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Amino acid Composition and Biological Value of milk of Galshinsky-Breed Cows During Adaptation

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***Abstract* − The aim of the research was to study, considering the modern requirements of amino acid, composition and biological value of milk proteins of Holstein cows, imported from the Netherlands, during the adaptation period in the agricultural holding "EkoNivaAgro" (Voronezh region) with the level of milk yield in the herd of 8700 kg, a mass fraction of 3.8% of fat and 3.35% of protein. As a result, a high concentration and a complete set of essential and non-essential amino acids of milk proteins were revealed, which indicates their biological value. Among the essential amino acids, there is the greatest number of phenylalanine + tyrosine (PHE + TYR), lysine (LYS), leucine (LEU), Val. The content of tryptophan was insignificant. The sum of the essential amino acids exceeded their amount in the ideal protein. However, the amino acid number varied in a rather wide range: from 60.6 % in tryptophan (TRP) to 197% in the amino acids phenylalanine + tyrosine (PHE + TYR), which shows their imbalance. The limiting amino acid is tryptophan, which determines the level of use of other essential amino acids of milk. As a result, with high natural digestibility of milk proteins (96%), the human body will use only 59% of incoming essential amino acids.**

Key words− breed, amino acids, biological value, adaptation.

I. INTRODUCTION

The need for protein is the most important evolutionary component in the nutrition of humans and animals, caused by the need to provide an acceptable physiological level of receipt, especially (essential) amino acids.

The basis of a full complete protein having a full set of essential amino acids sufficient for protein biosynthesis in the human body is animal products (meat and meat products, fish, eggs, milk, seafood). Proteins of animal origin are absorbed by the body by 93-96 %.

It should be noted that milk proteins have the highest biological value, second only to egg proteins in this quality [1, 2, 3]. Milk proteins are well absorbed and digested by the body. Milk proteins are a good source of supply of amino acids, as well as sulfur and phosphorus. Milk almost completely provides the body with essential nutrients, and in the form that improves the process of assimilation in the digestive organs of children and adults.

Essential nutrients of milk for human include essential amino acids contained in proteins; trace elements: Mo, Fe, F, I, Cu, Mn, Se, Co, Zn; calcium in the form of Ca2+ ions in compounds; phosphorus and sulfur in the form of their compounds.

Milk proteins are synthesized in the secretory cells of the mammary gland from free amino acids in blood, immunoglobulins, and serum albumin in milk comes directly from the blood. They have a complex composition, diverse in structure, physical and chemical properties and biological functions.

Therefore, milk and milk products are vital and affordable food of the population. Milk is of exceptional importance in the nutrition of children, especially in the first period of their life. In the protein, the membranes of fat globules contain a significant amount of phospholipids, arginine and threonine-amino acids, normalizing the growth and development of the body.

In this regard, among the selection priorities of cow-milk quality indicators, the mass fraction of protein is nowadays in

the first place. Although this feature is most genetically determined, obtaining milk with a high content of complete protein is possible only with well-balanced diets for all controlled nutrients [4, 5].

Under conditions of industrialization of the Russian dairy industry, there is a certain tendency to increase animals of Holstein breed of foreign selection. However, the norms and technologies of feeding and keeping cows in agricultural enterprises do not fully correspond to the animal of intensive type of productivity. Highly productive cows, having a high metabolism, are more demanding to the quality of diets, which is not always provided in farms due to the shortage, and sometimes low quality voluminous feed (juicy and coarse feed), which determines the concentrate type of feeding. Diets with a high proportion of carbohydrates and starch without fiber are poorly absorbed by ruminants, which ultimately leads to a decrease not only in milk yield, but also in the quality characteristics of milk [6, 7, 8]. In addition, the process of adaptation, with the change of climatic conditions, is associated with a heavy load on the body of animals, which also has a negative impact on their productivity.

The Holstein breed is the most widespread in the world, and the most adapted in the conditions of loose technology of the maintenance. Having its own characteristics in different countries, it retains the main qualities: high milk production and good adaptability to modern industrial conditions of maintenance and milking. The European type of black-and-white cattle of Holland is characterized by a dense physique, with a high mass fraction of fat in milk. As a result of the increased use of Holstein bulls-improvers in black-and-white cattle, it is now characterized by a pronounced dairy type.

Long-term selection of milk productivity contributed to the fact that Holstein dairy cattle productivity dominates the function of self-preservation. Agricultural enterprises that purchase cattle abroad for the acquisition of modern dairy complexes meet with a number of problems. First of all, it is a high level of mortality and culling of imported animals because of various diseases. Typically, this figure should be at 10 %, but usually it is 30-50 %. This fact is due to the fact that the Holstein cattle is poorly acclimatized in new environmental conditions [9].

Feed quality and feeding are the main factors determining the productivity and health of Holstein cattle. Unfortunately, in many farms that purchased imported Holstein cattle, this is not given enough attention.

The norms and technologies of feeding and keeping of cows do not fully correspond to the animals of intensive type of productivity.

Highly productive cows, having a high metabolism, are more in demand by the quality of diets, which is not always provided in farms due to the shortage, and sometimes low quality voluminous feed (juicy and coarse feed) that determines the concentrate type of feeding. Diets with a high proportion of carbohydrates and starch, but not fiber, are poorly absorbed by ruminants, which ultimately leads to a decrease not only in milk yield, but in the quality characteristics of milk [6,7,8]. In addition, the process of adaptation, with the change of climatic conditions, is associated with a heavy load on the body of animals, which also has a negative impact on their productivity.

During the adaptation period, Holstein cows usually exhibit high milk productivity, but do not fully realize their potential [9, 10, 11]. At the same time, the biological value of the milk produced by Holstein animals, taking into account modern requirements, has not been studied.

In this regard, we have set a goal to study the amino acid composition and biological value of milk proteins of Holstein cows of Dutch selection during the adaptation period.

II. Objects And Methods Of Research

The research was conducted in the agricultural holding "EkoNivaAgro", Voronezh region.

Milking herd of dairy complex, formed by animals of Holstein breed of European selection, is contained in a loose technology of modern dairy complex. At the same time, the lack and low quality of bulky feed determined the type of concentrate feeding of cows at a specific weight of concentrates more than 50 %.

Milk yield per cow, the mass fraction of fat and protein during the experiment were respectively: 8700 kg, 3.8 and 3.35 %.

The object of the study was the full-aged cows of Holstein breed at 4-5 months of lactation, imported from the Netherlands.

The amino acid composition of milk samples (n=4) was determined by high-performance liquid chromatography on a liquid chromatograph Shimadzu LC-20 Prominence, (Japan) [12, 13].

To establish the biological value of milk proteins, the amino acid digestibility coefficient (Protein digestibility-corrected amino acid score (PDCAAS) recommended by FAO/WHO in 1993 and more acceptable for UN protein quality assessment was used [14 - 16].

This method allows you to control not only the concentration of essential amino acids, but also to determine the degree of digestion.

The amino acid digestibility coefficient of milk proteins (PDCAAS) was calculated by the formula:

$$PDCAAC = \frac{A_1}{A_2} \cdot K, \qquad (1)$$

where A_1-the amount of limiting amino acid in g / 100 g of milk proteins;

A_2-the amount of the same amino acid in 1 g/100 g of the "standard" protein;

K - % true digestibility of milk proteins (96 %).

The limiting amino acid (A_1) was determined by the minimum value of the amino acid number of a specific essential amino acid by the formula:

$$A_1 = \frac{A}{A_2}, \quad (2)$$

where A - the amount of essential acid in g / 100 g of milk proteins.

The formula of standard protein (A_2) approved by FAO/WHO in 2011, was used to assess the biological value of milk proteins [17]. As a standard, it adopted the need for essential amino acids in children at the age period of 2-5, which is considered to be the highest relatively to other age groups (table.1).

TABLE 1. FORMULA REFERENCE PROTEIN

Amino Acid	*Content, g/100 g*
Isoleucine (ILEU)	3.0
Leucine (LEU)	6.1
Lysine (LYS)	4.8
Methionine + cysteine (MET + CYS)	2.3
Phenylalanine + tyrosine (PHE + TYR)	4.1
Threonine (THR)	2.5
Tryptophan (TRP)	0.66
Valine (VAL)	4.0
Histidine (HIS)	1.6

The resulting digital material is processed by the method of variation statistics on algorithms [18] using a computer program Microsoft Office "Excel".

III. Results And Their Discussion

Proteins are the most valuable components of milk, the monomer units of which are amino acids. There were 20 amino acids in animal proteins, in milk proteins - 19. In food proteins there are 9 amino acids (threonine, leucine, phenylalanine, isoleucine, lysine, tryptophan, histidine, valine and methionine), which are essential for humans and animals.

Due to a certain concentration of essential amino acids and their digestibility in the gastrointestinal tract, milk proteins have a high biological value. They supply the body with phosphorus and sulfur. Milk contains more essential amino acids, such as methionine, tryptophan, isoleucine, than fish, meat and vegetable proteins. Milk proteins perform a wide variety of important vital functions in the body: the basis of structural units of the body (cell membrane protein), information transfer (hormones), body protection (lysozyme, immunoglobulin), etc. [1, 2, 3].

TABLE 2. AMINOACID COMPOSITION OF MILK PROTEINS OF HOLSTEIN COWS

Amino acid	*In nmol / ml*	*as % of total composition*	*of milk in % of total protein*
Essential amino acids (A)			
Isoleucine (ILEU)	204.05±10.024	0.16±0.007	4.20±0.016
Leucine (LEU)	390.40±18.106	0.21±0.091	8.04±0.019
Lysine (LYS)	298.93±13.943	0.27±0.011	6.86±0.015
Methionine +cysteine (MET + CYS)	99.50±7.168	0.10±0.007	2.45±0.080
Phenylalanine+ tyrosine (PHE + TYR)	294.39±13.823	0.32±0.013	8.09±0.019
Threonine (THR)	196.13±9.580	0.14±0.006	3.67±0.015
Tryptophan (TRP)	15.74±6.442	0.02±0.006	0.40±0.157
Valine (VAL)	277.57±13.366	0.20±0.008	5.10±0.014
Histidine (HIS)	90.69±4.158	0.08±0.003	2.15±0.005
∑(A)			41
Interchangeable amino acids (A^1)			
Aspartic acid + asparagine (Asp+ASN)	310.03±14.187	0.25±0.010	6.48±0.009
Serine (SER)	283.92±12.273	0.18±0.007	4.68±0.014
Glutamic acid + glutamine (Glu+GLN)	791.18±35.072	0.71±0.027	18.27±0.030
Alanine (Ala)	194.09±9.014	0.11±0.004	2.21±0.016
Arginine (Arg)	662.96±34.012	0.71±0.031	18.12±0.193
Proline (PRO)	430.46±18.145	0.30±0.011	7.75±0.003
Glycine (Gly)	131.99±4.718	0,06±0,002	1,56±0,016
∑ (A^1)			59,0
Ratio ∑(A) / ∑(A^1)	0.69		

TABLE 3. BIOLOGICAL VALUE OF MILK COWS BELCWGALCHINSKY BREED

Amino acid	*Essential amino acids, FAO / who scale*		*Essential amino acids of milk proteins*	
	Content, g / 100 g amino	*Acid number*	*Content, g / 100 g amino*	*Acid number, limiting amino acid**
Isoleucine (ILEU)	3.0	1.0	4.2	1.40
Leucine (LEU)	6.1	1.0	8.0	1.31
Lysine (LYS)	4.8	1.0	6.5	1.35
Methionine +cysteine (MET + CYS)	2.3	1.0	2.4	1.04
Phenylalanine+ tyrosine (PHE + TYR)	4.1	1.0	8.1	1.98
Threonine (THR)	2.5	1.0	3.6	1.44
Tryptophan (TRP)	0.66	1.0	0.4	0.61*
Valine (VAL)	3.9	1.0	5.1	1.31
Histidine (HIS)	1.6	1.0	2.7	1.69
∑	29.0	–	41.0	–
Digestibility of proteins, %	96.0			
Amino acid digestibility coefficient of proteins (PDCAAS), %	59.0			

a.

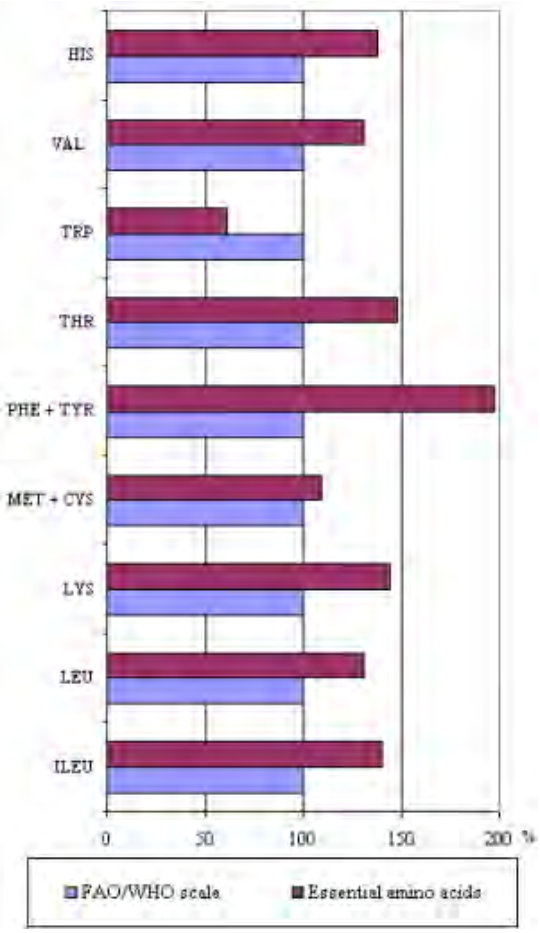

Fig.1 Aminogram of milk of Holstein cows

The human body can not synthesize essential amino acids, so they must enter the body as part of food proteins.

However, non-essential amino acids make up a large enough specific weight (up to 2/3) in the milk protein and perform very important functions in the human body, and some of them are no less important than essential.

It should be borne in mind that non-essential amino acids are formed in the human body, but internal factors are not able to fully provide them in the required amount, so they also have to come from food.

When studying the amino acid composition of milk proteins of Holstein cows, the results of which are summarized and presented in table 2, a complete set and a high concentration of essential and non-essential amino acids confirm its biological value.

Among the essential amino acids, the largest amount in cow milk was found to be phenylalanine + tyrosine (PHE + TYR), lysine (LYS), leucine (LEU), valine; the content of tryptophan (TRP) was insignificant.

High concentrations in proteins of non-essential amino acids had glutamic acid + glutamine (Glu+GLN) and Arginine (Arg), and the lowest alanine (Ala), Glycine (Gly). Total proportion of essential amino acids (41.0 %) in proteins is much smaller than the non-essential (59.0%), so their amino acid index was only 0.69, whereas in domestic (Simmental, red-mottled and Holstein) breed it is much higher – from 0.93 to 1.16 [19-25].

The biological value of proteins for the human body is determined by its balance of essential amino acids in relation to the ideal protein (FAO/WHO scale), as well as the efficiency of its use by the human body. Therefore, for the

synthesis of most proteins of the human body not only a sufficient amount of each essential amino acid is necessary, but also their ratio is also important. In the best case, their content should be in harmony with the proteins of the human body. Imbalance of these essential nutrients leads to disruption of the formation of its own proteins. The lack of at least one essential amino acid limits the use of all other amino acids in the life of the body [26].

Table 3 and Fig. 1 show the calculated indicators of biological value of milk proteins of the studied animal population during the adaptation period. They indicate the excess amount of essential amino acids in milk proteins over their amount in the ideal protein.

However, their amino acid number varies in a rather wide range: from 60.6 % in tryptophan (TRP) - to 197% in amino acids phenylalanine + tyrosine (PHE + TYR), which indicates their imbalance.

Therefore, the limiting amino acid is tryptophan. It determines in this case the level of use of other essential amino acids of milk of the studied population. As a result, with high natural digestibility of milk proteins (95%), the human body will use only 57% of the incoming essential amino acids.

Essential amino acid tryptophan is involved in protein synthesis as a structural element, and is a precursor of many physio-active compounds. Its quantity in proteins of animal origin is determined by the conditions of keeping animals and poultry [27-31].

The deficit of tryptophan in milk proteins was also revealed in highly productive cows of the Simmental breed of the Austrian selection during the adaptation period, but the biological value of milk proteins increased when the corresponding feed additive was included in their diet [28].

It is considered that ruminants are less than other animals sensitive to the lack of essential amino acids in the diet, as they can synthesize them by microflora. However, the body of highly productive cows can not fully provide themselves with bacterial proteins; to meet their needs, it is necessary to use other sources of proteins, including in diets scarce amino acids, it is possible to reduce the need for protein by 15-20 %, increase milk productivity of cows and reduce feed costs of products [27-33].

IV. CONCLUSIONS

Thus, the study of amino acid composition and biological value of milk proteins of Holstein cows imported from the Netherlands during their adaptation allowed establishing the following:

cow milk had a high concentration of essential and non-essential amino acids and a complete set of them, which confirms its biological value;

among the essential amino acids, the largest amount in cow milk contained phenylalanine + tyrosine (PHE + TYR), lysine (LYS), leucine (LEU), valine, tryptophan saturation (TRP) was insignificant. High concentrations of glutamic acid + glutamine (Glu+GLN) and Proline (PRO), and the lowest alanine (Ala), glycine (Gly) were among the non-essential amino acids. The total share of essential amino acids (41.0 %) in proteins is much less than the non essential ones (59.0%), so their amino acid index was only 0.69;

although the sum of essential amino acids in milk proteins exceeded their number in an ideal protein, their amino acid number varied in a rather wide range: from 60.6 % in tryptophan (TRP) to 197% in amino acids phenylalanine + tyrosine (PHE + TYR), which indicates their imbalance;

the limiting amino acid is tryptophan, which determines the level of use of other essential amino acids of milk. As a result, with high natural digestibility of milk proteins (95%), the human body will use only 57% of the incoming especial amino acids.

To increase the biological value of milk proteins, it is necessary to optimize the diets of highly productive cows, including also in their composition, the scarce amino acids.

During organization of full feeding of dairy cattle, the paramount importance is to be given to the quality of the food, especially the volume of hay, haylage, silage. The low quality of the main feed makes it necessary to balance the diets due to the increased consumption of concentrates, which is unprofitably economical and harmful to animal health.

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