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THE DIGESTIVE UTILIZATION OF METHIONINE FROM PROTEIC RAW MATERIALS WITH SUPPLEMENTARY ADDITION OF DL-METHIONINE MARKED WITH RADIOACTIVE ISOTOPE ³⁵S

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Abstract

The aim of this study was to evaluate the influence of supplementary addition with DLmethionine a proteic raw materials used in the compound feed on the retention rate and apparent digestibility of methionine, as well as utilization in the organism of their amino acid. The results indicates that the organism use shortly the methionine from fodders, when the ratio between methionine and lysine was reduced (0,96%), indicating an ratio unhinged between the two essentials amino acids. The apparent digestibility of methionine to the soybean meal (86,26%) was improved through supplementation with DL-methionine, analogous to fish meal (89,12%).

Key words: methionine, digestibility, retention rate.

INTRODUCTION

Like lysine, an essential amino acid in the animal nutrition is methionine, sulphur amino acid which interferes in the lipids metabolism, in the thyroids' function, in the livers' protection (Batterhan, 2001), being critical amino acid for swine and poultry (Baker, 1994).

It has been observed that in the presence of methionine the nitrogen is used economically and the synthesis of corporal protein is stimulated, influencing in this way the rhythm of animal growing (Marin, 2007; Stoica and Stoica, 2001).

In food the methionine can substain fully cysteine. This is explained by the fact that cysteine can be synthetized in the organism because of methionine (Church and Pond, 1996; Dragotoiu, 2003).

As lysine, the methionine is produced on synthetical way, the results regarding the using of this amino acid in the animal's food being very different (Brinegan et al., 2000; Peace et al., 1986; Sprinson and Rittenberg, 1990; Susenbeth, 1998). So, the aim of the present study is to establish the retention rate and the apparent digestibility of methionine and also the evolution of the contained of methionine in blood, liver, intestine, muscles, in the conditions in which methionine marked with radioactive isotope ³⁵S has been used.

MATERIAL AND METHODS

The researches have been done in the Centre of Medical Researches – Fundeni on a number of 88 rats divided in four batches as much as similar as corporal weight. The rats have been kept in cages, two animals/cage, in optimal conditions of temperature and humidity.

The feeds have been established in conformity with the necessary energy and nutritive substances of the rats, starting from the chemical composition and the content in amino acids of the feeds used in the structure of the compound (table 1).

The administrated feeds have been constituted of an base mixture without nitrogen, but equilibrated in other nutritive parameters, made with starch wheat, sugar, sunflower oil, mineral and vitamin mix, being completed at the experimental batch I with soya meal (27,50%) and 0,24% DL-methionine marked with radioactive isotope ³⁵S, at the experimental batch II with 36% sunflower meal and 0,12% DL-methionine ³⁵S, at the experimental batch III with 19,20% fish meal and 0,08% DL-methionine ³⁵S. At the all of the three experimental batch II and 12,23% at the batch III) and the methionine level was the same at all the four batches (0,40%).

Table 1

The structu	re and the recipes para	ameters used in the exp	erience
Ingredients (%)	Batch I	Batch II	Batch III
Starch wheat	52,26	43,88	60,72
Cellulose powder	5,00	5,00	5,00
Sugar	3,00	3,00	3,00
Sunflower oil	6,00	6,00	6,00
Mineral mix	5,00	5,00	5,00
Vitamin mix	1,00	1,00	1,00
Soya meal	27,50	-	-
Sunflower meal	-	36,00	-
Fish meal	-	-	19,20
DL-methionine	0,24	0,12	0,08
TOTAL	100,00	100,00	100,00
	Recipes pa	rameters	-
EM (kcal/kg)	2883	2784	2932
PB (%)	12,29	12,24	12,23
Lysine (%)	0,673	0,418	0,812
Methionine (%)	0,402	0,402	0,400
Treonine (%)	0,475	0,432	0,491
Triptophan (%)	0,162	0,158	0,127
Valine (%)	0,585	0,640	0,566
Leucine (%)	0,943	0,777	0,793
Isoleucine (%)	0,572	0,482	0,501
Phenilalanine (%)	0,572	0,500	0,438
Cistine (%)	0,173	0,198	0,104
	Ratios b	etween:	
EM/PB (kcal/g%)	234,6	227,4	239,7
Methionine/PB (%)	0,033	0,032	0,033

The structure and the recipes parameters used in the experience

From the each batches were sacrificed three animals at 3, 6, 10, 15, 21 and 27 days to establish the way of repartition of marked methionine in rats' organism. The radioactivity of DL-methionine marked with the ³⁵S isotope from the samples of feeds, faecals, urine, blood, liver, intestine, muscles have been measured with the spectrometer with liquid scintillation Beckman LS-6500.

For establishing the digestibility coefficients of methionine from administrated feeds of the experimental batches in which have been used proteic raw materials have been organized experiences with radioactive isotopes, the time of the digestibility experiences been for 11 days, from which 5 days has been the preparing period and 6 days the control period. The rats have been kept in individual cages, the harvesting of urine and faecals has been made separately. The determination of methionine from urine and faecals has been made at the end of the experimental period, being used with the amino acids analyzator AAA 339 M.

The experimental dates have been statistical expounded, using the Student test.

RESULTS AND DISCUSSION

For looking after the way of repartition of methionine in the rats' organism after the ingest of the marked amino acid has been measured the evolution of the methionine radioactivity from blood, liver, intestine and muscles (table 2).

Table	2
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Batch	Sacrifice at:	DL-methionine in:			
		blood	liver	intestine	muscle
	1 day	8,08 <u>+</u> 0,08	30,57 <u>+</u> 0,35	64,49 <u>+</u> 0,68	14,45 <u>+</u> 0,12
	3 days	21,04 <u>+</u> 0,29	97,99 <u>+</u> 1,29	140,93 <u>+</u> 2,42	27,04 <u>+</u> 0,56
	6 days	45,14 <u>+</u> 0,45	142,54 <u>+</u> 2,21	197,69 <u>+</u> 3,96	59,39 <u>+</u> 0,85
L	10 days	88,86 <u>+</u> 1,48	249,37 <u>+</u> 4,36	231,97 <u>+</u> 4,51	100,66 <u>+</u> 1,98
	15 days	127,12 <u>+</u> 2,78	352,46 <u>+</u> 7,82	277,69 <u>+</u> 7,29	139,56 <u>+</u> 3,40
	21 days	154,79 <u>+</u> 3,51	385,19 <u>+</u> 10,52	303,66 <u>+</u> 6,31	232,15 <u>+</u> 6,52
	27 days	182,72 <u>+</u> 6,12	416,98 <u>+</u> 13,55	359,63 <u>+</u> 8,69	253,51 <u>+</u> 5,62
	1 day	13,30 <u>+</u> 0,15	38,84 <u>+</u> 0,49	58,04 <u>+</u> 0,86	9,84 <u>+</u> 0,17
	3 days	40,16 <u>+</u> 0,56	115,97 <u>+</u> 1,89	157,64 <u>+</u> 2,93	36,47 <u>+</u> 0,54
	6 days	74,17 <u>+</u> 1,30	230,63 <u>+</u> 2,88	221,50 <u>+</u> 3,93	68,87 <u>+</u> 0,96
L_{II}	10 days	97,80 <u>+</u> 2,01	238,27 <u>+</u> 4,86	230,52 <u>+</u> 4,09	115,45 <u>+</u> 2,28
	15 days	153,16 <u>+</u> 2,98	347,67 <u>+</u> 7,82	296,33 <u>+</u> 6,29	149,36 <u>+</u> 3,22
	21 days	158,66 <u>+</u> 3,61	321,24 <u>+</u> 8,28	321,69 <u>+</u> 7,67	165,94 <u>+</u> 4,02
	27 days	157,70 <u>+</u> 4,73	311,37 <u>+</u> 10,43	285,01 <u>+</u> 7,12	198,18 <u>+</u> 3,49
	1 day	19,67 <u>+</u> 0,31	31,38 <u>+</u> 1,07	50,34 <u>+</u> 1,09	9,43 <u>+</u> 0,14
	3 days	47,22 <u>+</u> 0,81	112,89 <u>+</u> 2,10	184,95 <u>+</u> 3,44	31,48 <u>+</u> 0,55
	6 days	73,12 <u>+</u> 1,37	176,36 <u>+</u> 3,09	240,14 <u>+</u> 4,44	79,99 <u>+</u> 1,34
L _{III}	10 days	91,01 <u>+</u> 1,54	246,49 <u>+</u> 4,11	281,49 <u>+</u> 5,47	128,31 <u>+</u> 2,64
	15 days	153,35 <u>+</u> 2,98	315,66 <u>+</u> 6,71	303,98 <u>+</u> 5,69	182,16 <u>+</u> 3,41
	21 days	171,03 <u>+</u> 4,60	427,73 <u>+</u> 11,51	340,57 <u>+</u> 7,47	251,70 <u>+</u> 3,87
	27 days	198,30 <u>+</u> 6,94	496,87 <u>+</u> 17,42	393,87 <u>+</u> 11,82	274,28 <u>+</u> 6,86

The evolution of the total radioactivity of methionine in rats' organism (DPM/g) (x 10^3)

At the experimental batch I, which has been administrated only one source of protein, respectively soya meal added with 0,24% DL-methionine, it has been established a progressive increase of lysine radioactivity, in the first ten days of experience comparative with the other batches, because of an accumulation of methionine in blood to be observed later, reaching at the end of the experimental period at 182,72 DPM/ml, value which is close to the one registered in the third experimental batch (198,30 DPM/ml), in which's food participate fish meal and 0,08% DL-methionine. A lower value has been determinated at the second experimental batch in which's food participated sunflower meal and 0,12% DL-methionine, a balance of measured radioactivity (153,16 DPM/ml at 15 days, 158,66 DPM/ml at 21 days and 157,70 DPM/ml at 27 days) has been observed.

Progressive developments of radioactivity have been observed at the liver at the experimental batches I and III, where at the end of the experience values of 416,98 DPM/g at the first batch and 496,87 DPM/g at the third batch have been established. The second batch registered a quicker development of radioactivity until the 15 day of experience (347,67 DPM/g), afterwards a decrease has been observed, reaching at 311,37 DPM/g in the 27 day, which is because of lower using of methionine in food because of imbalance between methionine and lysine, this ratio being of 0,96%.

By adding methionine in the soya meal which is known, that it has less amino acid, a ratio has been established between methionine and lysine (0,59%), similar results being obtained with fish meal, which contains protein with an equilibrated content of amino acids. In these conditions, methionine has been better valorificated by the organism being eliminated in a lower proportion.

Regarding the evolution of methionine radioacitivity in the rats' intestine, a closer evolution of batches until the 15 day of experience has been remarked, afterwards bigger values have been registered at the end of the experimental period at the third batch with 393,86 DPM/g and the first batch with 359,63 DPM/g, comparative to the second experimental batch (285,01 DPM/g), decrease determinated because of a small diminution of food consumption.

During the experimental period it has been measured the methionine radioactivity from the muscles rats, established an increase of radioactivity at the first batch (253,51 DPM/g) and the third batch (274,28 DPM/g), oposite to the second batch which's measured value has been of 198,18 DPM/g.

The established values obtained experimentaly demonstrates an accumulation of radioactive methionine bigger than in liver and intestine, the resulted amino acids after digestion passing through the mucous

membrane of the small intestine, from where by porta vein are transported to the liver and from here by the general blood circulation reaching at the level of tissue and cells.

In the continuation of the researches, by the base of the obtained dates during the digestibility experiences, it has been established the coefficients of apparent digestibility and the retention rate of the methionine from the administrated feeds to rats from the experimental batches I, II and III (table 3).

Table 3

Batch	CDa(%)		Retention rate of lysine (%)	
	x+s _x	v%	x+s _x	v%
LI	86,26 <u>+</u> 0,88	4,19	57,84 <u>+</u> 0,55	3,84
L _{II}	84,46 <u>+</u> 0,94	4,49	43,89 <u>+</u> 0,50	4,57
L _{III}	89,12 <u>+</u> 0,89	3,91	58,35 <u>+</u> 0,55	3,77

The coefficients of apparent digestibility and the retention rate of the methionine from the proteic raw materials

By adding soya meal with DL-methionine, a digestibility coefficient of methionine (86,26%) has been obtained, higher then the soya meal (84,46%) registered an closer to the fish meal (89,12%).

Regarding the retention rate of the methionine, it has been observed a similar proportion of using in organism of methionine in case of soya meal and fish meal (57,84% and 58,35%), both supplemented with DLmethionine, the differences being not significant statistically.

CONCLUSIONS

1. The coefficients of apparent digestibility of the methionine established in experimental period have been evidenced a improvement of the utilization of methionine in case of soya meal additioned with synthetical methionine (86,26%).

2. The retention rate of methionine has been improved by the supplementation of the synthetical amino acid DL-methionine of soya meal (57,84%), this been near of fish meal with DL-methionine (58,35%). It can been remarked that soya meal added with DL-methionine can substitute with better results the fish meal in the recipes of compound feeds.

3. The used of methionine in the rats' organism is influenced by the ratio between this amino acid and another essential amino acid represented by lysine in the conditiond in which protein and methionine are at the same level (12% PB and 0,40% methionine) at the I, II and III experimental batches.

4. The measured values of the radioactivity of methionine marked with the isotope 35 S indicates that in the organism is deposit the methionine in bigger quantities in intestine and liver.

5. In consequence, the animal proteic raw materials can be replace with vegetal materials, on condition that the ratio between the essential amino acids to be provided.

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