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Comparative Analysis of the Amino Acid Composition of Beverages Based on Plant Raw Materials and Cow's Milk

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Abstract. Knowledge of the peculiarities of technological processes for the production of cow's milk analogues, optimal parameters for production, the chemical composition of raw materials and the resulting finished product, in particular the amino acid composition, plays an essential role in the development of new types of functional products, improvement of their stability, taste, and nutritional properties, and allows them to be enriched with additional components through various modifications. The purpose of the study is to compare the amino acid composition and balance of amino acids of various types of beverages based on plant raw materials: almond, rice and coconut, oatmeal with respect to the amino acid composition of cow's milk protein. The following methods were used: capillary electrophoresis – to determine the amino acid composition of the protein in analogues of milk and cow's milk, mathematical – to calculate the amino acid score and utility coefficient. The composition, organoleptic quality indicators, main technological stages, and safety of production of plant-based analogues of animal milk are presented. Three types of plant-based beverages were used for the study: almond with a mass fraction of fat of 1.5%; oatmeal with a mass fraction of fat of 2.5%; rice and coconut with a mass fraction of fat of 1.8%; and cow's milk with a mass fraction of fat of 2.5%. A comparative estimate of the mass fraction of amino acids valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine in the protein of beverages based on plant raw materials is presented relative to the mass fraction of amino acids in the milk protein, and the reference protein. The limiting amino acid in the protein composition of the studied analogues of cow's milk is methionine. The highest amino acid protein scores were observed in oatmeal drink; however, they did not exceed those of pasteurised milk. To assess the balance of the amino acid composition, the utility coefficient was determined. The findings are of practical importance, because they can be used in the production of functional products, in particular, analogues of cow's milk, to improve the composition, increase biological value, and enrichment with nutrients and additional components

Keywords: cow's milk analogues, plant-based drink, oatmeal drink, almond drink, rice and coconut drink, amino acid score, utility coefficient

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INTRODUCTION

Nowadays, in the processing industry, there is a tendency to increase the segment of natural products for healthy nutrition, accompanied by a continuous expansion of protein processing, the search for new, potentially suitable raw materials. Skripnichenko *et al.*, (2020) found that for this purpose, raw materials of plant origin are often used, which can increase the nutritional and biological value of products and have therapeutic properties, etc.

Studies by Min *et al.* (2018), Merzlov *et al.* (2021) indicate that one of the priority areas of research in the field of nutrition is the production of Non-Dairy Milk – a drink based on plant raw materials, which was created as an alternative to animal milk, including to solve the problem of protein deficiency in economically undeveloped countries. However, such drinks are becoming increasingly popular today.

According to the authors of Khalupa-Krebzdak *et al.* (2018) the advantages of consuming such a drink are the absence of cholesterol, low caloric content, high content of fibre, isoflavonoids, antioxidants, monounsaturated, and polyunsaturated fats, which is important, especially when following a diet. R. Bocker *et al.* (2022), J. Poore *et al.* (2022), C. Short *et al.* (2021) found that the technological process for the production of beverages based on plant raw materials is environmentally friendly and contributes to reducing carbon emissions into the environment compared to the technology for the production of cow's milk.

With the constant consumption of plant analogues of milk, there is a possibility of a nutritional imbalance associated with protein and trace element composition. McClements *et al.* (2021) showed that such products are characterised by low digestibility of essential substances, and instability of the amino acid composition, compared with animal products, contain many antinutrients, such as trypsin inhibitors, phytic acid, and inositol phosphates.

Sarangapany *et al.* (2022), Silva *et al.* (2020) investigated various technologies for the production of plant-based beverages and found that for the stability of the amino acid composition and increasing the nutritional value of the final product, minerals, vitamins, various food additives are additionally applied to the product, for example, flavourings, dyes, stabilisers (guar gum, gellan gum, xanthine gum, etc.) or, as indicated in studies by Kundu *et al.* (2018), several types of raw materials can be used simultaneously. Sarangapany *et al.* (2022), Bocker *et al.* (2022) studying the mass fraction of protein and amino acid composition of beverages based on plant raw materials proved the dependence primarily on the production technology, in particular, the use of innovative processing technologies (ultrasound, high-pressure processing, the use of pulsed electric field, ultraviolet and microwave radiation), which affects the nutritional, organoleptic quality indicators and product safety, but no less important were the quality indicators of the raw materials used (variety, climatic conditions and geographical area of cultivation).

Studies by Han *et al.* (2021) and Sarangapany *et al.* (2022) showed that a mandatory stage in the production of cow's milk analogues is the extraction of water- and salt-soluble protein fractions from raw materials. First, pre-grinding of raw materials is carried out, followed by the preparation of a suspension and filtering it, to obtain a consistency similar to cow's milk. This technological operation ensures the colloidal stability of the product. McClements and Grossmann (2021) and Short *et al.* (2021) proved the importance of using technological operations of homogenisation and pasteurisation to increase microbiological stability, inactivation of enzymes, increase the storage time of the product, in addition to providing pleasant sensory properties of the product for the consumer. Pasteurisation is performed by heat treatment, but Aydar *et al.* (2020) suggest that the use of high temperatures (from 60°C to 130°C) changes the physical, chemical, and organoleptic properties and nutritional value of beverages. Analysing research data by Han *et al.* (2021), an extremely important step in the production of beverages based on plant raw materials is the choice of technological parameters, which is associated with the need to block or break down anti-nutrients: protease inhibitors, lectins (for example, during the use of legumes) specific alkaloids or other substances that can cause protein deposition. McClements and Grossmann (2021) investigated the technological operations of steaming, calcination, and soaking in slightly alkaline solutions, which can increase the efficiency of protein and dry matter extraction in beverages, increase the colloidal stability of the obtained emulsions in the production of beverages from various types of legumes and nuts.

Silva & Smetana (2022) found that one of the main factors influencing the quality of a plant-based drink is the temperature regime. During the production of animal milk analogues from legumes, the use of ultra-high temperatures allows inactivating trypsin, but the use of such modes at the stages of blanching and roasting affects the fractional composition of proteins, reducing their solubility and extraction indicators. The main criterion in assessing the biological value and physiological role of amino acids is their ability to provide protein synthesis in the body.

The purpose of the study is to investigate the amino acid composition of the protein of plant-based milk analogues: almond, rice and coconut, and oatmeal, and to conduct a comparative analysis of amino acid balance in relation to cow's milk protein. To achieve this goal, the following tasks were set: to investigate the amino acid composition of cow's milk analogues: rice and coconut, almond, and oatmeal; to compare the amino acid composition of beverages with the amino acid composition of cow's milk; to determine the amino acid score of the products under study and compare it with the amino acid score of cow's milk; to calculate the utility coefficient.

MATERIALS AND METHODS

For the study, samples were taken in accordance with DSTU 4856:2007 (2008) of three types of plant beverages: rice and coconut with a mass fraction of fat of 1.8%, almond and oatmeal with a mass fraction of fat, respectively, 1.5 and 2.5 %, and cow's milk with a mass fraction of fat of 2.5% in accordance with DSTU ISO 5538:2004 (2005).

The mass fraction of amino acids in the products was determined in the laboratory for the control of feed additives and premixes of the State Scientific Research Control Institute of Veterinary Medical Products and Fodder Additives in Lviv by capillary electrophoresis according to the method of I.Y. Kotsyumbas (2013). Capillary electrophoresis is based on electrokinetic phenomena — electromigration of ions and other charged particles and electroosmosis. The study was carried out using the Kapel-105/105M capillary electrophoresis system, equipped with special software based on a personal computer. The method is based on the hydrolysis of samples by the acid method (hydrochloric acid for 16 hours) with the transition of amino acids to free forms of phenylisothiocarbamyl derivatives (PTC derivatives), their further separation and quantitative determination by capillary electrophoresis. This method was used to determine the mass fraction of amino acids: lysine, phenylalanine, leucine and isoleucine, methionine, valine, and threonine. The obtained indicators were compared with the amino acid composition of cow's milk.

The biological value of proteins in beverages based on plant raw materials and cow's milk was determined using the method described by Peshchuk & Nosenko (2011) comparing the composition of products with the composition of the "ideal" protein and calculating the score by determining the ratio of the amount of each amino acid (g) in 1 g of protein of the studied product to the amount of the same amino acid in 1 g of the "ideal" protein. The amino acid with the lowest amino acid score is an amino acid that limits the biological value of protein.

Using the methods developed by academician N.N. Lipatov (2001), which are based on the development of the Mitchell-Block principle and allow estimating the amino acid composition, its balance and the degree of digestibility of amino acids. The ratio of the lowest amino acid score to the highest was determined (the utility coefficient was established) according to the method described by M. Hayes (2020).

RESULTS AND DISCUSSION

The amino acid composition of plant-based beverages was studied: oatmeal with a mass fraction of fat of 2.5%, almond with a mass fraction of fat of 1.5%; rice-coconut with a mass fraction of fat of 1.8 %; and cow's milk with a mass fraction of fat of 2.5%. It was found that the protein of pasteurised milk has a higher mass fraction of the amino acids leucine, valine, isoleucine, lysine, and threonine, respectively, by 39.7%; 15.6%; 42.7%; 43.1%, and 15.2%. The mass fraction of phenylalanine and methionine in cow's milk was 13.8% and 25.1% lower, respectively, compared to the reference protein index (Table 1).

***Table 1**. Amino acid composition of cow's milk and beverages based on plant raw materials M ± m, n = 12*

Name of amino acid	Mass fraction of amino acid, mg/1 g of protein				FAO/WHO reference protein, mg/1 g of protein
	Product name				
	Pasteurised milk	Oatmeal drink	Rice and coconut drink	Almond drink	
Valine	57.8 ± 4.41	32.4 ± 0.02	8.1 ± 0.03	20.0 ± 0.01	50.0
Isoleucine	57.1 ± 3.31	26.7 ± 0.01	5.7 ± 0.02	17.0 ± 0.03	40.0
Leucine	97.8 ± 6.26	53.0 ± 0.09	11.3 ± 0.01	35.0 ± 0.01	70.0
Lysine	78.7 ± 6.08	19.0 ± 0.03	5.4 ± 0.03	10.0 ± 0.02	55.0
Methionine, mg	26.2 ± 2.36	9.8 ± 0.005	3.5 ± 0.02	4.9 ± 0.02	35.0
Threonine, mg	46.1 ± 2.66	21.7 ± 0.02	7.5 ± 0.02	16.0 ± 0.01	40.0
Phenylalanine, mg	51.7 ± 4.64	36.5 ± 0.02	8.5 ± 0.04	26.9 ± 0.015	60.0

***Source:** compiled by the authors*

Comparing the amino acid composition of plant-based beverages with the amino acid composition of pasteurised milk and reference protein, there is a low mass fraction of amino acids: valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine, which is associated with the low content of these amino acids in the composition of plant proteins and the technological stage of production of this product (soaking and extraction process). In all the studied analogues of cow's milk, the predominance of the mass fraction of the amino acid leucine in comparison with other amino acids was noted. Thus, in oatmeal drink, the mass fraction of leucine exceeds the indicator of almond and rice and coconut drinks by 1.5 and 4.7 times, respectively, but its amount is significantly less than in pasteurised milk by 1.84 times.

In rice and coconut drink, the mass fraction of the amino acid leucine exceeds the mass fraction of

valine by 39.5%, isoleucine by 98.2%, lysine by 2.09 times, methionine by 3.2 times, threonine by 1.5 times, and phenylalanine by 32.9 %. The mass fraction of the amino acid leucine in almond drink exceeds the mass fraction of valine by 75.0%, isoleucine by 2.06 times, lysine by 3.5 times, methionine by 7.1 times, threonine by 2.2 times, and phenylalanine by 30.1%.

The findings indicate that the limit for all the studied analogues of milk is the amino acid methionine, the smallest mass fraction of which is in the rice and coconut drink. Compared to oatmeal and almond drinks, the amount of it in this product is less by 2.8 and 28.5%, respectively. In oatmeal drink, the mass fraction of this amino acid exceeds other indicators of plant-based beverages and amounts to 9.8 mg/1 g of protein and remains 2.6 times less compared to cow's milk. The amino acid score of pasteurised milk according to the following indicators: valine, isoleucine, leucine, lysine, and threonine exceeds 100, respectively, by 15.6%; 42.7%; 39.7%; 43.1% and 15.2%, which indicates the completeness of protein (Table 2).

***Table 2**. Amino acid protein score of pasteurised milk and plant beverages M ± m, n = 12*

	Amino acid score, %			
Name of the amino acid	**Pasteurised milk**	**Oatmeal drink**	**Rice and coconut drink**	**Almond drink**
Valine	115.6 ± 19.2	64.8 ± 0.024	16.2 ± 0.06	40 ± 0.011
Isoleucine	142.7 ± 21.3	66.7 ± 0.02	14.2 ± 0.068	42 ± 0.11
Leucine	139.7 ± 22.1	75.7 ± 0.027	16.1 ± 0.12	50.0 ± 0.13
Lysine	143.1 ± 23.5	34.5 ± 0.018	9.8 ± 0.031	18.2 ± 0.12
Methionine	74.9 ± 12.3	28.0 ± 0.017	10.0 ± 0.022	14.0 ± 0.07
Threonine	115.2 ± 19.1	54.2 ± 0.03	18.7 ± 0.06	40.0 ± 0.09
Phenylalanine	86.2 ± 14.5	60.8 ± 0.02	14.2 ± 0.072	44.8 ± 0.01

***Source:** compiled by the authors*

Drinks based on plant raw materials have a significantly lower amino acid score of all the amino acids under study. Analysing the obtained data, it is worth noting that the amino acid score of pasteurised milk protein is limited by the amino acid methionine and phenylalanine, the amino acid score of which is the smallest (<100%) and is 74.9% and 86.2%, respectively. Deviation of the indicator both below and above 100% is undesirable, since this indicates an inefficient use of amino acids by the body for the synthesis of its own proteins.

The first limiting amino acid is methionine. The difference in amino acid score was less than valine by 40.7%, isoleucine by 67.8%, leucine by 64.8%, lysine and threonine by 68.3% and 40.7%, respectively. The amino acid score of the second limiting amino acid phenylalanine is 29.4% less than valine, 56.5% less than isoleucine, 53.5% less than leucine, 56.9% less than lysine, and 29.4% less than threonine.

Analogues of animal milk – rice and coconut, almond, and oatmeal drinks have a low amino acid score compared to pasteurised milk, which is associated with a lower mass fraction of amino acids in these products. Lysine and methionine in the rice and coconut drinks have the lowest amino acid score, which is 8.4% and 4.0% less compared to the almond analogue, respectively, and 3.5 and 2.8 times less compared to oatmeal, respectively.

Oatmeal drink is characterised by the highest amino acid score in leucine and surpasses the analogues of rice and coconut and almond by 4.66 times, by 51.4%, respectively. The leucine index in oatmeal is 10.9% higher than the amino acid score of valine, 9.0% higher than isoleucine, 2.2 times higher than lysine, 2.7 times higher than methionine, and higher than threonine and phenylalanine by 21.5% and 14.9%, respectively.

Rice and coconut drink has the highest amino acid score of threonine, exceeding valine by 2.5%, isoleucine by 4.5%, leucine by 2.6%, lysine by 8.9%, methionine by 8.7%, phenylalanine by 4.5%. In the almond drink, the highest amino acid score was observed for leucine, which exceeds the index of valine, isoleucine, lysine, methionine, threonine, and phenylalanine by 10.0; 8.0%, 2.75 and 3.57 times and by 10.0; 5.2%, respectively. The utility coefficient was calculated to assess the balance of the amino acid composition (Table 3).

***Table 3**. Utility coefficient*

Product name	**Utility coefficient, %**
Rice and coconut drink	9.8
Almond drink	13.9
Oatmeal drink	27.9
Pasteurised milk	74.7

***Source:** compiled by the authors*

The higher the value of the utility coefficient, the better the balance of the amino acid composition, and the more efficiently amino acids can be used to synthesise the body's own proteins. The highest utilitarianism coefficient was observed in the studied pasteurised milk. This figure is 74.7%. Drinks based on plant raw materials have a lower utility coefficient relative to cow's milk. Thus, the lowest coefficient of rice and coconut drink is 9.8%, the indicator is less than that of almond and oatmeal by 4.1 and 2.85 times, which indicates a low level of protein digestibility of this product. In an almond drink, the utility coefficient is lower compared to pasteurised milk and oatmeal drink by 5.4 times and 2.0 times, respectively. Oatmeal drink is characterised by the highest utility coefficient among plant-based beverages compared to rice and coconut and almond analogues of cow's milk by 2.85 and 2.0 times, respectively.

Analysing the results obtained, it can be concluded that the studied plant-based drinks oatmeal, almond and rice and coconut in the protein composition have a lower mass fraction of the amino acids valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine compared to cow's milk. The amino acids limiting for all the studied milk analogues are methionine and lysine, the smallest mass fraction of which is found in rice and coconut drinks.

Plant-based oatmeal drink contains the largest mass fraction of the amino acids valine, isoleucine, leucine, lysine, methionine, threonine, phenylalanine compared to rice and coconut and almond drinks, however, the indicators are lower compared to cow's milk. The largest mass fraction of the amino acid leucine, this indicator is higher compared to almond and rice and coconut drinks by 54.4% and 4.7 times, respectively, but its amount is less than in pasteurised milk by 84.5 %. In addition, oatmeal drink is characterised by the highest utility coefficient in comparison with rice and coconut, and almond plant-based analogues of milk.

Singhal *et al.* (2017) and Atalar (2019) found that a rice-based drink is rich in vitamins A, B12, and D and is high in dietary fibre, contains more selenium and manganese compared to other types of beverages. But compared to cow's milk, almond, rice and oatmeal drinks have a lower mass fraction of protein, amino acids, calcium and argentum. Comparing the amino acid composition of plant-based beverages with the amino acid composition of pasteurised milk and reference protein, there is a low mass fraction of amino acids: valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine.

Munekata *et al.* (2020), Han *et al.* (2021) and Paul *et al.* (2020) suggest that a key role is played by essential amino acids, which are not synthesised by humans and animals, and are found in very small amounts in plant products (limited or deficient). The researchers have proven that lysine and methionine are the limiting amino acids for most milk analogues. The study results indicate that the first limiting for all the studied analogues of milk is the amino acid methionine, the smallest mass fraction of which in the rice and coconut drink – 3.5 mg in 1 g of protein of the drink, the second limiting amino acid is lysine, the smallest mass fraction of which in the rice and coconut drink – 5.4 mg in 1 g of protein.

Jeske *et al.* (2017) and Fawzi *et al.* (2022), comparing the obtained data, prove that the protein of a drink made from soy is similar in composition to cow's milk protein, and the protein of an oatmeal drink is the most complete in amino acid composition. Analysing the data of this study, it is found that the plant-based oatmeal drink also contains the largest mass fraction of the amino acids valine, isoleucine, leucine, lysine, methionine, threonine, phenylalanine compared to rice and coconut and almond drinks, however, the indicators are lower compared to cow's milk.

Pua *et al.*, (2022), Raikos *et al.* (2020), Vagadia *et al.* (2018) prove that an important indicator of raw materials used in the production of plant drinks and analogues of dairy products, which characterises the nutritional and biological value is not only the chemical composition and ratio of nutrients, but also the composition and balance of amino acids. There is a certain relationship – the higher the value of the utility coefficient, the better the balance of the amino acid composition. According to the results of the conducted studies, it was noted that oatmeal drink has the highest indicator of the utility coefficient relative to the indicator of rice and coconut and almond analogues of milk. The value was 2.85 and 2.0 times higher, respectively.

Analysis of the obtained data shows that oatmeal drink is more complete in terms of amino acid composition of protein compared to rice and coconut and almond drinks, because it differs from analogues by a larger mass fraction of amino acids valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine, has the highest indicators of amino acid score and utility coefficient. Comparing the amino acid composition of the studied milk analogues with the amino acid composition of cow's milk, there is a low mass fraction of the amino acids valine, isoleucine, leucine, lysine, methionine, threonine, and phenylalanine. Limiting amino acids are methionine and lysine, the smallest mass fraction of which is found in rice and coconut drink.

CONCLUSIONS

It was researched that the development of insurance of the subjects of the closed cycle technology sphere would depend directly on the participation of the state and Insurers in this process. It was noted that for development of this type of insurance, efforts of all participants of the insurance market should be attracted. On the part of the state there should be the most incentive to insurance for enterprises working in the sphere of closed cycle technologies. Such an incentive may look like compensation of the part of the insurance cost for these enterprises. Therefore, enterprises will be able to

ensure their risks and have a better chance of successful existence and development, and the state will get another developed branch in the economy, which can become a significant part of the state's gross domestic product. Also, it was confirmed that under the condition of the consumer's awareness of insurance services under the conditions of the Contract of insurance the trust to Insurers would increase considerably.

It was established that the insurers themselves should develop new insurance products, which might be interested in terms of the subjects of the closed cycle technology sphere and significantly increase competition in the market of this type of insurance. However, to motivate Insurers first, it is necessary to increase the demand due to the compensation of part of the insurance cost. It was determined that in the long-term period of insurance of the subjects of the closed cycle technology sphere has all chances for successful development taking into account the development of the entire insurance market of Ukraine. All this will significantly improve the development of the closed cycle technologies and insurance in Ukraine. Thus, it is possible to confirm that under the condition of global insurance of subjects of the closed cycle technologies sphere and further development of such technologies, in the future the nature of risks of individual and legal persons, and accordingly the insurance sphere as a whole, will change considerably.

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Порівняльний аналіз амінокислотного складу напоїв на основі рослинної сировини та коров'ячого молока

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Анотація. Знання особливостей технологічних процесів виробництва аналогів молока коров'ячого, оптимальних параметрів за виробництва, хімічного складу сировини та одержаного готового продукту, зокрема амінокислотного складу, все це відіграє важливу роль під час розроблення нових видів продуктів функціонального призначення, а також для покращення їх стабільності, смакових та поживних властивостей, та дозволяє збагачувати їх додатковими компонентами шляхом різних модифікацій. Мета роботи – порівняльний аналіз амінокислотного складу та збалансованості амінокислот різних видів напоїв на основі рослинної сировини: мигдального, рисово-кокосового, вівсяного відносно амінокислотного складу білка коров'ячого молока. Були використані такі методи: капілярного електрофорезу – для визначення амінокислотного складу білка аналогів молока та коров'ячого, математичні – для розрахунку амінокислотного скору та коефіцієнту утилітарності. Наведено склад, органолептичні показники якості, основні технологічні етапи та безпечність виробництва аналогів молока тваринного походження. Для дослідження було використано три види напоїв на рослинній основі: мигдальне з масовою часткою жиру 1,5 %, вівсяне та рисово-кокосове з масовою часткою жиру 2,5 та 1,8 %, а також молоко коров'яче з масовою часткою жиру 2,5 %. Представлено порівняльну оцінку масової частки амінокислот валіну, ізолейцину, лейцину, лізину, метіоніну, треоніну, фенілаланіну у складі білка напоїв на основі рослинної сировини відносно масової частки амінокислот у складі білка молока, а також еталонного білка. Лімітуючою амінокислотою у складі білка досліджуваних аналогів молока коров'ячого є амінокислота метіонін. Найвищі показники амінокислотного скору білка напою вівсяного, проте, вони не перевищували показники молока пастеризованого. З метою проведення оцінки збалансованості амінокислотного складу визначали коефіцієнт утилітарності. Результати досліджень мають практичне значення, адже можуть застосовуватися за виробництва продуктів функціонального призначення, зокрема аналогів молока коров'ячого з метою покращення складу, підвищення біологічної цінності, збагачення поживними речовинами та додатковими компонентами

Ключові слова: аналоги молока коров'ячого, напій на рослинній основі, вівсяний напій, мигдальний напій, рисово-кокосовий напій, амінокислотний скор, коефіцієнт утилітарності