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Nitrogen conversion and apparent intestinal amino acid absorption in young bulls fed isonitrogenous diets with different nitrogen sources

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ABSTRACT

The experiment was carried out on 10 bulls of about 120 kg live weight equipped with rumen cannulas and re-entrant cannulas in the proximal duodenum and terminal ileum. The bulls were given a diet of maize whole plant meal, potato starch and mineral mixture. The diet was supplemented for group I with groundnut oilmeal (60% of dietary N), for group II with groundnut oilmeal (10.5% dietary N) and urea (48% of dietary N), and for group III with urea (58% dietary N). Diets I, II and III contained, according to the INRA system, 15% CP in DM; 234, 196, 186 g PDIN; 227, 158, 145 g PDIE, and 2.14, 2.11, 2.11 UFV, respectively. Daily feed intake was 2.25 kg.

The ammonia-N level in the rumen fluid 2 h after feeding was 8.2, 15.1 and 24.5 mg/100 ml in animals of groups I, II and III, respectively. The amount of N entering the duodenum was 20.1; 22.1 and 18.0 g, leaving the ileum 9.7, 10.4 and 7.3 g/day/kg of feed intake, in the respective groups. The apparent duodenal-N absorption from the small intestine was 52, 53 and 59%, respectively.

Total amino acids (g/day/kg feed) ingested with feed equalled 128, 62 and 49; entering the duodenum 107, 113 and 92; leaving the ileum 43, 48 and 32; absorbed from the small intestine 64, 65 and 60 in groups I, II and III, respectively. The apparent intestinal absorption of most amino acids entering the duodenum was from 50 to 70% in all groups, but absorption of cysteine was markedly lower, 25-40%. The amount of lysine absorbed from the small intestine in group I was similar to the amount ingested, but exceeded this amount by 2.4 and 3 times in groups II and III. Absorption of methionine was two times higher, indicating considerable conversion of dietary N in group II and III into microbial amino acids.

KEY WORDS: amino acid, absorption, intestine, nitrogen source, ruminant

INTRODUCTION

The amount of amino acids absorbed in the small intestine of ruminants depends on the quality and amount of ingested protein and its degradability in the rumen as well as on the source of energy that is easily available to microorganisms in the rumen (Ørskov, 1992; Pająk et al., 1992; Mebjeesh et al., 1996; van Straalen et al., 1997). Feed additives or treatment protecting protein from rumen degradation and technological feed processing also affect ileal amino acid digestibility (Kowalczyk and Otwinowska, 1982; Benchaar et al., 1994; Waghorn et al., 1994).

The objective of the present study was to estimate the amount of amino acids leaving the abomasum and their apparent digestibility in the small intestine of young bulls fed an isonitrogenous ration, but with different amounts of amino acids and non-protein nitrogen proportions.

MATERIAL AND METHODS

The experiment was carried out on 10 young Black-and-White bulls of about 120 kg liveweight equipped with permanent simple cannula into the rumen and with re-entrant cannulas situated at the proximal duodenum and terminal ileum.

The bulls were divided into three groups and given a basic diet of maize whole plant meal, potato starch and mineral mixture. The diet was supplemented for group I with groundnut oilmeal (60% of dietary N), for group II with groundnut oilmeal (10.5% dietary N) and urea (48% of dietary N), and for group III (4 animals) with urea (58% of dietary N). The dietary components and chemical composition are presented in Table 1. Diets I, II and III contained, according to the INRA (1989) system, about 15.5% CP in DM; 234, 196, 186 g PDIN; 227, 158, 145 g PDIE; and 2.14, 2.11, 211 UFV in 1 kg of the respective diets. Daily feed intake was 2.25 kg given in two equal portions at 7.30 a.m. and 4.30 p.m.

After feeding with the respective diets for two weeks the animals were housed in metabolic cages and a digesta sampling period lasting 3 consecutive days was started. Samples of rumen liquor were taken before and 2 and 4 h after the morning feeding for determination of pH, ammonia-N and volatile fatty acids. The digesta flowing into the duodenum and leaving the ileum was measured directly continuously in a 3-day collection and returned into the duodenum or ileum after taking samples of 3% from the duodenum and 5% from ileum for analysis. The daily bulked samples of digesta were analysed for pH, ammonia-N in fresh material, while the samples for determination of DM, total N, VFA and amino acid composition were frozen until analysis.

pH was measured potentiometrically, ammonia-N with the Conway (1962) diffusion method, total volatile fatty acids by steam distillation from solutions acidi-

TABLE I

Components and chemical composition of rations, %

	Group		
	I	II	III
Feeds			
dehydrated maize (whole plants)	76.0	76.0	76.0
groundnut oilmeal	18.0	3.0	–
potatoe starch	4.5	17.2	19.7
mineral mixture	1.5	1.5	1.5
urea	–	2.3	2.8
Chemical composition			
dry matter	92.59	92.27	92.21
ash	5.98	4.99	4.80
crude protein	15.98	15.48	15.31
protein from natural feeds	15.98	8.05	6.45
urea	–	2.55	3.04
ether extract	2.13	1.97	1.94
crude fibre	14.79	13.59	13.35
N-free extractives	61.12	68.85	70.42

fied with sulphuric acid, amino acids in feeds and digesta were determined with a Beckman Unichrom amino acid analyser after previous 6 N HCl hydrolysis of samples, methionine and cysteine after previous oxidation with performic acid.

RESULTS

The pH values, ammonia-N and volatile fatty acid concentrations in rumen liquor indicated that the nutrients entered normal metabolic pathways (Table 2).

TABELA 2

Ammonia-N(mg/100mL), VFA (mmol/L) concentration and pH value in the rumen liquor, (0 – before feeding, 2 and 4 h after feed administration)

Component	Group								
	I			II			III		
	sampling time								
	0	2	4	0	2	4	0	2	4
NH_3-N, mg/100 mL	8.1	8.2^{A}	4.1	6.1	15.1^{B}	5.8	6.0	24.5^{C}	5.0
VFA, mmol/L	73.2	87.6	79.2^{a}	78.6	90.0	89.5^{b}	80.4	94.4	94.6^{b}
pH	6.96	6.08	6.57	6.67	6.42	6.23	7.06	6.43	6.06

A, B, C – $P<0.05$; a, b, - $P<0.05$ between the groups 2 or 4 h after feeding

The ammonia level in the rumen liquor at 2 h after feeding diets containing urea increased ($P < 0.01$) as the amount of urea in the diet rose. Volatile fatty acids increased slightly ($P > 0.05$), corresponding to the rise in the level of easily fermentable starch.

In all of the groups the pH of duodenal and ileal digesta was similar and ranged from 2.2 to 2.5, and from 7.9 to 8.2, respectively, indicating normal function of the abomasum and small intestine. The amount of ammonia-N flowing with the digesta into the duodenum was small in animals of all groups, about 0.5 g/kg ingested ration.

The apparent digestibility of dry matter in the forestomachs was highest in group III, about 46% ($P<0.05$) and lowest in group II, about 31%, disappearance of total nitrogen in the forestomachs was most pronounced in animals of group III, 27% ($P>0.05$), least in group II, 24% (Table 3).

TABLE 3

Dry matter and total-N ingested, entering duodenum and leaving ileum (g/day/kg feed intake), and their apparent absorption in the forestomachs or in the small intestine, %

		Group		
		I	II	III
Ingested:	dry matter	923	923	922
	total-N	25.6	24.8	24.5
Entering duodenum:	dry matter	519^{A}	636^{B}	494^{A}
	total-N	20.1	22.1	18.0
Leaving ileum:	dry matter	408^{A}	484^{B}	348^{C}
	total-N	9.7^{a}	10.4^{a}	7.3^{b}
Apparent digestion:				
in forestomachs	dry matter	43.8^{b}	31.1^{a}	46.4^{b}
	total-N	25.4	23.9	26.5
in small intestine	dry matter	21.5^{a}	23.9^{a}	29.5^{b}
	total-N	52.0^{a}	52.9^{a}	59.4^{b}

a, b, c – $P<0.05$; A, B, C – $P<0.01$

The amount of total-N passing from the abomasum to duodenum (Table 3), expressed in grams per kilogram of ingested ration, was largest in bulls of group II and lowest in those from group III ($P>0.05$). The apparent digestibility of total-N and dry matter in the small intestine was highest in animals of group III and lowest in group I ($P<0.05$). The total amount of amino acids flowing to the duodenum (Table 4) in group II was similar to that in group I in spite of the bulls of this group ingesting considerably less ($P < 0.01$) amino acids than those in group I. In group I,

TABLE 4

Total amino acids ingested, entering duodenum, leaving ileum (g/day/kg of feed), and apparent absorption from small intestine, %

	Group		
	I	II	III
Amino acids, g/day/kg of feed			
ingested with feed	128^{A}	62^{Ba}	49^{Bb}
entering the duodenum	107^{a}	113^{a}	92^{b}
leaving ileum	43^{a}	48^{a}	32^{b}
Absorption of amino acids in intestine, g	64	65	60
Aminoacids entering duodenum related to ingested with feed, %	84	182	187
Absorption of aminoacids in intestine, %	60	57	65
Proportion of amino acids absorbed in small intestine to ingested, %	50	104	122

a, b – $P<0.05$; A, B – $P<0.01$

total amino acids flowing with duodenal digesta were 16% less than ingested, 82% less in group II, and 87% more in group III.

The apparent absorption of total amino acids from the intestine (g/day/kg feed) was slightly higher in groups I and II than in group III ($P > 0.05$), but when expressed as a per cent of the amount of amino acids entering the duodenum, it was highest in group III and lowest in the animals of group II (Table 4). The amount of total amino acids apparently absorbed from the small intestine compared with their intake was 50% less in group I, but 4 and 22% more than ingested in group II and III (Table 5).

Apparent absorption from the small intestine of most individual amino acids (Table 5) was from 50 to 70% in all groups but absorption of cystine was markedly lower, 25-40%. The amount of lysine apparently absorbed from the small intestine in group I was similar to the amount ingested, but in group II and III exceeded its intake by 2.4 and 3 times. Absorption of methionine in groups II and III was twice as high, but in group I, about 20% less than ingested.

DISCUSSION

The pattern of changes of pH values as well as the concentration of ammonia and volatile fatty acids in ruminal, duodenal and ileal digesta reflected the diet compositions and indicated that the digestive tracts of the animals in all of the groups were functioning normally. The significant increase in the ammonia con-

TABLE 5

The apparent absorption of individual amino acids from the small intestine and their proportion to ingested

Amino acid	Absorbed from intestine: leaving abomasum, %			Absorbed: ingested with feed, %		
	group			group		
	I	II	III	I	II	III
Asp	62	60	68	51	126	155
Thr	57	54	62	77	144	157
Ser	57	55	66	49	106	138
Glu	61	59	66	38	88	105
Pro	49	50	62	34	60	77
Gly	52	50	61	43	96	123
Ala	58	58	64	66	118	118
Val	55	51	59	56	100	105
Ile	60	57	62	67	129	126
Leu	62	59	69	53	88	100
Tyr	69	64	70	66	132	153
Phe	63	58	66	51	100	120
Lys	68	66	72	109	240	293
His	57	54	65	42	85	107
Arg	68	65	72	30	85	126
Cys	29	24	40	23	32	57
Met	67	69	71	78	151	149
Trp	47	44	50	53	117	128
Total AA	60	57	65	50	104	132

centration in the rumen liquor 2 h after feeding the animals of group II and III was caused by, respectively, the high content of rapidly hydrolysable urea in the diet (III, 58%; and II, 48% of urea origin nitrogen) delivering ammonia for growth of microorganisms. Only a small part of the ammonia produced in the rumen reached the duodenum, suggesting that most of it was converted into microbial protein or absorbed in the forestomachs.

The apparent digestibility of dry matter in the reticulo-rumen differed among the groups, ranging from 31% in group II to 46% in group III. The reason for such a wide variation is difficult to explain on the basis of the data obtained from the experiment. The results of respective nitrogen digestion were more consistent and ranged from 24 to 27% despite expectations that nitrogen from the forestomachs of animals fed diets with urea would disappear at a faster rate. The possible explanation may be that nitrogen of urea origin was effectively incorporated into microbial protein.

Data concerning the apparent digestibility of nitrogen in the forestomachs found in numerous papers usually oscillates in a wide range between -30 to +30% (Otwinowska and Kowalczyk, 1985; Waltz et al., 1989; Beever et al., 1990; Coomer et al., 1993). The reason for such wide dispersion of nitrogen apparent digestibility is the level and quality of nitrogen compounds in the diet. Negative values of digestibility are usually obtained when intake of nitrogen compounds is low and the inflow of nitrogen into the rumen with saliva and across the rumen wall from the blood prevails over its absorption from the reticulo-rumen. In cases of excess dietary nitrogen intake the opposite situation occurs and apparent digestibility in the reticulo-rumen is positive. In our experiment, with a large proportion of easily degradable groundnut protein and non-protein nitrogen in the diet, the apparent digestibility of nitrogen in the reticulo-rumen was rather high in all groups of animals, reaching a value of about 25%, suggesting excess nitrogen intake.

Apparent digestibility of dry matter in the small intestine increased with the level of urea in the diet, rising from 22% in group I to 30% in group III and corresponded to the digestibility of nitrogen (52 to 59%) in this part of the digestive tract (Table 3). Waltz et al. (1989) and Beever et al. (1990) reported a significant effect of protein source (maize silage, fish meal, soyabean meal, heated soyabean meal, maize gluten meal, blood meal or combinations of them) on intestinal dry matter and nitrogen digestibility in cattle fed diets with different high protein feeds. Protein of lower digestibility in the reticulo-rumen, e.g. fish meal, blood meal or heated soyabean meal was usually more digestible in the small intestine. A similar effect was found in our experiment with young bulls fed groundnut oilmeal or its substitute, urea.

The similar amounts of total amino acids entering the duodenum of animals in groups I and II suggest considerable conversion of urea nitrogen into microbial amino acids in bulls of group II. The smallest amount of total amino acids flowed through the duodenum of animals fed the diet in which 58% of nitrogen was in the form of urea and which supplied the lowest amount of amino acids. The reason for this was probably that the proportion of non-protein nitrogen in diet III exceeded the capability of the rumen population to convert it into microbial protein. The higher level of ammonia in those animals 2 h after feeding supports this explanation because a higher rumen ammonia level favours absorption of this compound from the rumen more than its conversion into bacterial protein. However, the efficiency of amino acid absorption from the small intestine of animals with the smallest amount of amino acids entering the duodenum was higher than in the other groups, perhaps due to the higher efficiency of enzymatic protein digestion.

The amount of total and individual amino acids apparently absorbed from the small intestine compared to the intake of amino acids, particularly of lysine which exceeded intake 2.4 and 3 times, and methionine-about two times (Table 5), con-

firms the relatively high conversion of non-protein nitrogen into amino acids even at high proportions, above 50%, of urea-nitrogen in the diet.

Apparent absorption from the small intestine of most of individual amino acids (Table 5) was from 50 to 70% in all groups, but absorption of cystine was markedly less. The range of absorption of individual amino acids estimated in our experiment is similar to reported by other authors (Waltz et al., 1989; Hussein et al., 1991; Kerry et al., 1992; Cecava et al., 1993; Benchaar et al., 1994 a,b). The low apparent absorption of cysteine from the small intestine found in our experiment (Table 5) agrees with the findings of other authors (Bowman and Paterson, 1988; Van Bruchem et al., 1989; Cecava et al., 1993; Schröder et al., 1997; Żebrowska et al., 1997). The reason for this is not clear, however. A possible partial explanation could be that endogenous cysteine may be delivered to the small intestine with digestive enzymes such as trypsin and chymotrypsin, which contain three to four times more cysteine than most feed or bacterial proteins (Tamminga, 1979) or from desquamation of the epithelium, which contains a rather high level of cysteine (Żebrowska and Kowalczyk, 1990).

It can be concluded that replacement of 50% of dietary nitrogen by urea nitrogen in diets for growing bulls has no adverse effect on the amount of amino acids absorbed from the small intestine when the diet contains enough energy easily available for bacteria. With 60% of dietary nitrogen in the form of urea, the amount of amino acids entering the duodenum is reduced but their digestibility in the small intestine is elevated. Nitrogen compounds of diets containing a high proportion of non-protein nitrogen are converted into microbial protein to a substantial extent, with a concomitant, significant increase of lysine and methionine entering the duodenum over their amount in the ingested feed.

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STRESZCZENIE

Konwersja azotu i wchłanianie aminokwasów w jelicie cienkim buhajków żywionych dawkami izoazotowymi o zróżnicowanych źródłach azotu

Doświadczenie przeprowadzono na 10 buhajkach o masie ciała około 120 kg z trwałymi przetokami do żwacza oraz mostkowymi w początkowym odcinku dwunastnicy przed ujściem przewodu żółciowo-trzustkowego oraz na końcu jelita biodrowego. Zwierzęta otrzymywały dawkę złożoną z rozdrobniolego suszu z całych roślin kukurydzy, skrobi ziemniaczanej i mieszanki mineralnej. Dawkę uzupełniono w grupie I śrutą arachidową (60% azotu dawki), w drugiej – poekstrakcyjną śrutą arachidową (10,5% azotu dawki) i mocznikiem (48% azotu dawki), w III – mocznikiem (58% azotu dawki). Dawki I, II i III zawierały, zgodnie z systemem INRA, 15% białka ogólnego w suchej masie; 234, 196 i 186 g BTJN; 227, 158 i 145 BTJE oraz 2,14; 2,11 i 2,11 JPŻ. Dzienne pobranie paszy wynosiło około 2.25 kg.

Poziom NH_3-N w płynnej treści żwacza wynosił 8,2; 15,1 i 24,5 mg/100 ml u zwierząt z grup I, II i III, odpowiednio. Ilość azotu ogólnego przechodząca do dwunastnicy wynosiła, odpowiednio w kolejnych grupach, 20,1; 22,1 i 18,0 g, a opuszczającego jelito biodrowe 9,7; 10,4 i 7,3 g/dzień/kg pobranej paszy. Pozorne wchłanianie azotu w jelicie cienkim obliczone w odniesieniu do ilości azotu wpływającego z treścią do dwunastnicy wynosiło 52, 53 i 59% w kolejnych grupach.

Suma aminokwasów pobranych z paszą (g/dzień/kg paszy) wynosiła 128, 62 i 49 g; przechodzących do dwunastnicy 107, 113 i 928 g, opuszczających jelito biodrowe 64, 65 i 60 g, odpowiednio w kolejnych grupach. Pozorne wchłanianie w jelicie cienkim większości aminokwasów osiągających z treścią dwunastnicę wynosiło od 50 do 70% we wszystkich grupach, natomiast pozorne wchłanianie cysteiny było znacząco mniejsze (25 do 40%). Ilość lizyny wchłanianej z jelita cienkiego u buhajków grupy I odpowiadała ilości pobranej z paszą, natomiast w grupie II i III była 2,4 i 3-krotnie, a metioniny dwukrotnie większa niż pobrancj, wskazując na wysoki stopień konwersji azotu dawki dla zwierząt grupy II i III w białko drobnoustrojów żwaczowych.