INFLUENCE OF MILK THISTLE SHOT ON QUALITY PARAMETERS OF THE SOUR-MILK BEVERAGE

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Abstract

Modern complicated ecological conditions cause a general necessity in improving the food structure of the population due to improving the quality, biological value and taste characteristics of products. Just that is why, the aim of the work was to study the influence of milk thistle shot on quality parameters of the sour-milk beverage – kefir. It was established, that it had the homogenous consistence with a broken clot and color from white to creamy with shot particles. The increase of the milk thistle shot dose to 3 % and 4 % results in the taste with the brightly expressed milk thistle smack and brown color with the expressed milk thistle content. The viscosity of kefir with fms 2,5 % changes during seven days of storage, although remains rather high at 8 day, namely 47 s. The increase of the viscosity of kefir with milk thistle shot is explained by its hygroscopic properties, in which result free moisture of the product is bound. The analysis of microbiological parameters in the process of storage of kefir with milk thistle shot allows to make a conclusion about the satisfactory sanitary condition of the new product and its harmlessness for consumer's health. In the kefir with milk thistle shot the general amount of amino acids grew by 11,6 %, including irreplaceable ones – by 10,1 %, replaceable – by 12,6 %, that indicates its biological value.

Addition of milk thistle shot to kefir didn't cause changes of the biological value of the protein component of the combined product. It is testified by the mean value of amino acid scores of control (129,3 %) and experimental (127,9 %) samples of kefir. Some growth of the valine score (3,6 %) can be noted.

So, the combined product is characterized by the balanced amino acid composition. Due to adding milk thistle shot, consumption of amino acids, necessary for synthesis of proteins and essential number of compounds, vitally important for the human organism, grew.

Keywords: sour-milk beverages, milk thistle, amino acids, organoleptic parameters, kefir.

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1. Introduction

The one of main directions of a conception of the state policy in the sphere of healthy nutrition includes elaboration of technologies of functional products for mass consumption, differentiated for prophylaxis of diseases and strengthening of protective functions of the organism, decrease of the risk of the effect of harmful substances, especially, for the population of ecologically unfavorable zones [1, 2]. Manufacturing of the wide assortment of treating-prophylactic food products, including milk ones causes a necessity to search for new sources of raw materials [3, 4].

Treating properties of food medical plants are conditioned by biologically active substances, including diverse vitamins, micro- and macroelements and enzymes [5]. Biologically active components of plants actively favor normalization of vitally important processes in the human organism, increase metabolism, provide the organism with necessary vitamins, mineral substances [6, 7].

Skillful combination of milk thistle shot as a supplement with the milk base has great perspectives in both biological-technical and social aspects.

Just that is why it is urgent to elaborate a technology of a sour-milk beverage using milk thistle shot that favors enrichment of these beverages with food fibers and biologically active substances.

2. Review of the problem

The problem of supplying the population with rational and balanced nutrition is very urgent today. Taking into account modern ecological conditions, a food ration of a human must contain natural biologically active substances, able to increase the resistance of the organism [8, 9]. Creation of new food products with a target destination due to using biologically active supplements, as opposite to traditional ones, allows to prevent and to correct results of human diseases [10, 11].

The special attention must be paid to natural vegetable supplements, because they give these milk products treating-prophylactic properties at the expanse of their natural ones [12, 13]. The use of such supplements allows to refill the deficit of food substances, to increase non-specific resistance of the organism to the effect of external unfavorable factors [7, 14].

Numerous authors offer the wide list of milk products with vegetable components, which use gives products with the planned structure and quality parameters [15, 16]. One of such supplements in the food industry is milk thistle, especially shot of fruits, which composition includes macro- and microelements (Ca content – 687 mg/100 g), amino acid, polyunsaturated fatty acids, record number of flavolignans, flavonoids, cellulose [17]. Thus, sylimarin in 100,0 g of shot – 4 g [18]. This all gives a possibility to name just this shot a lifebuoy for the liver and pancreas! With renewing the condition of a hepatic cell – pancreas condition improves, sugar level in blood normalizes, lipid metabolism improves, intestines' work normalizes. Clinical studies of shot of milk thistle fruits demonstrated the effectiveness of its use in the complex treatment of viral hepatitis B and C [19, 20].

That is why it may be stated unerringly, that milk thistle is useful for humans at the expanse of vitamins and other biologically active substances.

3. Materials and Methods

The experimental part of the work was conducted on the base of CAR Ivano-Frankivsk city milk plant" and department of milk and milk products technology of S. Z. Gzhytskyi national university of veterinary medicine and biotechnologies (Ukraine).

For the studies there were used kefir, produced according SSU 4417:2005 and kefir with milk thistle shot, produced by the elaborated recipe.

Milk thistle short according to TC U 15.6-24333456-001-2002 was used for the studies. Milk thistle shot must correspond to the following conditions: Outlook – comminuted seeds or clots with size 0,5–1 mm; Color – light-brown; Smell – fresh, herbal; Humidity – no more than 9 %; Caloric

value – 164 kcal; proteins – 15 g/100 g product; Lipids – 11 g/100 g product; Carbohydrates – 9 g/100g product.

The organoleptic estimation of kefir includes determination of such parameters as color, smell, taste, consistence.

The color was determined in a glass cylinder at day light.

The smell was determined at opening a tube with the product or at pouring from one tube to another.

The taste of the beverage was determined, taking a portion to the mouth, trying to moisten the whole oral cavity to the tongue root by it, exhaling air through the nose.

The consistence of the product was determined, slowly pouring it from the cylinder to another tube.

Measurements of viscosity were conducted according to SS 27709-88. This method determines the measurement of the dynamic viscosity by a viscosimeter, using the law of bead falling in the viscous medium.

The viscosity of a clot at producing the beverage was determined by the time of drain at 20 ^{o}C from a pipette with volume 100 ml with an output orifice with diameter 5 mm.

The drain time at the end of fermentation before mixing a clot must be no less than 20 s.

The titrated acidity was determined according to SS 3624-92. For that 10 cm^3 of the beverage or 5 g of it are measured or weighted in a conic flask (glass) with volume 100–250 cm^3, 20 cm^3 of distilled water are added, if the acidity of cream is determined, 30 cm^3 of water, if the acidity of the sour-milk beverage is determined. Then this all is mixed and 3–4 drops of phenolphthalein are added and the mixture is titrated by the solution of sodium hydroxide to the light rose coloration that doesn't disappear during 1 min.

For determining the consistence of the sour-milk beverage with fillings in the process of its storage up to 7 days, there was used the method of determining the consistence by the spread diameter.

Materials for the study:

– slide glass;

– millimeter sheet of paper;

– microbiological loop;

– sour-milk beverage.

The essence of the method is following: the plane of a slide glass was fixed horizontally on a laboratory table on a millimeter sheet of paper. The beverage was accurately mixed in a chemical glass during 1–2 min by a microbiological loop. Then it was scooped by the loop from the glass and put from height 5–6 cm on the slide glass, a drop of the well mixed beverage fell from the microbiological loop on the millimeter sheet. According to its indications, the diameter of beverage spread was determined, mm.

Determination of the number of sour-milk bacteria was conducted according to SS 10444.11-89.

Determination of the amount of yeast and moulds was conducted according to SS 26888-86.

4. Experimental procedures

Experimental samples of kefir with milk thistle shot were produced at the Ivano-Frankivsk milk plant. A control was kefir, produced by the reservoir method by the traditional technology. Milk thistle shot was brought in kefir at its cooling before ripening. The obtained samples were assessed organolepically at first (**Table 1**).

As it can be seen from the aforesaid, kefir with 1 % and 2 % of milk thistle shot, was characterized with the clean sour-milk taste and smell. It has the homogenous consistence with a broken clot and color from white to cream with shot particles. The increase of milk thistle shot dose to 3 % and 4 % results in the taste with the brightly expressed milk thistle smack and the color from white to brown with the expressed content of milk thistle.

Based on the organoleptic parameters of kefir with milk thistle shot, we chose kefir with 3 % of shot for further studies.

Table 1

Organoleptic characteristic of kefir depending on milk thistle shot dose

milk thistle shot dose, %	Smell and taste	Consistence, outlook	Color
–	Sour-milk, clean, soft, with the expressed milk smell	Homogenous with a broken clot	White, homogenous along the whole mass
1,0	Sour-milk. Clean, with the milk thistle smack	Homogenous with a broken clot	White, homogenous, with hardly noticeable shot particles
2,0	–//–	–//–	Cream with shot particles
3,0	Sour-milk. with the expressed milk thistle taste	Homogenous, dense with a broken clot	Light-brown with shot particles
4,0	Sour-milk. with the brightly expressed milk thistle taste	Homogenous, very viscous, with a broken clot	Intense brown with expressed shot

5. Results

For assessing the change of the consistence of kefir with milk thistle shot at storage, its viscosity and spread diameter were determined (**Table 2, 3**).

Table 2

Change of the conventional viscosity at storage

Drain duration, s / Name of products	Initial sample	3 day	5 day	8 day
Kefir with fms 2,5 % (traditional)	57	57	53	47
Kefir with fms 2,5 % with milk thistle shot	59	60	64	66

Table 3

Change of the spread diameter of kefir at storage

Spread diameter, mm / Name of products	Initial sample	3 day	5 day	8 day
Kefir with fms 2,5 % (traditional)	8,8	9,8	11,6	14,1
Kefir with fms 2,5 % with milk thistle shot	8,0	8,0	7,6	7,2

As it can be seen from the above data of tables 1, the structure of products that is their consistence changes at storage.

The viscosity of kefir with fms 2,5 % changes during 7 days of storage, although remains rather high at 8 day, namely 47 s. It can be explained by the acetic microflora in the composition of kefir that is a cause of the high viscosity even after ending of its storage term.

The viscosity growth of kefir with milk thistle shot is explained by hygroscopic properties of shot that result in binding free moisture of the product.

The acidity change of sour milk products at storage is an important characteristic for assessing their quality (**Table 4**).

The titrated acidity growth of products at storage testifies to intensification of the growth of lactobacteria, although the acidity value is within limits, fixed by the standard, even after ending of the storage term. It is interesting that the acidity growth is a bit more intense in kefir with milk thistle shot comparing with traditional one and at 8 day of storage it is by 5 ^{o}T higher than in kefir.

Table 4

Change of titrated acidity of kefir at storage

Spread diameter, mm / Name of products	Initial sample	3 day	5 day	8 day
Kefir with fms 2,5 % (traditional)	90	93	99	103
Kefir with fms 2,5 % with milk thistle shot	89	96	102	108

Based on the **Table 4** data, we can assume that milk thistle shot is a nutritive medium for the growth of lactate bacteria and their development is illustrated by the titrated acidity value.

For specifying storage terms of kefir with milk thistle shot, microbiological studies were carried out.

The following parameters were chosen as microbiological criteria of kefir safety: titre of bacteria of colon bacillus group and MAFAM in 1 g of the product (**Table 5)**. At setting the storage term of the new type of kefir, these parameters must not exceed maximal permitted values, cited in normative documents (SSU 4417:2005. Kefir. Technical conditions).

Table 5

Microbiological parameters of kefir at storage at temperature 4±2 °C

	Storage term, days					
	0	**2**	**4**	**6**	**8**	**10**
	Traditional kefir					
Bacteria of colon bacillus group (coliforms), absent in the product mass, g	0,1	0,1				
	0,01	0,01	0,01	0,01		
	0,001	0,001	0,001	0,001	0,001	0,001
Pathogenic microorganisms, including salmonellas	absent					
Microscopic fungi, CFU/g	3	3	4	5	10	31
Yeast, CFU/g	2	2	2	3	4	54
	Sour-milk beverage with milk thistle shot					
Bacteria of colon bacillus group (coliforms), absent in the product mass, g	0,1	0,1				
	0,01	0,01	0,01	0,01		
	0,001	0,001	0,001	0,001	0,001	0,001
Pathogenic microorganisms, including salmonellas	absent					
Microscopic fungi, CFU/g	2	3	3	4	13	49
Yeast, CFU/g	2	2	3	3	6	26

We studied microbiological parameters of kefir with milk thistle shot at once after its production and at storage at temperature (4±2) °C during 2, 4, 6, 8 and 10 days. For comparison traditional kefir with fms 2,5 % was used as a control sample.

The analysis of microbiological parameters at storage of kefir with milk thistle shot allows to make a conclusion about the satisfactory sanitary condition of the new product and its harmlessness for consumer's health.

Thus, at determining titre of bacteria of colon bacillus on Kessler's medium three solutions of the product were inoculated (0,1, 0,01 and 0,001 g). In all samples of kefir at storage (temperature

(4±2) °C during 8 days this index was more than 0,01 g. It must be noted, that the titre value of bacteria of the colon bacillus group of kefir with milk thistle shot didn't differ from control ones.

Based on the fact that according to the literary data, milk thistle fruits have the high protein content, its amino acid composition was studied for determining the biological value of this component of a supplement, used at kefir production.

As we can see from the obtained data (**Table 6**), 100 g of milk thistle shot comparing with kefir (**Table 7**) contains 4,86 times more general amount of amino acids, among them 4,34 times more irreplaceable ones and 5,22 times more replaceable.

Among irreplaceable amino acids, the most exceed was inherent to lysine (6,46 times), the least – to methionine (2,83 times). The content in milk thistle shot comparing with kefir was more in threonine in 4,85 times, isoleucine in 4,65 times, phenylanaline in 4,60 times, tryptophan in 4,38 times, leucine in 3,86 times and lysine in 3,58 times. The more noticeable differences were established as to the content of replaceable amino acids. Especially, concentrations of glycine, arginine, cystine in milk thistle shot exceeded indices of kefir in 11,84; 9,10 and 9,04 times. The exceed was a bit less for asparagine acid (6,87 times), alanine (5,94 times), serine (5,79 times), practically equal for glutamic acid (4,96 times) and histidine (4,94 times) and the least for proline (2,86 times) and tyrosine (2,88 times). The share of irreplaceable and replaceable amino acids from their total amount in milk thistle protein was 6,88 and 63,12 %, that is typical for vegetable proteins.

At the same time we assessed the biological value of milk thistle protein. The biological value of proteins is conditioned by their ability to provide plastic processes and synthesis of biologically active compounds. This sign characterizes their quality and is conditioned by the presence of irreplaceable amino acids in them, their ratio between each other and to replaceable ones, and also assimilability in the gastrointestinal tract (amount of digested protein).

The fullness of using amino acids depends on the balance of irreplaceable ones. The closer amino acid composition of food protein is to the one in proteins of the human body, the higher its biological value.

Table 6

Amino acid composition of milk thistle shot protein

Amino acids	g/100 g of milk thistle shot	% of the total amount of amino acids
Irreplaceable amino acids		
Threonine	0,568	4,06
Valine	0,834	6,10
Methionine	0,212	1,52
Isoleucine	0,740	5,28
leucine	1,092	7,80
Phenylalanine	0,658	4,70
Lysin	0,854	6,10
Tryptophan	0,184	1,31
Totally	5,162	36,88
Replaceable amino acids		
Asparagine acid	1,560	11,15
Serine	0,846	6,04
Glutamic acid	2,476	17,69
Proline	0,760	5,43
glycine	0,580	4,14
Alanine	0,600	4,29
Cystine	0,226	1,61
Tyrosine	0,432	3,09
Histidine	0,400	2,86
Arginine	0,956	6,93
Totally	8,836	63,12
Together	13,988	100,0

The lack of one irreplaceable amino acid in food causes incomplete assimilation of other ones.

As it was demonstrated by the study of the amino acid composition of kefir with adding milk thistle shot, the increase of the total amount of amino acids by 11,6 %, including irreplaceable ones by 10,1 % and replaceable ones – by 12,6 % takes place in its composition (**Table 7**).

Table 7

Amino acid composition of kefir with milk thistle shot

Amino acids	Kefir samples			
	Control		Experimental	
	g/100 g of kefir	% of the total amount of amino acids	g/100 g of kefir	% of the total amount of amino acids
Irreplaceable amino acids				
Threonine	0,117	4,00	0,130	4,05
Valine	0,129	4,48	0,150	4,67
Methionine	0,075	2,60	0,079	2,46
Isoleucine	0,159	5,52	0,176	5,48
leucine	0,283	9,83	0,308	9,59
Phenylalanine	0,143	4,97	0,159	4,95
Lysin	0,240	8,34	0,259	8,06
Tryptophan	0,042	1,46	0,047	1,46
Totally	1,188	41,26	1,345	40,72
Replaceable amino acids				
Asparagine acid	0,227	7,78	0,267	8,31
Serine	0,184	6,39	0,203	6,32
Glutamic acid	0,499	17,33	0,558	17,37
Proline	0,269	9,34	0,284	8,84
glycine	0,049	1,70	0,005	2,02
Alanine	0,101	3,51	0,116	3,61
Cystine	0,025	0,87	0,031	0,96
Tyrosine	0,150	5,21	0,159	4,95
Histidine	0,081	2,81	0,091	2,83
Arginine	0,105	3,65	0,131	4,08
Totally	1,691	58,74	1,905	59,28
Together	2,879	100,0	3,213	100,0

The content of all amino acids grew in the experimental sample of kefir. Among irreplaceable amino acids the level of threonine (11,1 %), valine (11,6 %), isoleucine (10,07 %), phenylalanine (11,1 %) and tryptophan (11,9 %) grew mostly and practically equally. The growth was less for leucine (8,8 %), lysine (7,9 %) and least for methionine (5,3 %).

The growth of concentrations of a series of replaceable amino acids was more noticeable, especially, glycine by 32,6 %, cystine by 24,0 %, asparagine acid – by 17,6 %, arginine – by 24,6 %, alanine – by 14,8 %, a bit less histidine – by 12,3 %. The content of proline (5,6 %) and tyrosine (6,0 %) grew least.

6. Discussion

Chemical and biological methods are used for determining the biological value of food protein. The chemical method of so-called amino acid score that allows to determine limiting amino

acids is mostly spread in practice. Determination of limiting amino acids is conducted by comparing with an "ideal" protein with balanced amino acid composition. For its calculation, there is used amino acid of the expert committee of the World health organization FAO/WHO, according to which, 1 g of an "ideal" protein contains (in mg) isoleucine 40, leucine 70, lysine 55, sulfur-containing compounds (methionine + cystine) 35, aromatic acids (phenylalanine + tyrosine) 60, tryptophan 10, valine 50. The value of amino acid score more than 100 % indicates the full satisfaction of the human need in this amino acid, and less than 100 % testifies to its insufficient satisfaction. Such amino acids are considered as limiting.

The value of amino acid score is calculated by the formula:

$$\text{Amino acid score} = \frac{\text{mg in 1 g of studied protein}}{\text{mg in 1 g of ideal protein}} \times 100\ \%.$$

At assessing the biological value of proteins, we must take into account their assimilability in the organism, understood as an amount of protein, digested in the gastrointestinal tract (percent of consumed). According to the literary data, the assimilability of animal proteins varies within 92–96 %, and vegetable – 60–80 %. The assimilability of protein of eggs and milk is the highest, the lowest one –of leguminous (40–60 %).

The assimilability of food proteins depends on their accessibility for digestive enzymes, specific for separate proteins, and their presence. It is important to note that consumption of vegetable and animal proteins increases the assimilability of first ones to 80–90 %.

The calculation of amino acid score testifies to the high biological value of milk thistle shot protein (**Table 8**), which integral (mean) index was 115,83 %.

Table 8

Biological value of milk thistle shot protein

Amino acids	Standard protein FAO/WHO g/100 g of protein	Milk thistle shot g/100 g of protein	Amino acid score, %
Isoleucine	4,0	5,28	132,0
Leucine	7,0	7,80	111,43
Sum of aromatic (phenylalanine+tyrosine)	6,0	7,79	129,83
Sum of sulfur-containing (methionine+cystine)	3,5	3,13	89,93
Threonine	4,0	4,06	101,5
Valine	5,0	6,10	122,0
Lysine	5,5	6,10	110,91
Triptophan	1,0	1,31	131,0
Totally			115,83

The limiting one was only the sum of sulfur-containing acids (89,93 %). Threonine score (101,5 %) was close to the standard protein. The most scores were for isoleucine (132,0 %), tryptophan (131,0 %), sums of aromatic amino acids (129,83 %) and valine (122,0 %) and a bit less for leucine (111,43 %) and lysine (110,91 %) (**Table 9)**.

Addition of milk thistle shot to kefir didn't cause changes of the biological value of the protein component of the combined product (**Table 9**). It is proved by the mean value of scores of the control (129,3 %) and experimental (127,9 %) samples of kefir. We must note some growth of valine score (3,6 %). So, the combined product is characterized by the balanced amino acid composition.

Table 9

Biological value of proteins in kefir with milk thistle shot

Amino acids	Standard protein FAO/WHO g/100 g of protein	Kefir samples			
		Control		Experimental	
		g/100g of protein	Amino acid score, %	g/100g of protein	Amino acid score, %
Isoleucine	4,0	5,52	138,0	5,48	137,0
Leucine	7,0	9,83	140,43	9,59	137,0
Sum of aromatic (phenylalanine+ tyrosine)	6,0	10,18	169,67	9,90	165,0
Sum of sulfur-containing (methionine+cystine)	3,5	3,48	99,43	3,42	97,7
Threonine	4,0	4,00	100,0	4,01	100,3
Valine	5,0	4,48	89,6	4,67	93,4
Lysine	5,5	8,34	151,64	8,06	146,5
Triptophan	1,0	1,46	146,00	1,46	146,0
Totally					

7. Conclusions

Kefir with milk thistle shot with fms 2,5 % corresponds to the actual standard SSU 4417:2005. Kefir. Technical conditions by organoleptic and physical-chemical parameters.

Kefir with milk thistle shot is a treating prophylactic product, because it contains vegetable biologically active substances.

The total sum of amino acids in kefir with milk thistle shot grew – by 11,6 %, including irreplaceable – by 10,1 %, replaceable – by 12,6 %, that testifies to its biological value.

It is recommended to add milk thistle shot, in amount 3 % of a mass of the normalized mixture in kefir at its cooling and mixing before package.

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