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The effect of soya-protein enriched with essential amino acids added to rumen of dairy cows in a form of rumen-protected tablets on the amino acid profiles of casein

S. Hadrová¹, L. Křížová¹, J. Šterc², O. Hanuš¹, J. Třináctý¹

¹*Research Institute for Cattle Breeding, Ltd., department Pohořelice, Czech Republic, e-mail:* <u>hadrova@vuvz.cz</u>

²University of Veterinary and Pharmaceutical Sciences, Ruminant Clinic, Brno, Czech Republic

Abstract

The aim of this study was to determine the effect of purified soya-protein HP300 enriched with the essential amino acids lysine, methionine and histidine added to rumen of lactating dairy cows either in a form of rumen-protected tablets or non-tableted mixture of the same composition on the casein content and yield and changes in the amino acid profiles of casein. The experiment was carried out on three lactating dairy cows H100 fitted with ruminal and duodenal cannulas. Cows were fed on a diet based on a corn silage, alfalfa hay and a supplemental mixture. The experiment was divided into 4 periods of 3 d (10 d preliminary and 3 d experimental period). In the first period one cow received the rumen-protected tablets (T group) and the other two received the non-tableted mixture (C group). In the subsequent period the rate of cows was antipodal. The casein content and yield was significantly higher in the T group in comparison to group C (P<0,05). The content of threonine was significantly higher in the T group than in the C group (3,98 vs. 3,87 %; P<0,05). With regard to differences in dry matter intake, the yield and composition of milk was recounted on DM intake.

Key words: dairy cows; rumen protection; amino acids; milk; casein

This study was supported by the National Agency for Agriculture Research of the Czech Republic, Project No. 1B44037

Introduction

Most of the amino acids absorbed by the mammary gland are used to synthesize milk proteins. Major proteins synthesized from amino acids in the mammary gland of cows are caseins (α_{S1} -casein, α_{S2} -casein, β -casein, κ -casein) and whey proteins (β -lactoglobulin, α lactalbumin and proteose-peptones). Remaining proteins found in the milk (bovine serum albumin and immunoglobulins) are absorbed directly from the blood (Bequette et al., 1998). Average concentration of individual fractions in bovine milk is approximately 50 % of α_s caseins, 36 % of β -casein and 14 % of κ -casein (Farrell et al., 2004). Most of the study proved that postruminally supplemented essential AA positively influenced casein synthesis, particularly in early lactation (DePeters et al., 1992). The aim of this study was to determine the effect of purified soya-protein HP300 enriched with the essential amino acids lysine (Lys), methionine (Met) and histidine (His) added to rumen of lactating dairy cows either in a form of rumen-protected tablets or non-tableted mixture of the same composition on the casein content and yield and changes in the amino acid profiles of casein.

Materials and methods

The experiment was carried out on three lactating dairy cows H100 fitted with ruminal and duodenal cannulas. The experiment was divided into 4 periods of 3 d (10 d preliminary and 3 d experimental period). In the first period one cow received the rumen-protected tablets (T group) and the other two received the non-tableted mixture (C group). In the subsequent period the rate of cows was antipodal. Treatment consisted of administrations of either rumen-protected tablets (T) or powder (C) containing purified soya-protein HP 300 and amino acids Lys, Met and His into the rumen of three lactating Holstein cows (1.–3. lactation) of mean live weight 523 kg.

Cows were fed individually twice daily (7.00 and 16.35 h) *ad libitum* a mixed diet based on a corn silage, alfalfa hay and a supplemental mixture. Cows were milked twice daily at 7.15 and 17.15 h. Milk yield was recorded and samples were taken at each milking during the experimental period. The samples of milk were conserved by 2-bromo-2-nitropropane-1.3-diol (Bronopol) and cooled to the 6°C and the basic compositions of milk were analysed by infrared analyser (Bentley Instruments 2000, Bentley Instruments Inc., USA).

Casein isolation was carried out following the conditions described by López-Fandiño et al. (1993). The lyophilized casein was dissolved in 10 ml Tris-HCl (pH 6,8) and sample buffer with 2-mercapthoethanol was added. For separation of casein fractions there were used separation gels (15% T, 2.6% C) and focussing gels (3% T, 2.6% C) (Laemmli, 1970) using Mini-Protean III Cell Electrophoresis apparatus (Bio-Rad Laboratories, Richmond, CA). The gels were coloured with Commassie Brilliant Blue R-250. For evaluation the platform ElfoMan 2.0 (Servis a prodej laboratorních přístrojů, Praha, CZ) was used. Quantification was performed on the basis of intensity of colouring and of individual areas casein fraction bands.

For AA analysis, the samples of lyofilized milk for acid hydrolysis were hydrolyzed with 6 mol.1⁻¹ HCl for 24 h at 110°C. The sulphur amino acids were determined as cysteic acid and methionine-sulphone by oxidation-acid hydrolysis. All hydrolyzates were separated on the automatic aminoanalyser (AAA 400, Ingos s.r.o., Praha, CZ) using Sodium citrate buffer system and quantified by reaction with ninhydrin. For determination of amino acids content the programme ChromuLan v. 0.7 (Ingos, Praha, CZ) was used.

Data resulting from the experiment were analysed using the GLM procedure of Statgraphics 7.0 package.

Results

Mean milk yield and the basic composition of cow's milk (dry matter of milk, protein, casein) is presented in the Table 1. The DM intake was significantly higher (P<0,05) for cows receiving supplement in the form of rumen-protected tablets (T group) with soya-protein and amino acids than in the control group (C group). Increased milk protein yield was followed closely by increases in casein content and yield in the T group (P<0,05). With regard to differences in DMI, the yield and composition of milk was expressed in dry matter intake. Milk yield/DMI was unaffected by the treatment (P>0,05). Casein yield/DMI was higher (P<0,05) in the T group. The relative amino acids content in casein is presented in Table 2. The relative content of Thr in casein was significantly higher in the T group than in the C

group (3,98 vs. 3,87 %; P<0,05). The yield of individual amino acids in casein is not presented in Table. Administration of soya-protein enriched with Lys, Met and His added to rumen of dairy cows in a form of rumen-protected tablets increased the yields of all amino acids in casein (P<0,05). Yields of amino acids of casein expressed in DM intake is presented in Table 2. Expressed amino acids yields in casein in both cases were significantly higher (P<0,05) in the T group.

Conclusions

The most limiting amino acids for synthesis of milk and milk protein have been reported to be Met and Lys. Inconsistent production responses to rumen-protected AA may be due to the possibility that several EAA are often co-limiting. In the present study we found out that the concentration of Thr in casein fraction was significantly higher (P<0,05) in the T group compared to C group. Presented results suggest that supplementation of diet with rumen-protected polymerically encapsulated tablets containing soya-protein and Lys, Met and His was effective because of influenced casein yield and amino acids composition of casein.

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Tab. 1: Milk yield and composition

Component	$T^{1}(n=18)$		$C^{2}(n=18)$	
	Mean	SEM	mean	SEM
Dry matter intake (kg)	16,14 ^a	0,31	15,69 ^b	0,36
Milk yield (kg/d)	17,77 ^a	0,36	16,63 ^b	0,47
Dry matter of milk (%)	11,80	0,16	11,72	0,19
Protein (%)	3,24	0,07	3,11	0,10
Protein yield (g)	572,44	7,55	515,79	20,44
Casein (%)	2,70 ^a	0,06	2,46 ^b	0,10
Casein yield (g)	477,96 ^a	11,08	407,82 ^b	18,95
Casein yield/DMI (g/kg)	29,76 ^a	0,78	25,91 ^b	0,88

^{a,b} means in the same row followed by the different superscripts differ (P<0.05) 1 experimental group; 2 control group

Tab. 2: The relative amino acids content in casein (%) and yields of amino acids of casein expressed in DM intake (g/kg)

Amino	The relative amino acids content in casein				Yields of amino acids of casein/DMI			
acid	$T^1(n=1)$	18)	$C^{1}(n=18)$		$T^{1}(n=18)$		$C^{1}(n=18)$	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Asp	6,55	0,04	6,50	0,04	1,95 ^a	0,05	1,68 ^b	0,06
Thr	3,98 ^a	0,02	3,87 ^b	0,04	1,19 ^a	0,03	1,00 ^b	0,04
Ser	5,43	0,02	5,40	0,03	1,62 ^a	0,05	1,40 ^b	0,05
Glu	21,35	0,25	20,87	0,28	6,34 ^a	0,14	5,41 ^b	0,20
Pro	12,06 ^a	0,10	12,40 ^b	0,14	3,59 ^a	0,10	3,21 ^b	0,11
Gly	1,60	0,02	1,62	0,01	0,48 ^a	0,02	0,42 ^b	0,01
Ala	2,67	0,02	2,68	0,02	0,80 ^a	0,03	0,69 ^b	0,02
Val	6,32	0,04	6,30	0,04	1,88 ^a	0,06	1,63 ^b	0,05
Met	2,62	0,02	2,68	0,03	0,78 ^a	0,02	0,69 ^b	0,02
Ile	4,76	0,03	4,78	0,03	1,42 ^a	0,04	1,24 ^b	0,04
Leu	8,84	0,05	8,93	0,06	2,63 ^a	0,08	2,31 ^b	0,08
Tyr	4,94	0,04	5,01	0,06	1,47 ^a	0,04	1,30 ^b	0,05
Phe	4,60	0,04	4,66	0,05	1,37 ^a	0,04	1,21 ^b	0,04
His	2,76	0,03	2,82	0,03	0,82 ^a	0,03	0,73 ^b	0,02
Lys	7,61	0,05	7,56	0,07	2,27 ^a	0,06	1,96 ^b	0,07
Arg	3,89	0,03	3,92	0,04	1,16 ^a	0,03	1,01 ^b	0,04

^{a,b} means in the same row followed by the different superscripts differ (P<0,05) 1 experimental group; ² control group