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THE ROLE OF MICROBIOTA AND THE TECHNOLOGICAL BASIS OF MICROBIOLOGICAL SAFETY OF FOOD PRODUCTS AND ANIMAL FEEDS

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Abstract. The microbiota of food products and animal feed not only ensures the safety of these products, but also significantly impacts the health and longevity of people who consume them. In animals, it influences their health, productivity, the quality of livestock products, and the profitability of the agribusiness. Therefore, during the production, transportation, and storage of feed ingredients and animal feeds, it is critically important to control quality indicators, especially microbiological safety, and to adhere to hygienic conditions at all stages of production, transportation, storage, and subsequent use of feeds. A key role in ensuring the satisfactory health of humans and animals is played by the microbiota, which populates specific areas of the digestive tract and forms the microbiome. Recent studies show that the mass of the microbiome constitutes 0.3% of the living organism's body mass. Despite its small mass, the microbiome plays a leading role in the vital functions of living organisms. A healthy microbiota positively contributes to digestion, nutrient assimilation, immunity support, and protection from pathogens. To ensure its development and maintenance, it is essential to enrich the diet with balanced prebiotic products, which helps to maintain a diverse and stable microbiome as a foundation for robust health and the prevention of many diseases. Therefore, the further improvement of food product quality and safety, and the efficiency of animal, poultry, and fish farming, lies in implementing hygiene measures in the production of animal feeds and establishing a healthy microbiota in animals. This is because this link in the "farm-to-fork" food production chain currently has the fewest measures to prevent microbiological contamination of feed raw materials and animal feeds. During the research, 151 samples of animal feed were analyzed. The samples were collected from feed mills in various geographical and climatic zones of Ukraine. The results of the bacteriological culture on appropriate nutrient media and microscopy showed that the microbial contamination of the samples ranged from 1.5×103 to 1×105 CFU/g. No Salmonella was detected in the finished feed samples. However, the most microbiologically contaminated areas of each of the studied feed mills were identified. The receiving device for grain raw materials and oilseed meals was found to be the most contaminated, with levels ranging from 3.5×106 to 4.0×107 CFU/g. An elevated level of microbiological contamination was observed in samples from feed deposits in the equipment after thermal treatment. In samples taken from the floors of the production facilities, an increased level of contamination by microorganisms was detected (8×105 CFU/g) compared to the finished feed, where it did not exceed 1.2–1.8×104 CFU/g. This indicates the necessity of implementing a system of microbiological hygiene measures at the feed mills.

Keywords: food products, animal feeds, microbiome, microbiological contamination, hygiene, production safety

Introduction. Formulation of the problem

Microorganisms accompany the lives of humans and animals at all stages of their development, influencing the state of health, longevity, and quality of life for people, as well as the health and productivity of animals, poultry, and aquatic organisms. They also affect the quality of raw materials, food, and feed products.

The implementation of modern principles for controlling the quality and safety of food and feed production relies on a systematic approach. This approach involves creating conditions that prevent pathogenic and opportunistic microbiota from entering food and feed, animal and poultry housing, any stage of food and feed production processes, feeding animals, poultry, and aquatic organisms, and also the storage, transportation, and processing of the resulting raw materials into food and feed, including the conditions for their subsequent transportation, storage, and use.

Applying HACCP principles allows not only for reacting to problems but also for actively preventing their occurrence, which is a key element in ensuring the microbiological safety and quality of food products. This approach is a major trend in the modern food industry. Even the most common opportunistic microorganisms found in plant-based raw materials for food and feed production, such as fungi of the Aspergillus and Penicillium genera, can cause diseases under certain conditions, primarily when the immune system of humans and animals is weakened. Their metabolites, known as mycotoxins, pose a direct threat to the health and lives of animals, poultry, aquatic organisms, and humans, as these toxic metabolic products can transfer into raw materials used for food production. Therefore, the microbiological safety of feed and food products is one of the main trends in ensuring their quality and safety for consumers. [1, 2].

The microbiota of food and feed not only determines the safety of these products for animals and subsequently for humans, but also significantly impacts animal productivity, the quality of livestock products, and the profitability of agribusiness. Therefore, during the production, transportation, and storage of feed raw materials and finished feeds, it is critically important to control quality indicators, with microbiological safety being paramount. Adherence to hygienic conditions is essential at all stages of production, transportation, storage, and subsequent use of feed.

The increasing importance of microbiological safety in ensuring the quality and safety of feed led to the introduction of the GMP+ Feed Certification scheme by the independent, non-governmental organization GMP+ International. This organization was founded in 1992 by members of the Dutch feed industry and was initially used on a national scale within the country.

Today, the GMP+ Feed Certification scheme is a recognized international system. While the implementation of GMP+ standards is voluntary, an increasing number of countries are making these certificates mandatory for the import of feed raw materials or finished feeds, thereby restricting access for foreign operators to their national feed markets. Consequently, Ukrainian producers of feed raw materials and compound feeds are increasingly adopting the GMP+ certification scheme. This scheme is designed to ensure the safety and quality of animal feed at all stages of the feed supply chain, from raw materials to finished feed. Furthermore, the impact of microbiological contamination levels – even from non-pathogenic microorganisms – on animal productivity, product quality, and business profitability is driving the study and implementation of technological foundations and a complex of hygiene measures at all stages of the production and use of feed and compound feeds.

Analysis of recent research and publications

The modern development of food production and food security, animal husbandry, poultry farming, fish farming, other aquaculture, and animal feed production is based on the principle of sustainable development. In 1987, G.H. Brundtland presented this as a type of development that can meet the needs of humanity without compromising the ability of future generations to meet their own needs [3]. According to Vaarst et al., this principle encompasses environmental responsibility, economic viability, and social responsibility [4].

The health status of animals, poultry, fish, and other industrially farmed aquatic organisms is a key factor in the development of animal husbandry, poultry farming, and aquaculture, as well as in the improvement of animal feed products and the high quality of food products [5, 6, 7]. A key role in ensuring a satisfactory state of health is played by the microbiota that inhabits specific parts of the digestive tract and forms the microbiome, which is now considered by many scientists to be another organ of living organisms [8, 9, 10].

The term microbiome was first defined in 1988 in the article "Mycoparasitism and plant disease control" by J.M. Whipps, Karen Lewis, and R.C. Cooke, which was published in the book Fungi in Biological Control Systems, edited by M.N. Burge. These authors defined the microbiome as a characteristic microbial community occupying a specific habitat with its own distinct physicochemical properties [11, 12]. While previous estimates suggested that the mass of the microbiome could be between 1 and 3% of the total body mass, more recent studies conducted by Ron Sender et al. indicate that its mass is approximately 0.3% of the body mass of a living organism [13]. It has been established that an imbalance in the gut microbiota (dysbiosis) is associated with the development of a wide range of diseases, including inflammatory bowel disease, metabolic syndrome, allergies, and even affects the "gut-brain axis" [14]. Thus, strategies aimed at the positive modulation of the microbiome through diet quality are becoming critically important for the prevention and treatment of these conditions.

Despite its small mass, the microbiome plays a leading role in the vital functions of animals. First, the state of the microbiome affects the health and reproductive functions of mammals and hydrobionts. The microbiota interacts with its host through key mechanisms such as forming a protective barrier against pathogens, maintaining the integrity of the mucosal barrier, facilitating nutrient metabolism, and modulating the immune response [15, 16]. Second, the microbiome significantly influences brain development and behavior. This is supported by comprehensive results from controlled laboratory experiments and clinical studies on animals and humans, which have shown that the gut microbiome affects neurodevelopmental processes, modulates stress responses, and influences cognitive functions via the gut-brain axis [17]. Third, the state of the microbiome influences animal productivity and the quality of livestock products. This is because specific microbial communities and their interaction with the host affect the efficiency of nutrient and bioactive substance digestion, animal growth rates, and even the formation of taste characteristics in the final products [18].

Considering the significant role of the microbiome in shaping the productivity and quality of products from livestock, poultry, fish, and other industrially farmed hydrobionts, in 2005, Professor S. Bilgili from the USA proposed evaluating the efficiency of broiler farming using an efficiency index. This index considers flock livability, average broiler body weight at the end of the growth period, duration of the growth period, and feed conversion ratio [19]. Subsequently, in 2009, this approach was adopted in EU countries as the European Production Efficiency Factor (EPEF) to assess the profitability of broiler production, as reflected in the Broiler Management Manual Ross-308 (UK) [20].

In studies conducted by E. Mavromati et al. in 2018 across eight poultry farms in EU countries, it was shown that the EPEF index varied from 218 to 292. This variation was attributed to the normalization of the microbiome and improved health status of the birds, resulting in better feed conversion and greater body mass over the same 39-day broiler growth period. As a result, one of the poultry farms with the maximum EPEF index not only achieved savings on compound feed costs but also produced an additional 29.9 tons of live broiler weight [21].

Thus, the state of the microbiome and the efficiency of its function significantly influence the profitability of the agricultural animal and poultry farming business. Therefore, it is critically important to pay attention to the factors that influence its formation, preservation, and function. In studies conducted by Sarah Haberecht et al., it was shown that the microbial colonization of the gastrointestinal tract of newly hatched chicks, as well as other birds and newborn mammals, begins at the moment of hatching (birth) and is shaped by environmental factors and feed microbiota [22]. The formation and state of the animal's microbiome are also influenced by the mode of birth, breed, age, stress, and the use of antibiotics, but the microbiota of the compound feed plays a key role [23]. Additionally, studies by Callie Selby showed that even in the absence of pathogenic and conditionally pathogenic microbiota in the feed, its total microbial contamination level can significantly affect the state of the microbiome and animal productivity [24]

Problem Statement. Thus, the problem of further improving the quality and safety of food products and the efficiency of animal husbandry, poultry farming, and fish farming lies in implementing hygienic measures in compound feed production. This includes shaping the compound feed microbiota to be favorable for the microbiome of animals, poultry, fish, and other hydrobionts, as well as using feed components with the best prebiotic index. Furthermore, it involves applying technological methods that enhance the prebiotic properties of individual components and compound feeds and improve the safety of products throughout the food chain.

Research materials and methods

For comparative analysis, the research materials included data from other authors, as well as samples of feed raw materials, dust samples from the production facilities of feed mills, and finished compound feeds.

To determine the level of microbiological contamination of feed raw materials, dust, and compound feeds, classical research methods were used, such as microbiological plating on nutrient media, microscopy, and the isolation and identification of microorganisms.

When sampling feed raw materials, dust, and finished products at feed mills, the requirements of the Law of Ukraine "On Feed Safety and Hygiene" from 2018 with amendments [25] were followed.

Results of the research and their discussion

The presence of the microorganism Salmonella in feed poses the greatest threat to the health of animals, poultry, and humans. This is because when it enters an animal's body through feed, it can be transferred to livestock products, which subsequently becomes a risk factor in food production. In studies conducted by F.T. Jones and K.E. Richardson, Salmonella was found in 100% of fishmeal samples and 10% of soybean meal samples collected from three feed mills in the USA. Notably, Salmonella was detected in both mash feed and feed that had been conventionally pelleted [26].

Contamination of compound feed with Salmonella is not uncommon, even in feeds that have undergone heat treatment. This is confirmed by data from EU member states, which show a national prevalence of Salmonella in poultry feed at 6% in one member state, while in most other countries, the prevalence is in the range of 0% to 1.5% [27]. Contamination rates have also been reported for feeds for poultry (up to 0.8%), pigs (up to 1.7%), and cattle (up to 2.4%) in EU countries [28]. According to the requirements of the GMP+ Feed Certification scheme, the level of microbiological contamination in finished compound feeds should not exceed 5×103 CFU/g [29]. A comparative analysis of data from a generalized study led by Anthony Christian et al. showed that the level of microbiological contamination in finished compound feeds varies significantly among different countries. According to data from Anthony Christian et al., the average levels of microbiological contamination (in CFU/g) in the analyzed samples of finished compound feeds, provided by feed mills from various countries worldwide, were: Argentina – $1…6*10^6$, Bangladesh –$2,4…3,3*10^{13}$, Brazil – $1,5…3,27*10^3$, Iraq – $5*10^6$, Iran – $6,3*10^3$, Nigeria – $2,19*10^5…3,6*10^7$, Poland – $1,1*10^3…1*10^6$, Saudi Arabia– $3,18*10^3$, Serbia – $6*10^6$, Slovakia – $1*10^3…2*10^7$, Turkey – $1*10^2…1*10^3$ [30]. A study conducted by C. Escobar Lobo found that the average level of microbiological contamination in compound feeds in the USA is no more than 1×103 CFU [31]. As is evident, among the countries whose feed mills consented to sample collection and analysis, the lowest levels of microbiological load were found in compound feeds produced in Brazil, Iran, Saudi Arabia, and Turkey, while the highest were in Bangladesh, Nigeria, Slovakia, Argentina, Iraq, Poland, and Serbia. This clearly indicates a close relationship between the level of technology and quality control systems in compound feed production and the level of their microbiological contamination.

From 2018 to 2025, we collected 152 samples of finished compound feed from 14 feed mills located in different parts of Ukraine. Results from bacterial plating on appropriate nutrient media and microscopy showed that the microbiological contamination of the samples was in the range of 1.5×103 to 1×105 CFU/g. Salmonella was not detected in 151 of the finished feed samples. It should be noted that none of the investigated feed mills used animal by-products (meat and bone meal, bone meal, and blood meal) or fishmeal as feed ingredients. The presence of Salmonella was found in one sample, which was most likely related to the feed mill's location on the territory of a poultry farm.

We collected samples of feed raw materials, compound feeds, and feed dust from various production areas and sections of three feed mills in Ukraine (their names are not disclosed in accordance with the inspection agreements) located in the Mykolaiv, Odesa, and Kharkiv regions. During the studies, certain patterns were identified.

At a feed mill with a capacity of 10 tons/hour located in the Mykolaiv region, samples of dust and spilled products were collected from the following locations: (1) a cyclone-type dust collector in the aspiration system of the grain raw material preparation line, (2) a scalper for cleaning grain raw materials, (3) the floor area near the extruder, (4) the surface of the hammer mill for grain grinding, (5) the surface of the compound feed component mixer, (6) the stagnant zones of the bucket elevator boot for conveying mash feed, (7) the surface of the grinder for oilseed meals, (8) the stagnant zones of the bucket elevator boot for conveying pelleted feed, (9) a distribution (swivel) pipe, (10) a fan that draws air from the pellet cooler.

No Salmonella was detected in any of the samples. The highest levels of microbial contamination were found in samples 2 (3.3×106 CFU/g), 5 (1.1×106 CFU/g), 6 (1.8×106 CFU/g), and 9 (3.5×106 CFU/g) (Fig. 1). These were dust samples collected from equipment used for the cleaning and preparation of grain raw materials. The lowest level of microbial contamination was observed in the sample collected from the spillage under the extruder. The CFU/g count for the finished compound feed was 5.0×103, which indicates the high effectiveness of using the extruder, particularly for addressing feed hygiene.

At a feed mill with a capacity of 10 tons/hour located in the Odesa region, samples of dust and spilled products of feed raw materials and compound feeds were collected from the following locations: the finished pellet sifter (1), the surface of the finished pellet sifter (2), the spout feeding mash feed into the die of the pellet press (3), the intake grate of the feed lysine hopper (4), the grate of the receiving pit for grain raw materials and oilseed meals (5), the aspiration system of the receiving pit (6), the drive-over surface of the receiving pit for grain raw materials and oilseed meals (7), a magnetic separator before grain raw material grinding (8), a pelleted feed grinder (9).

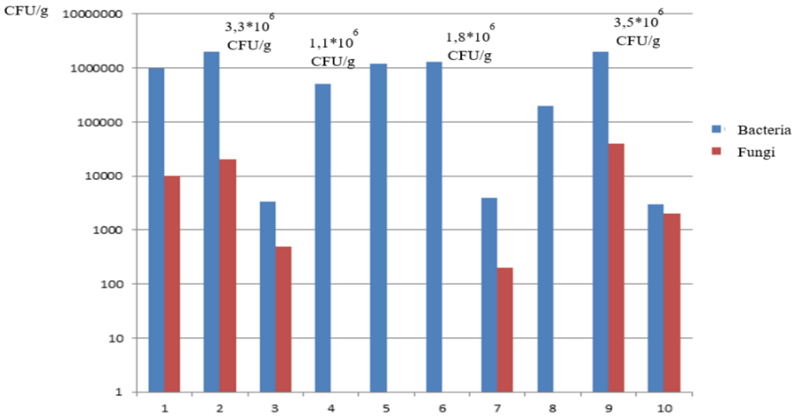

Fig. 1. The level of microbiota contamination of feed ingredient, dust, and compound feed samples collected at a feed mill in the Mykolaiv region .

During the microbiological analysis, no Salmonella was detected in any of the samples. The highest level of microbiological contamination was found in samples collected from the receiving pit, specifically from its drive-over surface, at the feed mill in the Odesa region, with a value of 4.0×107 CFU/g (Fig. 2).

The level of microbiological contamination in the finished compound feed did not exceed 1.3×104 CFU/g.

At a feed mill with a capacity of 10 tons/hour located in the Kharkiv region, samples of dust and spilled products of feed raw materials and compound feeds were collected from the following locations: the hopper above the pellet press (1), a hammer mill (2), the finished feed discharge device (3), the boot of the mash feed bucket elevator (4), the receiving pit for grain raw materials and oilseed meals (5), a pellet sifter (6), The batch grinding unit (7), the first-floor area (8), a compound feed component mixer (9). (Fig.3). No Salmonella was detected in any of the samples. The most microbiologically contaminated location was the receiving pit (7.8×106 CFU/g). An extremely high level of microbial contamination was also found in a sample of mash feed taken from the stagnant zone within the hopper above the pellet press (2.5×106 CFU/g). This is notable because the microbial contamination level in the mash feed before it entered the hopper was no more than 4.5×104 CFU/g. One possible reason for the microbial growth on the walls of the hopper above the pellet press could be the lack of thermal insulation for this hopper.

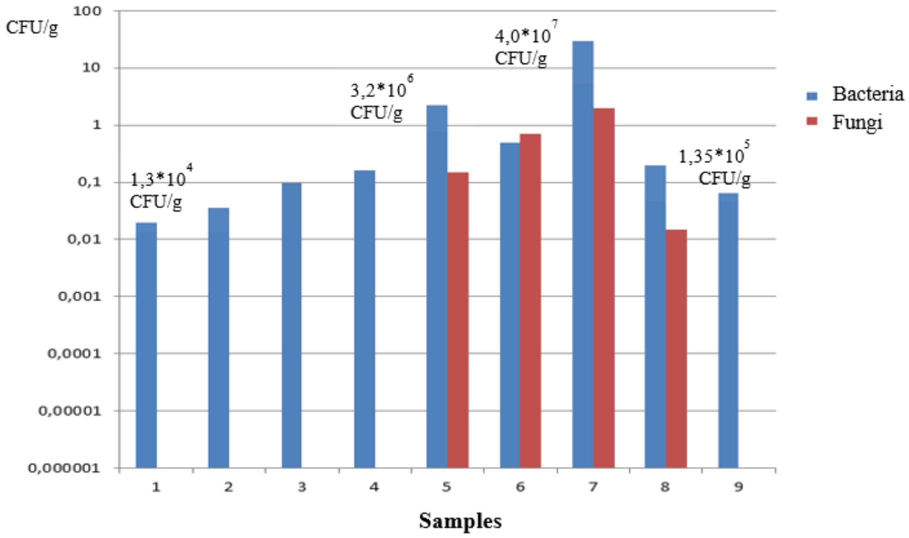

Fig. 2. Level of microbial contamination of feed raw materials, dust, and compound feed samples collected at a feed mill in the Odesa region.

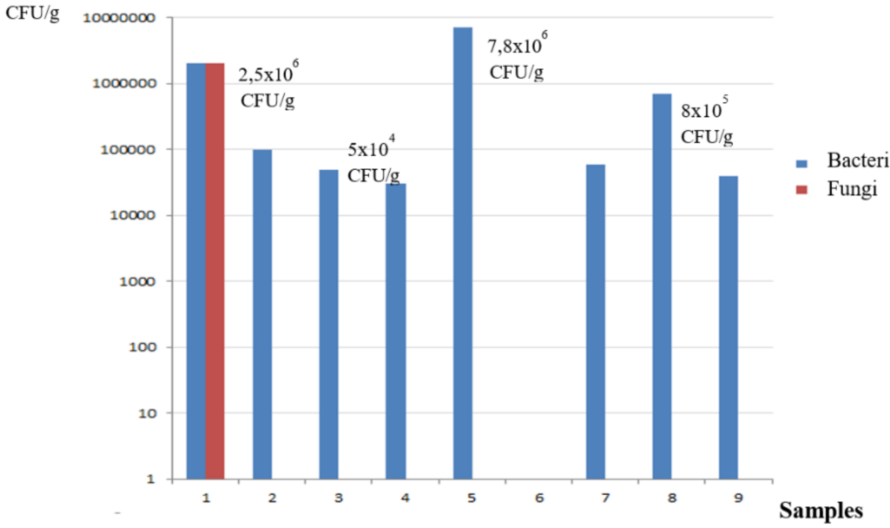

Fig. 3. The level of microbiota contamination of feed ingredient, dust, and compound feed samples collected at a feed mill in the Kharkiv region.

Conclusion

Based on the results obtained, it can be concluded that the most microbiologically contaminated area at the surveyed feed mills is the receiving pit for grain raw materials and oilseed meals. The contamination level here ranges from 3.5×106 to 4.0×107 CFU/g. This indicates the need for implementing measures such as regular surface cleaning, disinfecting vehicle wheels before entry, and optimizing aspiration systems to minimize dust emissions.

The use of an extruder is a highly effective method for feed hygiene, significantly reducing microbiological contamination in the final product, whereas stagnant zones in equipment, such as bucket elevator boots, contribute to the accumulation of microorganisms and increased contamination.

Attention should also be given to the design and sealing of technological and transport equipment, as well as the thermal insulation of operational hoppers. If processed feed is stored in these hoppers at a temperature that exceeds the workshop's air temperature by more than 5 °C, it can lead to the formation of condensation. When this condensation combines with dust deposits and feed accumulation in hard-to-reach areas, it promotes microbial growth.Among the key technological differences noted at the surveyed feed mills was the use of an extruder for processing finished feed at the plant located in the Mykolaiv region. The contamination level of the feed produced there did not exceed 5×103 CFU/g, a level characteristic of feed mills in the EU and the USA. In contrast, the processing of mash feed in the conditioners of pellet presses at the other two plants was insufficient to achieve a similar reduction in microbiological contamination.

An elevated level of microbial contamination was found in samples collected from the floors of the feed mill production facilities (8×105 CFU/g) compared to the finished compound feed, where it did not exceed 1.2×104 to 1.8×104 CFU/g. This highlights the need for implementing a system of personnel hygiene measures at feed mills. These should include defining specific traffic paths within workshops and between floors, and installing disinfection mats at entrances to production buildings and transitions between production areas. Such measures are crucial for preventing the cross-contamination of feed raw materials and finished feeds with dust and particles carrying a high microbiological load.

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РОЛЬ МІКРОБІОТИ ТА ТЕХНОЛОГІЧНІ ОСНОВИ МІКРОБІОЛОГІЧНОЇ БЕЗПЕКИ ХАРЧОВИХ ПРОДУКТІВ І КОМБІКОРМІВ

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Анотація. Мікробіота харчових продуктів та кормів для тварин не лише забезпечує безпеку цих продуктів, але й суттєво впливає на здоров'я та довголіття людей, які їх споживають. У тварин вона впливає на їхнє здоров'я, продуктивність, якість продукції тваринництва та прибутковість агробізнесу. Тому під час виробництва, транспортування та зберігання кормових інгредієнтів та кормів для тварин критично важливо контролювати показники якості, особливо мікробіологічну безпеку, та дотримуватися гігієнічних умов на всіх етапах виробництва, транспортування, зберігання та подальшого використання кормів. Ключову роль у забезпеченні задовільного здоров'я людей і тварин відіграє мікробіота, яка заселяє певні ділянки травного тракту та формує мікробіом. Сьогодні багато вчених розглядають мікробіом як ще один орган живих організмів. Недавні дослідження показують, що маса мікробіома становить 0,3% маси тіла живого організму. Незважаючи на свою невелику масу, мікробіом відіграє провідну роль у життєдіяльності живих організмів. Здорова мікробіота позитивно сприяє травленню, засвоєнню поживних речовин, підтримці імунітету та захисту від патогенів. Для забезпечення його розвитку та підтримки важливо збагачувати раціон збалансованими пребіотичними продуктами, що допомагає підтримувати різноманітний та стабільний мікробіом як основу для міцного здоров'я та профілактики багатьох захворювань. Тому подальше покращення якості та безпеки харчових продуктів, а також ефективності тваринництва, птахівництва та рибництва полягає у впровадженні гігієнічних заходів у виробництві кормів для тварин та формуванні здорової мікробіоти у тварин. Це пояснюється тим, що ця ланка у ланцюжку виробництва харчових продуктів «від ферми до виделки» наразі має найменше заходів для запобігання мікробіологічному забрудненню кормової сировини та кормів для тварин. Під час дослідження було проаналізовано 151 зразок кормів для тварин. Зразки були зібрані з комбікормових заводів у різних географічних та кліматичних зонах України. Результати бактеріологічного посіву на відповідні поживні середовища та мікроскопії показали, що мікробне забруднення зразків коливалося від $1{,}5\times10^3$ до 1×10^6 КУО/г. У зразках готових кормів сальмонели не виявлено. Однак було визначено найбільш мікробіологічно забруднені ділянки кожного з досліджуваних комбікормових заводів. Найбільш забрудненим виявився приймальний пристрій для зернової сировини та шроту олійних культур, рівні якого коливалися від $3{,}5\times10^6$ до $4{,}0\times10^{-}$ КУО/г. Підвищений рівень мікробіологічного забруднення спостерігався у зразках із запасів корму в обладнанні після термічної обробки. У зразках, взятих з підлог виробничих приміщень, виявлено підвищений рівень забруднення мікроорганізмами (8×10^{-6} КУО/г) порівняно з готовим кормом, де він не перевищував $1{,}2–1{,}8\times10^{-6}$ КУО/г. Це свідчить про необхідність впровадження системи заходів мікробіологічної гігієни на комбікормових заводах.

Ключові слова: харчові продукти, корми для тварин, мікробіом, мікробіологічне забруднення, гігієна, безпека виробництва.