Current trend in the development of biotechnology of a specialized dairy product for nutrition of athletes

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Abstract. The paper presents the analysis of modern domestic and foreign scientific and technical literature that addresses organization of healthy nutrition for athletes. The results obtained allow us to conclude that most of the studies and experiments have been performed in accordance with the target activities of athletes in a particular sport with different physical loads and specific diseases that reduce effectiveness of their training cycle. Relevance and significance of the development of the biotechnology for specialized food products for nutrition of athletes and professionals, as well as amateurs and the population regularly engaged in physical exercises is defined. The aim of the study is formulated, study objects and methods are described, scientific substantiation is provided, and a current trend in the development of biotechnological parameters of a specialized dairy (curd) product recommended for nutrition of athletes is determined. The study optimized the elemental composition of a new type of specialized product with two species of starter culture with probiotic cultures and special ingredients (concentrate of Milki land-WPC 80 whey proteins, glutamine, pollen) used to enrich the new product with free irreplaceable amino acids, vitamins, and minerals. This increased its digestibility and stability of the athlete’s gastrointestinal tract due to a specialized product that contains immobilized probiotic cultures in an amount of at least \(1 \cdot 10^8\) CFU. Chemical, amino-acid and vitamin composition of specific ingredients is presented, characteristics of their macro- and microelement composition are provided, and the relevance of their use is substantiated. Technological parameters for manufacturing a new product are determined, and a block diagram of the technology for manufacturing a curd product for the nutrition of athletes is presented.

1. Introduction

In complicated conditions of the incidence of COVID-19 throughout the world, in Russia, in countries of America, the European Union, Canada, Australia, and the Kingdom of Norway, the population shows an increased practical interest in a healthy lifestyle. A healthy lifestyle implies regular exercise, professional, amateur sports, fitness, and organization of regular balanced and specialized meals [1, 2]. Technological innovations in the development of food products for functional purposes and specialized food products are reported in numerous publications [3–10]. Both in Russia and abroad, scientists, specialists and nutritionists are engaged in the study of the effect of specialized nutrition on actual achievements.
of athletes [11]. However, commercially available supplements can be roughly divided into three different categories: health supplements, sports nutrition, performance supplements. Athletes and coaches choose medical supplements and sports nutrition with reliable information on their composition and purpose [12]. Most athletes’ diets are planned with regard to the tasks faced by the athlete in particular sport.

D. Sekulic et al. [13] believe that feeding has a significant impact on physical form and success of an athlete. Canadian athletes, of whom 88.4% use dietary supplements (DSU), vitamin C, protein, multivitamins and minerals, show consistent success [14]. In England, bodybuilding, where an athlete strives to acquire aesthetics, that is, muscle size, proportions and appearance, is very popular with professionals and amateurs. To keep fit, during the competition period, in addition to the usual diet, professionals consume foods rich in protein and calories, more carbohydrates and less fat to build and maintain the muscle mass [15]. In 2015–2017, G.L. Trakman et al. [16] developed a nutritional system, including the Nutrition for Sport Knowledge Questionnaire (NSQK) and Abridged Nutrition for Sport Knowledge Questionnaire (ANSQK), which aroused great interest among specialists. The study of the characteristics and properties of milk and dairy products expands the range of products for special purposes [17–20]. To obtain specific recommendations for improving the diet of athletes, scientists, together with specialists in dietitians of athletes, most often study the effect of milk, dairy products and milk proteins on performance and health of people, including athletes [21–30]. Analysis of foreign scientific and technical literature on healthy nutrition of athletes allows us to conclude that most studies and experiments have been performed in accordance with the target activities of athletes in a particular sport, who are exposed to different physical loads and have specific diseases that reduce the effectiveness of their training cycle. In this case, a healthy lifestyle program should imply that a person engaged in both professional and amateur sports must be healthy and physically resilient; his body must be resistant to stressful situations and immune to unfavorable environmental factors and nervous stress. The microbiota of an athlete must be healthy to withstand varying levels of nutrient intake during training and competition periods.

The above allows us to consider the development of the specialized food products biotechnology for nutrition of athletes, professionals, amateurs and the population regularly engaged in physical exercises relevant and significant.

The aim of the study is to develop a specialized dairy product biotechnology for nutrition of athletes.

2. Materials and methods

The main object of the study was raw cow’s milk in accordance with GOST 31449-2013. Starter cultures were used as biological objects:

- Probat 576 FRO500 DCU, Lactobacillus acidophilus, Lactococcus lactis, Lactococcus cremoris, Lactococcus biovar diacetilactis, and Leuconostoc mesenteroides cremoris cultures;
- AKO-1, the main cultures of Lactobacillus bulgaricus and Streptococcus thermophilus;
- Howaru Bifido FRO 100 DCU, culture of Bifidobacterium lactis;
- biopolymers: gelatin, pectin.

Special ingredients used were:
- whey protein concentrate Milkiland-WPC 80;
- glutamine in accordance with the current regulatory documentation;
- pollen in accordance with GOST 28887-90.

In experimental studies, standard research methods were used.

Immobilization experiments were carried out in a special box in the following sequence:
- activation of the biomass of cells of probiotic cultures in skim milk sterilized and cooled to a temperature of (38±1) °C, since the optimal temperature of the monocultures included in the association is (38±1) °C;
- preparation of a mixture of biopolymers performed at 20 °C;
- in the reactor, the association of the activated probiotic cultures was combined at a temperature of (33±1) °C with gel of biopolymers stirred for (15±5) min;
- the resulting mixture was dispensed into sterile forms;
- the forms were held in a special box is 15–20 minutes. As a result, thin films (membranes) were generated in the forms. Membrane were stored at a temperature of (4±2) °C.

The experiments were carried out in five repetitions. The results were processed by methods of mathematical statistics using standard MathCAD-14 professional software.

3. Results and discussion

Specialized food products should be considered as an essential addition to the diet of athletes or a standard diet of the population who are regularly engaged in sports and experience increased physical activity. It should be noted that an important and necessary component of the daily diet of all age groups of the population, including athletes, is milk and dairy products, whereas fermented dairy products prevail, including drinks, cottage cheese, curd products, and soft and semi-hard cheese [11].

Based on the above, low-fat cottage cheese was chosen as the milk base for production of a new product. Cottage cheese is a useful, dietary food product of high biological value, since it contains a large amount of proteins, essential amino acids, minerals, including calcium and phosphorus, and water-soluble vitamins [12].

The biological value and special properties of the curd product are due to the functional and special ingredients used. These ingredients increase immunity and endurance during training and competitive periods, contribute to a rapid recovery of the athlete’s body after exertion and to a stable work of the gastrointestinal tract owing to probiotics in the immobilized form. During development of biotechnological parameters for the new curd product, whole milk was heated up to (45±5) °C, subjected to bacterofugation and separated using a cream separator to produce skim milk with a mass fraction of dry substances of (9.5–9.8)% and cream with a mass fraction of fat of (15.0±0.5) and (20.0±0.5)%.

Skim milk was pasteurized at (90.0±2) °C, cooled to (30.0 ± 2) °C, and then low-fat cottage cheese was produced by the acid-rennet method with separation of the milk-protein base using a curd separator. The base had the following chemical composition,%; moisture (78–80); proteins (18.2–18.5); fat (1.0–1.1); carbohydrates (1.6–1.8); mineral substances (1.20–1.22).

Cream was pasteurized at (95±2) °C with an exposure time of 5 minutes and cooled to the fermentation temperature of (37±1) °C. Probat 576, ARO-1 and Howaru Bifido probiotics were added to the cooled mixture in a 1:1 ratio. After that, the cream was stirred for 30 minutes and left for fermentation.

The purpose of this stage of the study is to determine the type of cream and starter culture for subsequent use in production of a specialized curd product.

The process of cream fermentation was studied with regard to the following criterion requirements: fermentation time, active acidity (pH=4.6±0.5), pure creamy taste and low-viscosity consistency for uniform distribution in skim curd. The results of the study of cream fermentation are presented in tables 1 and 2.

### Table 1. Results of the study of fermentation of cream with fat content of 15.0 and 20.0% using Probat 576 and Howaru Bifido leaven.

| Duration of fermentation, h | Active acidity, pH | 
|---------------------------|-------------------| 
|                           | cream with fat content of 15 % | cream with fat content of 20 % |
| 0                         | 6.5               | 6.55               |
| 1                         | 6.3               | 6.40               |
| 2                         | 6.1               | 6.20               |
| 3                         | 5.5               | 5.55               |
| 4                         | 4.8               | 4.75               |
| 5                         | 4.6               | 4.50               |
Table 2. Results of the study of fermentation of cream with fat content of 15.0 and 20.0% using ARO-1 and Howaru Bifido leaven.

| Duration of fermentation, h | Active acidity, pH | Fat content, % | Viscosity, mPa·s |
|----------------------------|-------------------|----------------|-----------------|
|                            | cream with fat content of 15 % | cream with fat content of 20 % |
| 0                          | 6.50              | 6.55           |
| 1                          | 6.30              | 6.40           |
| 2                          | 6.10              | 6.20           |
| 3                          | 6.00              | 6.10           |
| 4                          | 5.80              | 5.85           |
| 5                          | 5.50              | 5.53           |
| 6                          | 4.90              | 4.95           |
| 7                          | 4.70              | 4.80           |
| 8                          | 4.65              | 4.60           |

Sensory properties of the tested products indicate that cream with fat content of 20% is more viscous and exhibits stable consistency compared to cream with fat content of 15%, which is more applicable to further product.

To confirm the results, the rheological properties of the tested products were studied on a RheomatR180 rotational viscometer at 10 ºC. The viscometer measures the torque resistance of the test product to rotation of the spindle of the measuring device at different rotation speeds and calculates the dynamic viscosity (table 3).

Table 3. Results of determination of the viscosity in fermented cream with fat content of 15 and 20 %.

| Experimental group No. | Leaven | Fat content, % | Viscosity, mPa·s |
|------------------------|--------|----------------|-----------------|
| Experimental 1         | Probat 576 + Howaru Bifido | 20 | 900 |
| Experimental 2         | ARO-1 + Howaru Bifido        | 20 | 850 |
| Experimental 3         | Probat 576 + Howaru Bifido  | 15 | 750 |
| Experimental 4         | ARO-1 + Howaru Bifido        | 15 | 740 |

Microbiological indicators of fermented cream with fat content of 15.0 and 20.0% are shown in figure 1.

Analysis of the experimental data of fermentation of cream with fat content of 15 and 20% and processing of the mathematical data show that it is more efficient to use milk cream with fat content of 20%, which is fermented with a two-component starter culture that consists of Probat 576 manufactured by Danisco (France) and Howaru Bifido.
To improve the protein composition of the product, we chose Milkiland-WPC80 whey protein obtained by ultrafiltration. The concentrate increases biological value of the product since it contains almost all essential amino acids. In addition, essential amino acids (Milkiland-WPC80) immediately enter the muscles for synthesis of new muscle tissue upon entering the human body. Milkiland-WPC80 has a positive effect on the immune system and optimizes insulin secretion (tables 4, 5 and 6).

Table 4. Chemical composition of whey protein concentrate.

| Component       | Content, % |
|-----------------|------------|
| Protein         | 80         |
| Fat             | 8          |
| Carbohydrates   | 6          |

Table 5. Characterization of macro- and micronutrient composition of whey protein concentrate.

| Component       | Content, mg |
|-----------------|-------------|
| Vitamins        |             |
| D, meg          | 2.050       |
| F               | 0.052       |
| Macronutrients  |             |
| K               | 384.000     |
| Ca              | 383.000     |
| Mg              | 56.400      |
| Ph              | 264.000     |
| Trace elements  |             |
| Zn              | 0.430       |

Table 6. Amino acid composition of whey protein concentrate.

| Essential amino acids | Content g/100 |
|-----------------------|---------------|
| Arginine              | 2.71          |
| Valine                | 5.71          |
Histidine  
Isoleucine  
Leucine  
Lysine  
Methionine  
Threonine  
Phenylalanine  

Essential amino acids

Alanine  
Aspartic acid  
Glycine  
Proline  
Serine  
Tyrosine  
Cysteine

The use of glutamine promotes synthesis of protein and glycogen in the human body, reduces the catabolic effect of glucocorticoids on muscles, and has a positive effect on the body’s recovery after physical exertion, which makes the product functional. The introduction of pollen increases the prophylactic effect since it is rich in dietary fiber and A, E, C, D, K vitamins, which have a positive effect on the immunity and contributes to the accelerated recovery of the body after physical exertion. In addition, introduction of vitamin-rich pollen increases the nutritive value of the product, which has a positive effect on the immunity and contributes to the accelerated recovery of the body after physical exertion (Table 7).

**Table 7. Vitamin composition of flower pollen.**

| Vitamin                | Content, mg /100 g dry matter |
|------------------------|-------------------------------|
| Carotene (A)           | 0.66-212.00                   |
| Thiamine (B₁)          | 0.55-1.50                     |
| Riboflavin (B₂)        | 0.50-2.20                     |
| Nicotinic acid (B₅, PP)| 1.30-2.10                     |
| Pantothenic acid (B₃)  | 0.32-5.00                     |
| Pyridoxine (B₆)        | 0.30-0.90                     |
| Biotin (H)             | 0.06-0.60                     |
| Folic acid (B₉)        | 0.30-0.68                     |
| Inositol (B₈)          | 188.00-228.00                 |

The amount of special ingredients was determined experimentally and then added to the cream before pasteurization. Figure 2 presents a block diagram of the technology for manufacturing a curd product for the nutrition of athletes.
Figure 2. Block diagram of the technology for manufacturing a curd product for the nutrition of athletes.

4. Conclusion
The results of analytical and experimental studies were used to provide scientific substantiation and to determine the current trend in the development of biotechnological parameters of a specialized dairy (curd) product recommended for the nutrition of athletes. The formulation of a new specialized product was optimized by using two types of probiotic cultures and special ingredients, which enriched the product with essential amino acids, vitamins, and minerals. In addition, consumption of a specialized product containing immobilized probiotic culture in an amount $<1 \times 10^8$ CFU/g increased its digestibility and stability of the athlete’s gastrointestinal tract.

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