compounds. (3) The C/N ratios of the digesta in stomach and intestines of Goat 1 and 3 were 41-81% lower than the three types of diets. This was because the digestible carbohydrates in the feed was assimilated and excreted as gaseous substances out of goat's body, which induced a decrease of C and a relative increase of N. (4) The diets with identical composition were fed to Goats 2 and 3. The digestibilities of different components in the feed were arranged as the following sequence: NDF (56.4 ± 4.9%) 〉 ADF (45.7 ± 3.9%) 〉 Lignin (3.0 ± 4.1%). (5) Goat 1 had higher digestibilities of NDF and C-compounds than Goats 2 and 3, because the contents of NDF and HC in basal diet were 10.1% and 5.0% higher than those in pre-experimental diet respectively. Thus, the digestibility of C-compounds in the feed for Goat 1 (74.75%) could be considered as the average digestibility of basal diet and experimental diet, and the average digestibility of C-compounds in the feed for Goat 2 and 3 (65.5 ± 2.5%) could represent the digestibility of experimental diet in which rice straw was a dominant component. These were coincident with the results obtained from the calculation of ^{15}N abundance.

Based on these results mentioned above, it may be found that for Goats 1 and 3 the recovery of ^{15}N in each goat's body was 38.5% and 23.6%, averaging 31.1 ± 10.5%; and the rates of total ^{15}N recovery for each goat were 73.3% and 57.5%, averaging 65.4 ± 11.2%. The recovery of ^{15}N in rice straw by feeding to goats was much higher than that by returning directly to soils. He et al. (1984; 1986) studied the fate of nitrogen in rice straw by using ^{15}N labelled rice straw incorporated directly to both gleyed paddy soil and normal paddy soil. The results showed that the recovery of ^{15}N by the rice plant was 19.4-23.1%, and the rate of ^{15}N remaining in the soil was 34.9-57.1% when only rice straw was applied; they were 25.1-29.6% and 37.7-61.2% respectively when 1/2 ^{15}N rice straw and 1/2 ^{14}N ammonium sulphate were jointly applied. It could be seen that more ^{15}N from rice straw was recovered by goat than by rice plant, and the economic value

TABLE 5. THE CONTENTS OF C AND N IN GOAT'S FEED AND FECES AND DIGESTIBILITIES OF CELLULOSE AND OTHER CARBOHYDRATES

Feed or Feces		NDF	ADF	LIG	N	C	C/N
		(g/kg)					
Basal diet		362	84	9.4	21.5	407	18.9
Pre-experimental diet		462	235	43.5	8.6	410	47.5
^{15}N-experimental diet		410	190	42.7	9.5	417	43.9
Feces	Goat 1[a]	483	273	109	20.3	374	18.4
	Goat 3[a]	518	312	106	19.0	358	18.8
	Goat 2[a]	490	313	111	18.5	360	19.5
	Mean	497	299	109	19.3	364	18.9
Gontents of rumen	Goat 1	513	297	129	29.1	325	11.2
	Goat 2	456	316	127	34.1	304	8.9
	Mean	485	307	128	31.6	315	10.1
Digestibility of the feed (%)	Goat 1	66.1	38.0	tr.	–	74.8	–
	Goat 2	59.9	48.4	5.9	–	67.2	–
	Goat 3	52.9	42.9	0.1	–	63.7	–
	Mean for Goat 2 and 3	56.4	45.7	3.0	–	65.5	–
Digestibility of basal diet for goat (%)		70.9	30.6	tr.	–	78.4	–

[a] 1055 g feces excreted by Goat 1;1582 g feces excreted by Goat 2 and 3060 g feces excreted by Goat 3.

of goat's body is much higher than that of rice grain and straw.

Concerning the causes for N loss, Wu and Ni (1990) quoted the data of Frissel (1978) to explain why 30.7% N was lost when ^{15}N-labelled ryegrass was fed to rabbits, and they estimated that about 12% N was lost from the animal's body in the digestion process, and about 20% N loss through ammonium volatilization. Denmed et al. (1974), Dean et al. (1975) and Frissel (1978) all attributed the causes of N loss to NH_3 volatilization and the N-compounds loss via the feces excretion. It seems to be possible, but the mechanism of N loss remains to be further studied.

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