ISSN 2074-9414 (Print)
ISSN 2313-1748 (Online)

https://doi.org/10.21603/2074-9414-2021-1-188-195
Original article
Available online at http://fptt.ru/eng

Quality Control of Colostrum and Protein Calf Milk Replacers

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Received: November 17, 2020 *Accepted: December 14, 2020*

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Abstract.
Introduction. Improving technologies and providing young farm animals with high-quality feed are the primary tasks for successful reproduction and maintenance of dairy cattle. The research objective was to assess the quality characteristics of colostrum and milk replacers, as well as their technological prospects.
Study objects and methods. The research featured colostrum, calf milk replacers (CMR), processing methods, and quality characteristics. The paper introduces an analysis of various sustainable processes of obtaining new CMRs.
Results and discussion. The article describes colostrum: recommended intake for young calves, qualitative characteristics, and control methods. It focuses mostly on the microbiological characteristics of colostrum, as well as on its role in developing the immune system of calves and the prospects of enzymatic regulation of its functional properties. Enzymatic regulation is based on deep proteins hydrolysates and a highly active serine protease (alcalase). The authors studied variants of using various enzyme preparations and bacterial starter cultures for obtaining hydrolyzed and fermented colostrum, analyzed the main process indicators of milk replacers with intermediate moisture content, and tested various methods for assessing the fatty acid and protein composition of concentrated milk replacers. Production methods proved to have a significant impact on the indicators in question.
Conclusion. Reproduction of the dairy herd genetic potential depends on the diet of the young farm animals, and so does the economy of agricultural production. Enzymatic processing of raw materials proved to be the most promising approach for obtaining products with improved functional properties. Deep colostrum hydrolysates can also be an important part of functional foods for children, athletes, in dietary foods, etc.

Keywords. Colostrum, whole milk replacers, hydrolysis, antigenicity, immunoglobulins, enzymes, fatty acid composition

Funding. The research was part of a state assignment of the Ministry of Science and Higher Education of the Russian Federation (Minobrnauka), topic No. AAAA-A20-120011500098-1.

For citation: KharitonovVD, AsafovVA, IskakovaEL, TankovaNL, HalavachTM, Kurchenko VP. Quality Control of Colostrum and Protein Calf Milk Replacers. Food Processing: Techniques and Technology. 2021;51(1):188–195. https://doi.org/10.21603/2074-9414-2021-1-188-195.

УДК 637.1
Оригинальная статья http://fptt.ru

Регулирование качественных показателей молозива и белковых композиций заменителей молока

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Дата поступления в редакцию: 17.11.2020 *Дата принятия в печать: 14.12.2020*

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Аннотация.
Введение. Совершенствование технологий, связанных с обеспечением молодняка сельскохозяйственных животных

качественными кормами, направлено на обеспечение воспроизводства и поддержание молочного стада. Цель работы – комплексная оценка качественных характеристик молозива и заменителей молока, а также технологических подходов в этой области.
Объекты и методы исследования. Молозиво и заменители цельного молока (ЗЦМ), методы их обработки; анализ процессов получения новых видов ЗЦМ, направленных на ресурсо- и энергосбережение.
Результаты и их обсуждение. Приведены потребности телят в молозиве, обсуждены качественные характеристики первичного молока и подходы к их регулированию. Особое внимание уделено микробиологическим показателям первичного молока, значению молозива в формировании иммунитета молодняка. Охарактеризованы возможности ферментативного регулирования функциональных свойств молозива в результате гидролиза белков с применением высокоактивной эндопептидазы (алкалаза). Рассмотрены варианты использования различных ферментных препаратов и бактериальных заквасок для получения гидролизованного и ферментированного молозива. Проведен анализ основных процессовых показателей ЗЦМ с промежуточной влажностью. Обоснована целесообразность применения ряда методик для определения жирнокислотного и белкового состава концентрированных ЗЦМ. Отмечено, что способы получения ЗЦМ оказывают существенное влияние на значения анализируемых показателей.
Выводы. Воспроизводство и реализация генетического потенциала молочного стада, а также экономика сельскохозяйственного производства определяются факторами, связанными с питанием молодняка сельскохозяйственных животных. Ферментативная обработка сырья является перспективным подходом для получения продуктов с улучшенными функциональными свойствами. Практическое применение глубоких гидролизатов молозива представляется целесообразным, в том числе в составе специализированных продуктов детского, спортивного и диетического питания.

Ключевые слова. Молозиво, заменители цельного молока, гидролиз, антигенность, иммуноглобулины, ферменты, жирнокислотный состав

Финансирование. Работа выполнена в рамках государственного задания Министерства науки и высшего образования Российской Федерации (Минобрнауки России) (тема № АААА-А20-120011500098-1).

Для цитирования: Регулирование качественных показателей молозива и белковых композиций заменителей молока / В. Д. Харитонов, В. А. Асафов, Е. Л. Искакова [и др.] // Техника и технология пищевых производств. – 2021. – Т. 51, № 1. – С. 188–195. https://doi.org/10.21603/2074-9414-2021-1-188-195.

Introduction

Dairy production depends on the dairy cattle reproduction, while effective calf management depends on the structure and quality of feeds early in life. Ideally, good-quality colostrum, milk, or its replacers should provide 9–12 g of daily liveweight gain per 1 kg. Poor weight gain is associated with poor immunity. However, if daily liveweight gain exceeds 15 g, it often causes obesity and affects the development of the mammary gland, which reduces milk production in adulthood.

Good-quality calf starters prolong dairy cow's productive lifetime and increase its fertility. Second, third, and fourth calves are known to grow into more valuable cows capable of higher milk yields and fertility. The determining factors for effective calf rearing are their health, weight, and first calving age, which depend on the quality of colostrum and feeds in the first three months of life [1].

During this time, one calf consumes 300–350 L of milk. However, feeding whole milk to dairy calves reduces the marketability of dairy production and increases the cost of herd management.

Calf milk replacers (CMR) can increase the marketability and reduce these costs [2]. In Russia, CMR market volume is 120 000 tons per year. The market demands are satisfied by 30%, of which import accounts for 20%.

Russia possesses sufficient resources of milk whey to produce CMRs. The rapidly increasing pork and soybeans production can provide animal fat and high-quality vegetable protein for CMR formulations.

A good CMR fulfills the following functions:
– providing controlled weight gain;
– replacing as much milk in calves' diet as possible;
– offering opportunities for using any whey, except for salted one;
– introducing fat in sufficient quantity;
– ensuring high microbiological indicators; and
– increasing productivity while demanding little investment.

CMRs for young farm animals can be classified according to the main components:
– skim milk and vegetable/animal fats;
– whey protein concentrates and vegetable/animal fats; and
– whey, vegetable protein concentrates, and vegetable/animal fats.

These three CMRs have the same feeding efficiency, but the plant-based CMRs are more economical. They include protein concentrates that need special quality standards. For instance, they should contain a particular set of available amino acids but no such antialimentation factors as oligosaccharides, allergenic proteins, various inhibitors that might affect feed digestion, etc. These factors can be eliminated by using special technologies, e.g. protein purification or molecular weight change. To ensure proper development of skeletal system,

a good CMR should contain available macro- and microelements, which need to be introduced in such a way as to ensure effective utilization.

Acid whey poses a certain challenge: it needs acidity regulators that provide the required osmotic pressure in the intestinal tract of animals.

If all the above factors are taken into account, new technologies and formulations of plant-based CMRs can ensure necessary weight gain and productivity.

As for soy-based CMRs, which are based on soy flour, milk, and cereal flour, they have a lot of disadvantages as they cannot be applied to very young animals and may reduce their productivity in the future [1, 2]. The research objective was to provide comprehensive assessment of the qualitative indicators of colostrum and milk substitutes, as well as to define possible technological options in this area.

Study objects and methods

The studies were carried out on the premises of two organizations: Laboratory of Resource-Saving Processes and Functional Products at the All-Russian Scientific Research Institute of the Dairy Industry (Moscow, Russia) and the Research Laboratory of Applied Biological Issues at Belarusian State University (Minsk, Belarus).

The research featured colostrum, and calf milk replacers (CMR) produced by the All-Russian Research Institute of Dairy Industry. The assessment methodology was based on physicochemical, biochemical, and sanitary-hygienic indicators, as well as on the criteria for the optimal weight gain of baby calves. The study made it possible to define the efficiency factors, to develop control methods for quality indicators and composition, and to analyze the processes of obtaining new resource-saving and energy-efficient CMRs [3].

The study involved contemporary approaches to defining the biologically active properties of native, fermented, and hydrolyzed colostrum, i.e. the fluorometric method for antioxidant activity, the impedimetric method for antimicrobial effect, and the Ames test for antimutagenic effect [4, 5].

Results and discussion

Colostrum quality assessment. Colostrum is a valuable source of highly-concentrated biologically active substances of protein nature:

– lysozyme, lactoperoxidase, and lactoferrin that perform the functions of nonspecific immunity;

– immunoglobulins responsible for specific protection in the first hours after birth; and

– transfer factors that serve as a signaling environment for triggering the mechanism of specific body defense.

The quality of colostrum defines health status, weight gain, and calf raising economy. The list of important factors includes the IgG content, bacterial contamination, and storage conditions.

High-quality colostrum for calf feeds should meet the following requirements: IgG content ≥ 50 g/L of immunoglobulins; amount of colostrum = 10% of the body weight (≈ 4 L); feeding time after calving ≤ 6 h; microbiological purity ≤ 100 000 CFU/mL [6, 7].

Bacterial contamination of colostrum can cause various diseases. Moreover, bacteria prevent immunoglobulin absorption. In this regard, microbiological control methods for colostrum microflora are priority scientific issues because colostrum quality on dairy farms needs to be improved. Heinrichs *et al.* studied heat treatment of colostrum in order to reduce bacterial and pathogenic growth while increasing IgG absorption. The scientists also established the upper limit of immunoglobulin absorption from colostrum and its substitutes, as well as assessed the possibility of introducing lactoferrin and sodium bicarbonate into maternal colostrum [8].

The IgG concentration in the blood was found to affect calves' resistance to diseases. The amount of absorbed immunoglobulins depends on their consumption with colostrum, as well as on the efficiency of absorption from the intestinal tract [9–11].

The calf should receive the first portion of colostrum within the first hour after birth, and it should be at least 70 mL/kg of body weight. The rate of feeding colostrum during the first day depends on the IgG content (Table 1) [12].

Colostrum immunoglobulins that a calf receives in the first hours after birth lengthen the period of passive immunity. Endogenous antibody synthesis sufficient to protect calves from infectious diseases usually develops at 1–3 months [9, 10].

A sufficient amount of high-quality colostrum allows lacto- and bifidobacteria to colonize the intestines. The normal intestinal microflora of calves consists of equal numbers of lactobacilli, bifidobacteria, and Escherichia, while the staphylococcus population is half as low [13].

Colostrum contamination occurs in the udder or teat canal. It can also be caused by poor sanitary conditions. Bacterial contamination of colostrum starts at 100 000 CFU/mL, but experts notice negative effects already at 50 000 CFU/mL [14].

Table 1. Amount of colostrum in calves' diet, depending on the age and IgG concentration

Time after birth, h	Colostrum feeding rate at a particular IgG concentration, L			
	25 g/L	50 g/L	75 g/L	100 g/L
1	4.0	2.0	1.3	1.0
3	—	2.5	1.6	1.3
6	—	2.9	1.9	1.5
9	—	—	2.2	1.7
12	—	—	2.5	1.9
15	—	—	2.8	2.2
18	—	—	—	2.4

Low-temperature treatment at 63–66°C decreases the level of microbiological contamination of colostrum without affecting its immunomodulatory properties [15].

These methods reduce colostrum deficiency by reserving it from healthy cows starting with the third lactation. Mature cows produce better colostrum compared to heifers. The density of their milk is higher by 0.02 g/cm^3, IgG content – by 73.4–122.2 mg/mL, IgA and IgM – by 8 and 6 mg/mL, respectively, and the mass fraction of dry matter – by 2.4% [12].

One of our previous research showed that raw colostrum obtained from farms in the Moscow Region did not meet the above requirements for microbiological parameters [3]. One of our previous studies featured the possibility of reducing the bacterial contamination of colostrum by its fermentation with *Lactobacillus acidophilus* (strain No. 630, not viscous). The method improved microbiological indicators, increased the amount of lactic acid cultures, and raised the antioxidant activity of fermented colostrum because the bacterial proteolytic system was broken down by proteins [4].

Colostrum surpasses whole milk in nutritional value and composition of biologically active components [16–19], which makes it a promising raw material for functional and diet foods for children, athletes, etc. One of our previous studies also featured the physicochemical and biologically active properties of colostrum fermented with *L. acidophilus* and treated with proteolytic enzyme (alkalase) [4].

The research defined the peptide composition, antioxidant activity, antimutagenic properties, and antimicrobial action of skim colostrum samples subjected to hydrolysis and fermentation. Enzymatic hydrolysis appeared to provide better proteolysis (17.4%) than fermentation with *L. acidophilus* (7.5%). The antioxidant activity of the peptide fractions of hydrolyzed and fermented colostrum increased by 4.1 and 2.0 times as compared to the original colostrum (dry solids content). As the proportion of the peptide fraction and the degree of colostrum proteolysis increased, the antiradical activity increased and the level of induced mutation (in the Ames test) decreased. Low molecular weight fractions of hydrolyzed colostrum developed specific peptides with antimicrobial activity.

As a result, they were found to be more effective against *Escherichia coli*, test strain ATCC 8739, than against *Staphylococcus aureus*, ATCC 6538 gram-positive strain [4]. Complexation of hydrolysates with cyclic oligosaccharide (β-cyclodextrin) made it possible to preserve the antimutagenic effect, increase the antibacterial effect of colostrum peptides, and improve their organoleptic properties [5]. The research results proved relevant when introducing inclusion complexes into functional products.

The abovementioned studies allowed us to obtain samples of hydrolyzed and fermented colostrum with confirmed biologically active properties [4, 5]. Therefore, fermentation and hydrolysis proved able to regulate the functional properties of products, which can be used in developing new feeds for young farm animals.

Analysis of approaches to assessing the quality indicators of new types of milk replacers. Various CMRs can increase marketability and reduce costs in dairy farming [3, 20].

The types of CMRs that are popular in contemporary dairy farming are based on:
– skim milk and non-dairy fats;
– whey protein concentrate and non-dairy fats;
– whey and specialized concentrates and isolates of soy protein and non-dairy fats; and
– whey, soy flour, and non-dairy fats.

The first three types are complete balanced feeds adapted to the needs of calves. They provide an average daily weight gain of 650–900 g. The fourth is intended for older calves. For an adapted feed, it contains too much lectins, oligosaccharides, and allergens.

According to the production method, CMRs are divided into whole milk replacer, which is a liquid product dried on a spray dryer, and regenerated milk prepared by dry mixing of ingredients, including fat.

Each method has its own advantages and disadvantages. Regenerated milk is more technologically advanced in terms of production. However, fats have to be introduced since calf feeds should not contain free fats. If free facts are present in the feed, calves absorb it poorly and develop slowly. As a result, regenerated milk has a low fat percentage (12%). The fat:protein ratio is 0.5:1, while in milk it is 1.2:1. Thus, calves may not receive the energy necessary for growth and have to consume more feed to replenish it.

The main condition for the CMR production is that replacers should be cheaper than whole milk and provide a required growth rate.

The correct economic assessment of CMR cost is the cost of feed per 1 kg of weight gain. It justifies the use of whey-based replacers, specialized concentrates, and isolates of soy proteins and non-dairy fats. These CMRs are as effective as CMRs based on skim milk, but their raw material composition is significantly cheaper. Low energy consumption and specific capital investments are important factors in CMR production.

CMRs with intermediate moisture are quite advantageous as they make it possible to:
– reduce spray drying costs;
– use any kind of whey, except for salted one;
– add fat in any required amount;
– start a low investment production;
– use methods of fermentation of protein fractions;
– obtain products with specified probiotic properties;
– achieve good recombination before use; and
– obtain a product with a high degree of fat emulsification and a minimum content of free fat.

CMR production for young farm animals has acquired industrial scales in most developed countries. In Russia, it started in the 1970s [20, 21].

Specialists dealing with CMR technologies focus on regulating their composition and functional properties and calculating the rational content of the mass fraction of solids. One of the topical areas of research is the development of new methodological approaches to the CMR technology, e.g., how to avoid drying stage and produce concentrated CMR. The production methodology and quality assessment of concentrated CMRs include the following stages:

1. Technological and processing options and parameters for obtaining various products based on dry and concentrated CMRs. Parameters that provide the required levels of optimization, composition, quality, and physicochemical properties involve technological modes at all production stages, including biotechnological transformation of raw materials components during processing.

2. Parameters that ensure increased ecological safety, as well as low energy and resource consumption.

3. Interconnection between quality indicators and properties of dry and concentrated products, production modes, and energy consumption.

4. Parameters that assess the end use based on objective testing, including zootechnical research.

5. Indicators of economic efficiency.

This methodology requires the basic technological regimes that determine the optimal ranges of the mass fraction of dry substances at various stages of dehydration and in the final product. These technological modes depend on the physicochemical and thermophysical properties of the product during production [22].

These specific features mean that the methodology for concentrated CMR production must observe some specific approaches. For instance, the efficiency of dehydration decreases during the first stage following the increase in the concentration of the mass fraction of dry substances. During vacuum evaporation, the heat transfer coefficient decreases as the mass fraction of dry substances increases during thickening. Another limiting factor is that viscosity increases together with the concentration of the mix, which increases energy consumption and can terminate the dehydration process [20].

Dehydration usually presupposes a two-stage scheme of sequentially mounted reverse osmosis units. As a result, it is possible to estimate the approximate permissible concentrations of the condensed product. For example, the limiting concentration of milk whey is 25% when using reverse osmosis plants and 57% when using vacuum evaporators. These values depend on the properties of the mix before concentration and require experimental testing.

The mass fraction of the dry matter of concentrated CMRs can be controlled by adding additional components to the mix – dry or texturized. These processes in combination with direct methods of concentration make it possible to control the mass fraction of dry substances in a rather wide range. The upper values are affected by viscosity, gelling properties, and stratification, which depend on temperature and some other factors.

Another important task of concentrated CMR production is to achieve maximal storage stability.

This indicator can be improved using several effective techniques, e.g. pasteurization or cold storage, which inhibits bad microflora. Other methods of inhibiting pathogenic microflora include fermentation, preservatives, heat sterilization, and ultraviolet treatment [20].

From the point of view of production economy, long shelf life is not important since it increases additional costs for intermediate storage and related equipment. The optimal storage temperature for milk replacer is determined by its delivery radius and the technical capabilities of each customer.

The approximate storage time for concentrated CMRs is based on some assumptions.

The first assumption is that the production time and use in practical conditions do not fluctuate. For example, the production time does not exceed two days, even taking into account its storage at the plant and a 20% time cushion. The time the CMR spends on the farm is approximately the same. Thus, the rational value of the storage stability of concentrated CMRs depends only on the delivery radius and time. The delivery time takes no more than one day within a radius of up to 400 km. Therefore, delivery time hardly affects the need to increase the storage stability of concentrated CMRs.

What is more important is customer's interest in creating a stable, long-term supply of a concentrated CMR on the farm. As a result, producers may want to stock the CMR for the subsequent targeted delivery to the customer in particular amounts.

Like many other multicomponent products with intermediate moisture content, CMRs require a complex multifactorial production process that can be implemented in various ways using various technological and technical methods of raw material processing (Fig. 1).

Figure 1 shows that the basic principles of processing multicomponent products remain the same, while specific technological methods are constantly being improved. Recent studies in the field of animal and plant biotransformation revealed some new plant raw materials and more effective means that facilitate and control emulsification of vegetable fats and protein hydrolysis [4, 5, 23–31].

Such innovations help to achieve these goals (Fig. 1) if based on the analysis of a set of interrelated process parameters at all production stages.

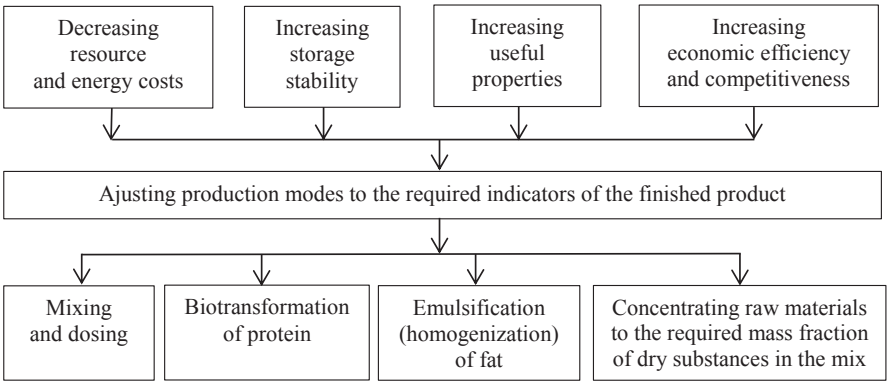

Figure 1. Basic processing procedures and required indicators of concentrated milk replacers

New types of raw materials and processing methods require new methods for assessing properties, quality, and physicochemical parameters of the product, e.g. the degree of emulsification of fats and changes in the antigenicity of plant proteins at particular production stages. For instance, the main indicators of high-fat CMRs include the dispersed composition of vegetable fats, assessed by the average diameter of the fat globules and their level of monodispersity in the CMR.

Another indicator is the protein coating of the fat globules. However, no official indicator has been developed so far for assessing protein globule membranes in concentrated CMRs. Qualitative assessment of this indicator and change patterns in fat stability in various processing modes are associated with dairy fat without protein membranes. In liquid dairy products, such fat is usually called destabilized, whereas in dry dairy products, it is called free fat with surface free fat [26]. However, these indicators are not decisive for assessing the properties of concentrated CMRs, for the reasons mentioned below.

Free fat without globule membranes produce almost no oxidative taste in dry whole milk during six months of storage, even at 30°C. In dairy fats, the rate of oxidative spoilage is highly inertial and depends on the state of fatty acids, antioxidants, oxygen contact surface, processing conditions, and storage conditions. In concentrated CMRs based on milk fats, their oxidation degree is always relatively low because they are based on fresh milk [27].

Taking into account these factors and the relatively short shelf life of concentrated CMRs, which does not exceed several weeks, the oxidation degree of the fat phase can be considered a limiting factor only for the initial value immediately after production. However, increased oxidation is an important factor when producing concentrated CMRs with non-dairy animal and vegetable fats.

As a result, the oxidation degree of non-dairy fats is a limiting factor: the values of this indicator should be equal to the dairy fat of pasteurized milk. Thus, when producing a concentrated CMR, the oxidation degree of fat phase should be determined before adding it to the mix, not before feeding it to baby calves. Based on the data mentioned above, the analysis of the total content of unprotected fat, destabilized or free, should take place after the methodology is improved and sample preparation is thoroughly studied.

Additional tests required for fat phase assessment include the assessment of its fatty and amino acids, as well as the dispersion of fat globules, which should correspond to the average size of fat globules in raw milk.

The assessment methodology for protein properties of a concentrated CMR also has its peculiarities because of its plant origin and different initial composition. In this regard, reducing the level of anti-nutritional substances in the protein composition is of great importance. As a result, assessing the antigenic properties of the protein and its polypeptide profile is also extremely important.

These indicators affect the zootechnical indicators when feeding milk replacers to young farm animals. Therefore, they should be controlled at various production stages, using traditional or novel techniques of biotransformation of the original protein raw material.

Conclusion

The main factor that determines the efficiency and economy of feed for young farm animals is the bioavailability of its protein and fat components.

The present research analyzed the qualitative indicators of colostrum and milk replacers. The analysis revealed that some of their physicochemical properties need additional control, i.e. the concentration of immunoglobulins, the polypeptide and amino acid profile of the protein component. The sanitary and hygienic indicators of primary milk can be controlled using various approaches, e.g. thermal inactivation or fermentation of colostrum with acidophilus bacillus, which reduces the number of pathogens.

The animal and vegetable fats that are part of milk replacers have some specific properties. Therefore,

their assessment should be based on the methods for determining the average diameter of fat globules, fatty acid composition, and the initial oxidation state of fats. A complex study of the fat phase requires methods that make it possible to establish the peptide and amino acid profile, as well as to assess the level of anti-nutritional and antigenic properties. In concentrated milk replacers, the qualitative and quantitative indicators of the protein and fat components depend on the methods and parameters their production.

Only by controlling the quality of colostrum and using high-quality whole milk replacers, farmers can provide rapid weight gain, earlier calving, high productivity, and prolonged lactation.

Contributon

V.D. Kharitonov supervised the project and wrote the paragraph about concentrated milk replacers. V.A. Asafov collected the material on colostrum and wrote the respective paragraph. E.L. Iskakova performed the experimental research on the hydrolysis of plant proteins, processed the obtained data, and described the results. N.L. Tankova performed the experimental studies of colostrum fermentation, processed the data, and described them. T.M. Halavach conducted the experimental research on enzymatic colostrum hydrolysis, described the properties of the obtained peptides, processed the data, and described the results. V.P. Kurchenko supervised the project, collected research material, and performed its synthesis.

Conflict of interest

The authors declare that there is no conflict of interests regarding the publication of this article.

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