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NEW METHODS OF MANURE DISPOSAL AND USE OF PROCESSING PRODUCTS IN PIG BREEDING

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The research was conducted in the laboratory of innovative technologies and experimental facilities of Institute of Pig Breeding and AIP NAAS and in the conditions of LIGA COJIAP LLC. In the course of the research, they evaluated and improved the "stepping technology" of vermiculture, developed a method of extracting young dungworms from compost, determined the possibility of vermiculture in large-sized packages of the "Big-Bag" type, and the expediency of using vermighumus and "Nanoverm" in the rations of young pigs. Vermihumus was obtained by utilizing manure in a continuous reactor to obtain vermiproduction using California worms (Eisenia Foetida). Raw vermighumus was extracted in the ABC-100 vortex bed apparatus and the biological preparation "Nanoverm" was obtained. It is determined that the technology has been developed vermiculture in containers compared to that in the fields in open ground increases the output of vermiculture by 14.78% and vermihumus by 23.36%.

The developed method of extracting young worms from compost, compared to the known one, ensures the separation of young individuals from sexually mature ones from compost. allows to separate young individuals from sexually mature ones. For the effective disposal of manure as a biological reactor for the production of compost and vermiproduction, a large-sized package "Big-Bag" was used, as well as a thermal case, which is four horizontally stacked reconstructed used truck ramps, covered with a polyurethane cover, in the middle of which a thermostat and a two-meter heating cable are installed. The developed device provided air temperature in the zone of vital activity of worms in the cold period of the year within the range of 13-18℃. Before feeding, raw vermighumus was mixed with compound feed and placed in the feeder, and "Nanoverm" - in a trough with water. For weaned piglets, the dose of vermighumus was 80-120 g per head per day at the age of 28-45 days, and Nanoverm - 2 ml, for 46-60 days 2.5 ml, 61-75 days - 3 ml. It was determined that the introduction of vermighumus and "Nanoverm" into the diet of weaned piglets had a positive effect on their growth energy and survival. In terms of live weight, the piglets outnumbered the control analogues at the age of 60 days by 13, 11 and 6.96%, and at the age of 90 days - by 14.2 and 11.52%, respectively. The cost of additional main products obtained from feeding vermihumus and "Nanoverm"

to piglets was UAH 343.3/head in the first experimental group, and UAH 278.53/head in the second.

Key words: disposal, vermicomposter, vermihumus, "Nanoverm", weaned piglets, living mass, preservation, interior.

НОВІ СПОСОБИ УТИЛІЗАЦІЇ ГНОЮ ТА ЗАСТОСУВАННЯ ПРОДУКТІВ ПЕРЕРОБКИ У СВИНАРСТВІ

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Дослідження проведенні в лабораторії інноваційних технологій та експериментальних об'єктів Інституту свинарства і АПВ НААН та в умовах ТОВ «ЛІГА СОЛІАР». В процесі дослідження проводили оцінку та удосконалення «крокуючої технології» вермикультивування, розробляли спосіб виїмки молодняку гнойових черв'яків із компосту, визначали можливість вермикультивування у великогабаритних упаковках типу «Big-Bag», та доцільність застосування вермигумусу і «Нановермі в раціонах молодняку свиней. Вермигумус отримували шляхом утилізації гною у реакторі неперервної дії для отримання вермипродукції з використанням каліфорнійських черв'яків (Eisenia Foetida). Вермигумус-сирець екстрагували в апараті вихрового шару АВС-100 і отримували біопрепарат «Нановерм». Встановлено, що розроблена технологія вермикультивування в контейнерах порівняно з такою у в буртах на відкритому грунті є підвищує вихід вермикультури на 14,78% і вермигумусу на 23,36%.

Розроблений спосіб виїмки молодняку черв'яків із компосту, порівняно із відомим, забезпечує відділення із компосту молодих особин від статевозрілих. дозволяє відділяти молодих особин від статевозрілих. Для ефективної утилізації гною в якості біологічного реактора для виробництва компосту та вермипродукції була застосована великогабаритна упаковка «Big-Bag, а також термо футляр, який являє собою чотири горизонтально складені реконструйовані вживані скати вантажних автомобілів, накритих кришкою з поліуретану, в середині яких установлено термостат і нагрівальнтй двометрового кабель. Розроблений пристрій забезпечував в в зоні життєдіяльності черв'яків температуру повітря в холодний період року в межах 13-18^{0}С. Вермигумус-сирець перед годівлею перемішували із комбікормом і вносили у годівницю, а «Нановерм» - у корито з водою. Для відлучених поросят доза вермигумусу склала на голову за добу у віці 28-45 днів 80-120 г , а «Нановерму -2 мл, на 46-60 днів 2,5 мл, 61-75 днів - 3 мл. Встановлено, що введення в раціон віднятих поросят вермигумусу і «Нановерму» позитивно вплинуло на їх енергію росту і збереженість. За живою масою відняті поросята переважали контрольних аналогів у віці 60 днів на 13, 11 і 6,96 %, а у 90-денному – відповідно на 14,2 і 11, 52 %. Вартість додаткової основної продукції, отриманої від згодовування вермигумусу і «Нановерму» поросятам склав у першій дослідній групі 343,3 грн/гол, а в другій – 278,53 грн/гол.

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Ключові слова: утилізація, вермикомпостер, вермигумус, «Нановерм», відняті поросята, жива маса, збереженість, інтер'єр.

Introduction. The modern practice of industrial complexes and farms highlighted one of the environmental problems associated with the utilization of animal waste products (Ivanov V.O., Voloshchuk V. M., 2019; Maksym V., D. Solomonko Lytvyn R., 2021). One of the progressive methods of manure disposal is vermicomposting. The development of this direction in agro-ecological production makes it possible to develop a system of diversification of vermitechnologies for obtaining organic products in the field of pig farming, as well as to solve a number of urgent environmental problems, namely: utilization of manure by vermicomposting, obtaining high-quality, environmentally friendly organic fertilizer, increasing soil fertility, strengthening the fodder base, obtaining safe organic pork (Senchuk M.M. Senchuk M.M., 2018; Senchuk M.M., 2020).

In the process of processing biohumus, earthworms (vermiculture) release a valuable product - caprolite, which was named vermihumus (Sherman R.. 2018). According to a number of scientists, an important characteristic of vermighumus as an environmentally friendly fertilizer is its macro- and microelement composition (from 14 to 18 and 25%) of nitrogen, phosphorus, calcium, magnesium, copper and zinc. The highest content of Mn (94–148 mg/kg) and Fe (74–195 mg/kg), less Zn, S, B, and the amount of Co and Cu did not exceed 1 mg/kg were noted in the vermihumus obtained after the processing of cattle manure. By means of vermicomposting, high-quality, environmentally friendly organic fertilizer (vermighumus) is obtained, which is used to increase soil fertility, strengthen the fodder base and grow safe organic pork (Maxim V., et. al., 2021; Przemieniecki, S.W. et. al., 2021).

One of the features of vermighumus is that it significantly reduces emissions of polluting gases (Naushin Y. et. al., 2022; Spiehs M.J. et. al., 2019). In this connection, vermiculture should be considered a new element of the technological process of production and processing of pig products. Bioprocessing of agro-industrial complex waste by the vermicomposting method helps to improve the ecological condition of the environment, as well as to obtain effective ecologically safe bioadditives (Dyudiaeva O.A., Rutta O.V., 2024; Maksym V. et. al., 2021). Along with this, there is more and more information in foreign and domestic literature that vermihumus is an effective source of protein replenishment in fodder production and animal feeding (Shatalin D.B., 2017; Hesami Y. et. al., 2020).

Analyzing the state of existing technologies in pig breeding, it should be noted that the recycling system and the diversification system are practically not developed for obtaining organic products of fodder and pork in conditions of agro-ecological production

In this regard, (Valyavska K.V. et. al., 2023; Zhuk P.V., 2022, Hussein I.A., Mona S.M.M., 2018) the further improvement of vermiculture technology through the development and implementation of new technical means for deep processing of vermihumus and the use of the resulting products in animal husbandry and fodder production, solving a number of important environmental problems, is urgent.

The purpose of the research is to develop new methods of manure disposal and the use of processing products in pig breeding.

Research materials and methods. Experimental research was conducted in the period 2020-2024. at "LIGA SOLAR" LLC, Zaporizhzhia, OJSC "Agroprime Holding". and in the laboratory of innovative technologies and experimental livestock facilities of the Institute of Pig Breeding and APV of the National Academy of Sciences, Research

took place in four stages. At the first stage, evaluation and improvement of the "stepping technology" of vermiculture was carried out.

At the second stage, a method of extracting young dungworms from compost was developed. At the third stage, the possibility of vermiculture in large-sized packages of the "Big-Bag" type was determined. At the fourth stage, the expediency of using vermihumus and "Nanoverm" in the diets of young pigs was determined. Vermiculture technology was carried out in accordance with methodological recommendations (Senchuk M.M. Senchuk M.M., 2018; Senchuk M.M., 2020, Sherman R.. 2018).

Vermiproduction was obtained by using vermiculture technology in continuous reactors (fig. 1).

Fig. 1. **3The content of humic substances in vermihumus was: humic acids-3.6% or 36.0 g/l, fulvic acids-3.0% or 30.0 g/l, humic substances-6.6% or 66.0 g/l.**

Biopreparation "Nanoverm" was obtained by extracting the obtained vermighumus in the ABC-100 vortex bed apparatus (fig. 2).

Fig. 2. **General view of the equipment for obtaining "Nanoworm"**

In "Nanoverm" the content of humic substances was: humic acids-0.56% or 5.6 g/l, fulvic acids-3.53% or 35.3 g/l, humic substances-4.09% or 40.9 g/l.

To determine the expediency of using vermighumus and "Nanoverm" in the rations of young pigs, three sub-experimental groups were formed (table 1).

Table 1

Scheme of feeding weaned piglets

Group	n	Feeding conditions of weaned piglets
Control	30	Standard combined feed according to existing norms
I-Experimental	30	Standard combined fodder + vermighumus (80-120 g per head per day)
II-Experimental	30	Standard compound feed + dietary supplement ("Nanoverm" 3-4 g per head per day)

The fattening productivity of young animals was determined according to the relevant methodical recommendations of Institute of Pig Breeding and AIP NAAS. The economic efficiency of research results was determined according to (Andriychuk V. G. (2002). The research results were processed using the methods of variational statistics (Chepur S.S., 2023).

Research results. "Stepping technology" allows vermicomposting to be carried out both outdoors and indoors (*Fig.* 3). It consists of several operations: preparation of the substrate, formation of the starting edge, settlement of the edge with a population of worms, control of adaptation, feeding, collection of worms, raw vermicompost, creation of conditions for humification and mineralization of vermicompost. separation of vermicompost and its storage.

Fig. 3. **Sides of the "stepping technology" of vermiculture in open ground and indoors**

Preparation of the substrate consists in preliminary natural composting of humus for 2-3 months outside in open ground. First, the basic substrate is prepared from the pre-fermented organic substrate, and a pillow of the future vermiburt is arranged from it on the concrete floor. A starting deck made of substrate with a width of 80-100 cm and a height of 20-30 cm is laid out on it. The length depends on the size of the room or composting area.

Then this substrate is inhabited by a population of compost worms of the Eisenia fetida species, at the rate of 2-2.5 kg of compost per 1m2. After 7-10 days, a 7-10 cm

layer of fodder substrate is layered on top of the burt and moistened. These operations are repeated regularly until the height of the side reaches 60-90 cm. From this moment, layering of the prepared fermented fodder substrate for compost worms and moistening is carried out only on one side of the formed stationary side. Also, fertilizing vermiculture with fodder substrate is started by layering only on one side.

Periodically moisten with water by sprinkling or spraying (humidity of the substrate should be 75-85%). The burt, which is in the room, is covered with agrofibre, and if it is outside - with straw. The layer of straw is 10-15 cm in summer, and 50 cm in winter. Periodically moisten with water by sprinkling or spraying (humidity of the substrate should be 75-85%). The burt, which is in the room, is covered with agrofibre, and if it is outside - with straw. The layer of straw is 10-15 cm in summer, and 50 cm in winter. In this method, compost worms continuously migrate for the nutrient substrate from one side of the burt to the other, forming an active zone of vermicomposting and vermiculture (Birnbaum J. A., 2015).

It should be known that after the formation of vermihumus, the next layer of top dressing must be applied, into which compost worms also continuously migrate, forming the next active zone of vermicomposting and vermicultivation, while a layer of finished vermihumus is removed from the opposite, respectively left or right, side of each side. The raw vermicompost collected from the sides is sent to the fermentation and humification area (closed soil; ventilated room without direct sunlight). It is stacked at a height of 1.5-2 m for further ripening and slow natural drying to a humidity of about 40% for 1-2 months. The disadvantage of this method is that compost worms are tasty food for rats and mice (Zaitseva V.G., Nesterenko O.V., Chernyshenko G.O., etc., 2020). As a result, the output of vermihumus and vermiculture decreases. The basis of the useful model is the task of improving the method by preventing contact between rodents and worms.

This task is achieved by forming ridges from separate grid containers filled with compost and vermiculture, and after completion of vermicomposting and vermiculture, new containers filled with nutritious substrate without worms are placed next to them (Fig. 4).

To implement the method, a container (fig. 1-5) was developed, which consists of a metal frame (1), in the whole bent pyramidal trapezoid, four walls (2), which are made of lattices, and the fifth - contains a hinged door (3) with a latch (4) and movable hinges (5). Fig. 1 shows the device in an axonometric projection in working condition, in fig. 2 - in a non-working condition, with the door open, in Fig. 3 - a cross section of a row of containers filled with compost and vermiculture, in figs. 4 and 5 - a diagram of filling a container with compost and vermiculture. Example 1. A substrate of organic composition made of pre-fermented organic material (cow, pig manure or other organic waste) is placed through the open door (2), which is fixed on the frame (1), into a container (fig. 1, 2), inserted into a support (fig. 4, 5), and from one of the walls (2) a population of compost worms is planted in the substrate.

Next, a self-propelled stacker forms ridges with a certain distance between them on an open site for vermicomposting. After the completion of vermicomposting and vermiculture and the formation of fresh vermihumus in the container, a second container filled with fresh nutrient substrate is tightly placed next to it, where the worms from the first container migrate. from the first container, forming the next active zone of vermicomposting and vermiculture (3).

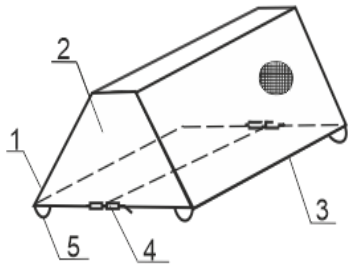


fig. 1

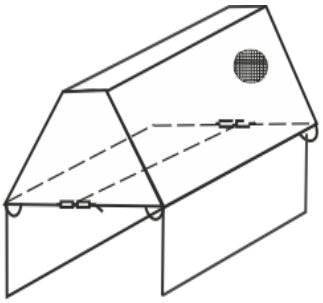


fig. 2

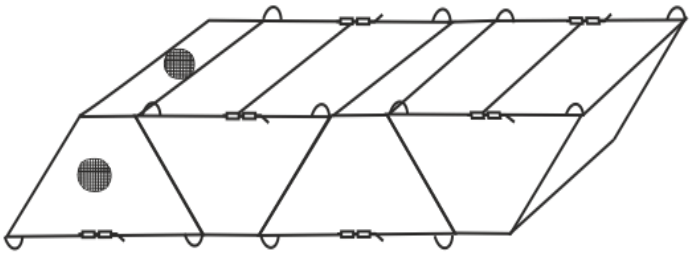
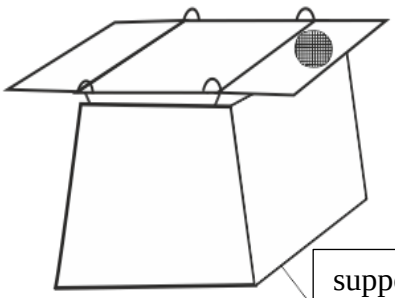
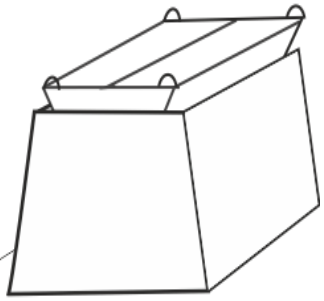


fig. 3





support for the container

fig. 4

fig. 5

Fig. 4. **General appearance of containers for ridge vermitechnology**

After the completion of vermicomposting and vermiculture and the formation of fresh vermihumus in the second container, a third container with a nutrient substrate is tightly placed to it again, where the worms from the second container migrate, etc. Containers with missing worms and ready biohumus are lifted by self-propelled stacker by hinges (5) and transported as intended. New containers with fresh substrate are placed in place of the evacuees and the vermicomposting process continues.

Given that the cells of the latticed metal wall (2) have a small size (up to 1 cm in diameter), the worms are completely protected from the penetration of rats and mice, which significantly increases the yield of vermiculture and vermihumus. Comparative characteristics of the efficiency of different methods of vermiculture and vermicomposting are given in table 2.

The data in table 2 indicate that the use of a new method of vermiculture and vermicomposting increases the output of vermiculture by 14.78% and vermihumus by 23.36%.

Table 2

Effectiveness of different methods of vermiculture and vermicomposting

Indicator	Prototype	Proposed method
The volume of manure in the container, m^3	1.0	1.0
Introduced vermiculture, kg	1.5	1.5
Received vermiculture, kg	11.5	13.2
Output of vermighumus, kg	50.5	62.3

A number of methods are used to quickly collect vermoproducts (Sherman R., 2018). For example, in the USA, a method of collecting dung worms from compost has been developed, which consists in adding bait - peat, rotten pieces of vegetables, coffee grounds, pieces of paper - to individual mesh bags used in households to store various vegetables, and then placing them in the top layer of compost, into which vermiculture was previously loaded for processing (Evans T., 2015). In the process of vermicomposting, worms from the compost crawl into mesh bags, where they consume the bait described above. After processing humus and bait, bags with worms at different stages of development (young, sexually mature) are removed from the processed compost and sent to their destination. The disadvantage of this method is that it does not allow separating young individuals from sexually mature ones. In scientific and industrial practice, such a technological operation is absolutely necessary.

Therefore, in order to improve the method, the bait is placed in bags made of mosquito net (the size of the cells is no more than 1.4 mm), and rotten fruits (pears, apples, apricots, plums) with a moisture content of no more than 70% are used as bait. In addition, the bag's stay in the compost should not exceed 5 days.

Example. Prepared compost with vermiculture (dung worms Eisenia fetida) is placed in bags made of mosquito net (the size of the cells is no more than 1.4 mm), filled with bait - rotten fruits (pears, apples, apricots, plums) with a moisture content of no more than 70%. Such humidity provides optimal conditions for the life activity of worms. Observations showed that worms consumed rotten fruits (pears, apples, apricots, plums) much better than rotten vegetables (table 3). Observations showed that young worms (up to 20 days old) freely penetrate through the cells of the mosquito net (diameter of the cell 4 mm), and at the age of 40-45 days they penetrate through the cells with tension, changing the thickness of the body due to its significant elongation. Periodically, but no later than 5 days, the bags were taken out of the compost and sent to their destination, and the formed vermicompost was sifted through a vibrating screen and thus received pure vermicompost and adults. Why do you need to remove the bags from the compost no later than 5 days? This is due to the fact that after 4-5 days, young worms become adults (puberty in worms also occurs at the age of 50-60 days). If the age at the time of introduction of worms into the bag is 45 days, and the period of stay is more than 5 days, the young individuals at the age of 45 days will become sexually mature (5+45 = 50 days), which will nullify the success of the method. This method of extracting young worms from compost can also be used for worms of other species under conditions of similarity of morpho-physiological parameters (age, body length, body thickness, sexual maturity).

The advantage of the proposed method is that it ensures the separation of young individuals from sexually mature ones from the compost. Therefore, recently, scientific institutions of various countries of the world have been intensively searching for methods and ways of removing, processing and using manure from large pig farms, which involve its complete utilization. The solution to this problem lies primarily in the fact that

livestock complexes become a source of raw materials for obtaining additional agricultural products (Bogatova D. R., 2018; Skorobogatov M.M., Kutserubova O. I., 2011).

Table 3

The attractiveness of the bait for worms according to the prototype and the new method

Indicator	Type of bait in a bag	
	moss, rotten vegetables, coffee grounds, pieces of paper	**rotten fruits (pears, apples, apricots, plums)**
The number of young worms in the bag 5 days after applying the worm bag, %	38	52
The number of young worms in the bag 10 days after applying the worm bag, %	64	88

The modern stage of manure disposal using vermiculture is a significant and effective biological method. In this connection, the further development and improvement of new technical means of vermiculture is relevant. As a biological reactor for the production of compost and vermiproduction, we used a large-sized package "Big-Bag" (0.75 x 0.75 x 1.25), which is widely used for packaging packaged and bulk building materials, ore, vegetables, fruits, mineral fertilizers, etc. (Fig. 5).

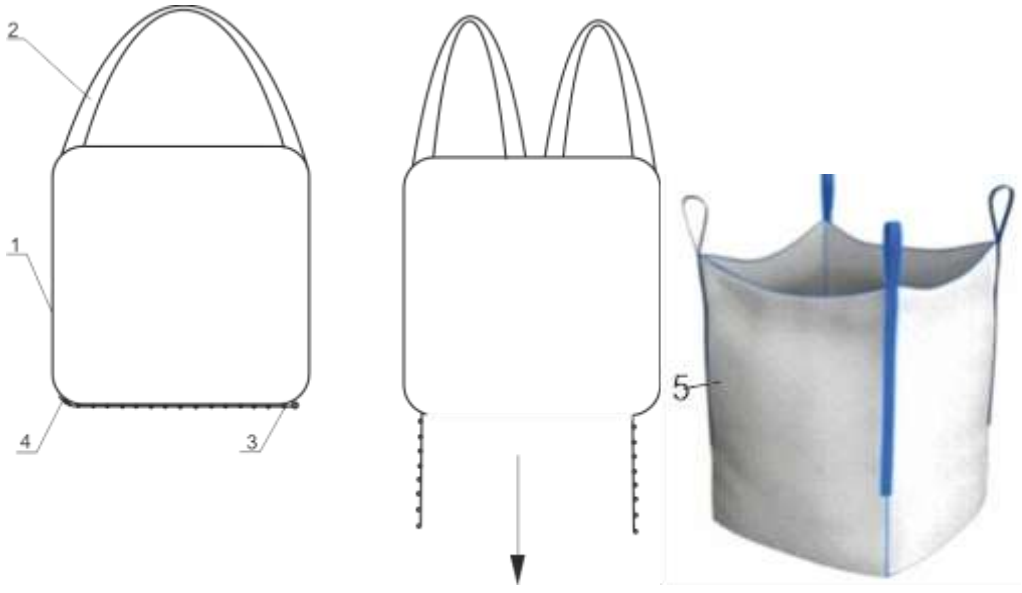


Fig. 5. **Vermicomposter with flexible walls: 1-case with polypropylene walls, 2- handles for gripping with a crane, 3- rings, 4-cord, 5- "Big-Bag" packaging.**

The task is implemented in the following way. With the help of a conveyor, fresh manure is loaded in layers through the neck of the package and watered with an aqueous solution of the biological preparation "Complesyn" on top. which is intended for quick processing of manure and obtaining vermihumus suitable for the settlement of

vermiculture. The drug was first diluted with warm water (20 g per 1 liter) and after 15 minutes evenly sprayed manure, which was added to the package at the rate of 10 liters per 1 m^3 of manure. According to the manufacturer's recommendations, the composting process should be carried out in the spring-autumn period, which takes place within 6-8 weeks, depending on the outside temperature. In our research, an additional task was set: to carry out composting in the cold period of the year (autumn-spring). A case was used to store each package, which is four horizontally stacked reconstructed used truck ramps, covered with a polyurethane cover (Fig. 6).

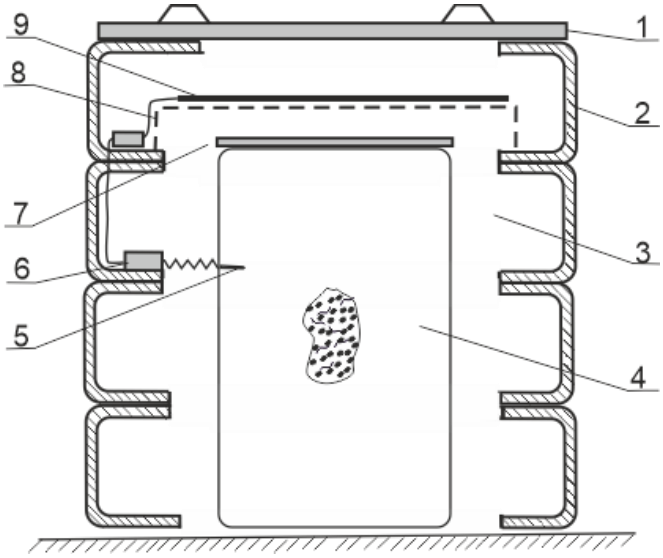


Fig. 6. **Case for packaging "Big-Bag with substrate": 1 - cover, 2 - slope, 3 - cavity, 4 - packaging, 5 - capillary tube, 6 - thermostat, 7 - polyurethane board, 8 - grid, 9 - heating element.**

An installation consisting of a thermostat and a two-meter heating cable (30 W/m) was used to heat the substrate. The thermostat contains a case with a handle, a canister with freon, connected to a spring and a capillary tube and designed to maintain a temperature in the range of -30...+ 30 ^{0}C. To prevent the manure from drying out in the "Big-Bag" packaging, a wooden board was placed on it, and a heating unit was placed on top. A capillary tube was inserted into the substrate and a temperature of 30 ^{0} C was set on the body using a handle. With this parameter of the thermostat and the average temperature of the outside air in the range of +0.4...-3 ^{0}C, the composting process lasted five weeks.

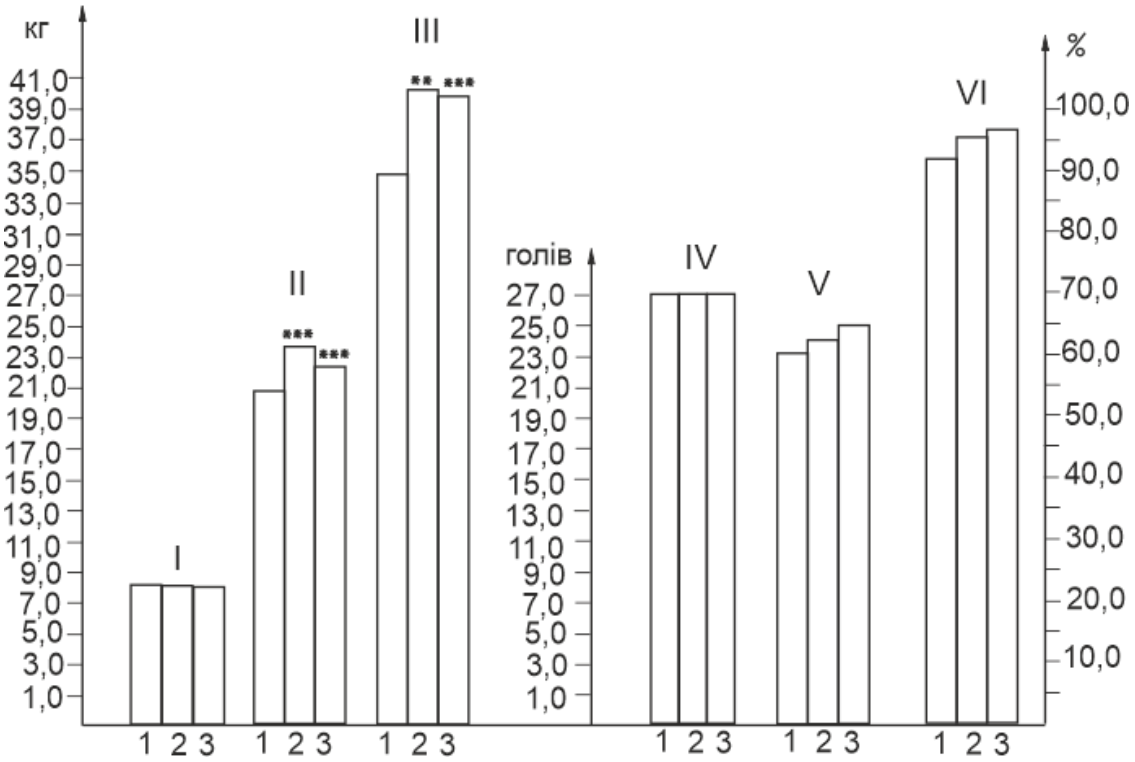
Next, a brood box with California worms (0.78 kg) was added to the prepared compost (70-80%) from above. The temperature in the living area of worms, depending on the temperature of the outside air, fluctuated between 13-18 ^{0}C.

The vermicomposting process lasted five months. At the end of vermicomposting, the mass of worms was 5.48 kg, and the mass of vermihumus was 0.5 m^3 .

To obtain liquid vermighumus or "vermichayu"several liters of specially purified structured water are poured onto the substrate, which flows through the entire substrate, is saturated with living microflora, soil microorganisms, spores, soil antibiotics, micro- and macroelements, humates, fulvic acids, amino acids, phytohormones, enzymes, vitamins, plant growth and development hormones and drains through the spill valve located at the bottom of the package . After the end of the compost processing process, the mother vermiculture is selected for further breeding.

To do this, a box with a new nutrient substrate is placed on the surface of the formed vermighumus, to which the worms willingly move. Next, the box with the mother culture is placed in a large package with prepared compost. Vermighumus, together with cocoons and other worms, is poured into vehicles, taken to fields to improve soil fertility, or used as a feed additive.

In order to establish the possibility of using feed additives from vermighumus and "Nanoverm" in the rations of young pigs, we conducted a production inspection. The results of the research are shown in Fig. 7.



*Note. **P>0.99, ***P>0.999 compared to the control group*

Fig. 7. **Indicators of productivity and survival of weaned piglets: I- number of piglets at the time of rearing (28 days), goal, II- live weight of piglets at the age of 28 days, kg, III- live weight of piglets at the age of 60 days, kg, IV- live weight of piglets at the age of 90 days, kg, V- Number of piglets at the age of 90 days, goal VI- Survival at the end of the 3-month age %.**

It was found that the weaned piglets of the first and second experimental groups exceeded their peers at the age of 60 days by 13.11 and 6.96% in live weight, and at the age of 90 days by 14.12 and 11.52%, respectively. The animals of the experimental groups also had a higher survival rate by 4.3 and 8.9%, respectively, which also confirms the biological activity of humates in growing young pigs. The cost of additional main products obtained from feeding vermighumus and Nanoverma" to piglets was 343.3 UAH/head in the first experimental group, and 278.53 UAH/head in the second. The results of hematological studies of young pigs showed that when adding vermigums in the amount of 50-80 g/head per day and "Nanoverm - 1-2 ml/ head per day to the main compound feed of suckling piglets of the first experimental group at the age of 90 days, the hemoglobin content increases by 6.27% and 8.75% compared to the control.

Table 4

Biochemical indicators of blood, g/l

Group	Age, days	Indicator		
		total protein	albumin	globulin
Control	28	42,21±2,43	25,25±3,92	14,68±3,24
	60	43,32±2,45	26,45±1,21	16,45±1,57
	90	54,02±2,24	28,01±1,96	22,41±2,02
1-experimental	28	47,21±2,23	28,25±2,02	16,68±1,24
	60	49,32±2,45	27,81±1,21	18,45±1,44
	90	59,71±2,74	34,21±1,36*	25,47±2,02*
2-experimental	28	47,21±2,13	29,25±1,91	18,68±1,34
	60	45,32±2,25	30,1±2,11	21,45±1,57
	90	62,14±2,14*	34,82±2,04*	28,47±1,12*

*Note. *P≤0.05; compared to the control group*

At the age of 90 days, the level of hemoglobin increases by 8.75% ($p<0.01$) compared to the indicators of the control group. Our data are consistent with the materials of foreign authors (Kovacik A, Sladecek T., Massán M. et.al., 2022). Feeding vermigmus and Nanoverm to piglets had a positive effect on protein metabolism and some indicators of animal resistance. (table 4).

Discussion. Improving technologies for the utilization of farm animal manure is a logical development of agricultural production, the final link of which is recycling and diversification (Skorobogatov M.M., Kutserubova O.I., 2011, Hussein I. A.S., Mona S.M. M. (2018). In our opinion, most of all, this process will be accelerated due to the introduction of innovative methods and techniques (Ivanov V.O., Voloshchuk V. M., 2019). Based on the goal, at the first stage of research, technological solutions for the production of vermihumus and worm products were evaluated. It should be noted that they were held in the nursery of LLC "Liga Solar" (Zaporizhia), which in its production activities is engaged in deep processing of manure and sale of the obtained products. When comparing the two most common technologies ("stepping edges" and "stationary edges") it was established that they have common opportunities for implementation both in open areas and in closed places without additional financial investments due to the rapid growth of the worm population. the technology of vermiculture in stepping edges compared to stationary ones in the calculation per 1 m^2 allows to increase the production of vermihumus by 1.68 times, vermiculture by 1.87 times. According to a number of authors (Zaytseva V.G., Nesterenko O.V.,. Chernyshenko G.O, etc., 2020), the considered technologies have a general drawback, which is due to the fact that worms attract rats and mice, which significantly reduce the yield of vermiculture. Therefore, in order to prevent contact with rodents, we have developed a method and a device for its implementation. Its essence is that the ridges are formed from separate lattice containers filled with compost and vermiculture, and after completion of vermicomposting and vermiculture, new containers filled with nutritious substrate without worms are placed next to them. In addition to the above-mentioned technologies, a new method was developed that allows you to lure worms from vermighumus, separating young individuals from sexually mature ones. In contrast to the known method (Evans T. 2015), we proposed a more perfect method because it ensures the separation of young individuals from sexually mature ones from the compost. This does not happen in the known method. Worms at various stages of development crawl into the mesh bags.

At the next stage, the large-sized packaging "Big-Bag" was studied as a biological reactor for the production of compost and vermiproduction in warm and cold seasons. The obtained vermighumus was used in feeding pigs. In the course of the study, it was established that the young of the II and III experimental groups exceeded their counterparts in terms of live weight by 4.02 and 8.76%, respectively. %. There are a number of reports in the scientific literature that confirm the positive effect of vermiguvmus on the results of pig breeding (Valyavska K.V., Geysun A.A., Matrosov O.S., 2023; Shatalin D.B., 2017).

An innovative method was used to obtain the biologically active substance - "Nanoverm" (trade name) from vermighumus - extraction in the AVS-100 vortex field apparatus. This method increases the content of humic substances in the finished product, compared to the prototype, 3.32-9.98 times higher. The introduction of "Nanoverm" into the diet of young pigs helped to increase their growth energy and the humoral defense of the body, which affected their survival. As you know, globulins provide humoral protection of the animal body. According to the content of globulin days, the piglets of the experimental groups slightly exceeded the control analogues, which can be explained by the influence of vermighumus and nanoverm, which were fed during the post-weaning period, which caused an increase in the synthesis of immunoglobulins. Our conclusions are consistent with research (Valyavska K.V., Geisun A.A., Matrosov O.S., 2023), which established that, due to the effects of sodium humate, succinic acid and trace elements, a probable increase in the proportion of T-lymphocytes was observed, which indicates an increase in the ability of the cellular link of immunity to respond to antigenic stimulation. The obtained data on the positive effect of humic substances are consistent with research conducted by domestic and foreign researchers (Byambas P., Hornick J.L., Marlier D. et.al., 2019; Rai S.N., 2019). The issues considered in the article give reason to consider them as an example of diversification in the agricultural sector, which is fully confirmed with the opinion of Bogatova D. R. (2018).

Conclusions

1. The technology of vermicomposting of "stepping edges" has been improved, which consists in the fact that ridges are formed from separate lattice containers filled with compost and vermiculture, and after completion of vermicomposting and vermiculture, new containers filled with a nutrient substrate without worms are placed next to them, as a result of which the output of vermiculture increases by 14.78% and vermihumus by 23.36%.
2. A method of extracting, separating from compost, young individuals from sexually mature worms from vermighumus has been developed, which consists in the fact that the bait is placed in bags made of anti-mosquito mesh with a mesh size of no more than 1.4 mm, and rotten fruits (pears, apples, apricots, plums) with a moisture content of no more than 70% are used as bait. In addition, the bag's stay in the compost should not exceed 5 days.
3. The technology of vermicomposting in large packages of the "Big-Bag" type in the cold period of the year has been developed. It was established that the obtained vermighumus when introduced into the concentrate diet of fattening young pigs in a dose of 5 and 10% contributes to an increase in live weight by 4.02 and 8.76%, respectively.
4. The introduction of vermighumus (80-120 g/head) and "Nanoverm" (2.5 ml per head) into the diet of weaned piglets of the first and second experimental groups contributed to an increase in live weight at the age of 60 days by 13.11 and 6.96%, and at the age of 90 days by 14.12 and 11.52%, respectively. The animals of the experimental groups also had a higher survival rate by 4.3 and 8.9%, respectively, which also confirms the biological activity of humates in growing young pigs.

5. The experimental research materials given in the article should be considered as an example of diversification in the agricultural sector.

References

Andriychuk V. G. (2002). Ekonomika silskohospodarskykh pidpryiemstv [Economics of agricultural enterprises] Textbook. 2nd ed., add. and recycled K.: KNEU, 624 p. (In Ukrainian).

Birnbaum J. A. (2015). Vermicomposting and vermiculture systems for cold climates. URL: https://www.canr.msu.edu/hrt/uploads/535/78622/Vermicompos-ting-Systems-19pgs.pdf.

Bogatova D. R. (2018). Diversification as a factor in the competitiveness of agricultural enterprises. URL: http://www.economy.nayka.com.ua/pdf/10_2018/159.pdf.

Byambas P., Hornick J.L., Marlier D. et.al. (2019). Vermiculture in animal farming: A review on the biological and nonbiological risks related to earthworms in animal feed. Vol. 5. Issue. 1 https://doi.org/10.1080/23311843.2019.1591328.

Chepur S.S. (2023). Biometriia: navchalnyi posibnyk [Biometrics: study guide]. Uzhhorod: "Hoverla" UzhNU Publishing House,. 196 p. (In Ukrainian).

Dyudyaeva O.A., Rutta O.V. (2024). Greening of the food industry through the introduction of vermiculture technology in agriculture. *Environmental sciences*. No. 2(53). WITH. 22-28.

Evans T. (2015). Harvest Worm Castings the Easy Way. https://myurbangardenoasis.wordpress.com/2015/03/23/harvest-worm-castings-the-easy-way.

Hesami Y., Esmaielzadeh L., Torshizi M. A. K. (2020). Effect of diets containing earthworm powder and vermihumus on egg production, hatchability, blood parameters and immunity of Japanese breeder quails. *J Anim Physiol a Anim Nutr*.. 105(4). https://doi.org/10.1111/jpn.13453.

Hussein I. A.S., Mona S.M. M. (2018). Solid waste issue: Sources, composition, disposal, recycling, and valorization. Egyptian Journal of Petroleum. Vol. 27, Issue 4, , P. 1275-1290.

Ivanov V.O., Voloshchuk V. M. (2019). New in the technology of production and processing of livestock products. Monograph. Poltava "Firma Techservice" LLC. 434 p.

Kovacik A, Sladecek T., Massán M. et.al. (2022). Impacts of humic acids in nutrition on haematological and biochemical parameters of brown hares. *Journal of Microbiology Biotechnology and Food Sciences*. DOI:10.55251/jmbfs.9549 .

Maksym V., Solomonko D. Lytvyn R. (2021). Economic efficiency of processing organic livestock waste into biohumus. Scientific Bulletin of the LNU of Veterinary Medicine and Biotechnology. Series: Economic Sciences. Vol 23 No 98.: https://doi.org/10.32718/nvlvet-e9805.

Naushin Y., Milleni J., Alok K. P.(2022). Emission of greenhouse gases (GHGs) during composting and vermicomposting: Measurement, mitigation, and perspectives Author links open overlay panel . Energy Nexus. Vol. 7, 100092/https://doi.org/10.1016/j.nexus.2022.100092.

Przemieniecki, S.W. Zapałowska A., Skwiercz A. .et. al. (2021). An evaluation of selected chemical, biochemical, and biological parameters of soil enriched with vermicompost. Environ Sci Pollut Res Int. vol. 28(7). C. 8117–8127. https://doi.org/10.1007/s11356-020-10981-z PMCID: PMC7854409.

Rai S.N. (2019). Haya Vermiculture and Vermicomposting: Agriculture in Fiji. Islands: Haya Saudi J Life Sci. Dubai, United Arab Emirate. https://doi.org/10.21276/haya.2019.4.2.6.

Senchuk M. M. (2020). Technological design in organic production: Educational and methodological guide for independent work and practical classes of students of the agrobiotechnological faculty / BSAU. Bila Tserkva, 94p.

Shatalin D.B. (2017.). Earthworms (Lumbrisidae) of forest and urboecosystems of the Dnieper steppe: structural and functional organization of groups and ecological aspects of vermiculture: diss. ... candidate s.-g. Sciences: 03.00.16. Dnipro 187 p.

Sherman R. (2018). Vermiculture: A Farmer's Guide. Chelsea Green Publishing Co. 256 p.

Skorobogatov M.M., Kutserubova O.I. (2011). Diversification as one of the ways to improve the efficiency of enterprises in modern conditions. Economic Herald of Donbass. No. 3. P. 18-21.

Spiehs M.J., Woodbury B.L. Parker D.B. (2019). Ammonia, Hydrogen Sulfide, and Greenhouse Gas Emissions from Lab-Scaled Manure Bedpacks with and without Aluminum Sulfate Additions. Environments, 6(10), 108; https://doi.org/10.3390/environments6100108.

Valyavska K.V., Geisun A.A., Matrosov O.S. (2023). The influence of biologically active substances on the reproductive function of vermiculture. Current issues of biotechnology, ecology and nature management: International scientific conference, April 27-28. P. 76-78.

Vasanthi P. J. (2019). Efficacy of Different Substrates on Vermicompost Production: *A Biochemical Submitted: Published:* . DOI:10.5772/intechopen.86187.

Zaitseva V.G., Nesterenko O.V., Chernyshenko G.O. et. al.. (2020). Vermiculture, its importance in solving environmental problems and improving agricultural conditions. *Scientific bulletin of construction*. Vol. 101, No. 3. P. 222-228. doi.org/10.29295/2311-7257-2018-101-3-222-228.

Zhuk P. V. (2022). Agricultural waste in Ukraine: volumes of generation and issues of recycling. https://doi.org/10.36818/2071-4653-2022-3-4

Zralý Z., Písaříková B. (2010). Effect of Sodium Humate on the Content of Trace Elements in Organs of Weaned Piglets. *Acta Vet.* Brno 79: 73-79. URL: https://doi.org/10.2754/avb201079010073.

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FACTORS OF FEED PRODUCTION IN THE CONTEXT OF CLIMATE CHANGE

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The article examines the current state and dynamics of the development of the feed production industry in Ukraine and considers the impact of climate change factors and force majeure on its development.

It is determined that climate change has been observed not only all over the world, but also in the conditions of Ukraine. They are accompanied by an increase in Heat Supply and aridity of the growing season, uneven distribution of precipitation throughout the year.

It is established that over the past 20 years, the structure of sown areas of agricultural crops has changed significantly. In 2000, cereals accounted for 50.2%, technical 15.4%, and fodder 26.0 %. However, over the years, prices for cereals and technical crops (except sugar beet) have significantly increased, and the number of animals has significantly decreased. Therefore, in the total structure of sown areas, the area of industrial crops increased in 2021 to 32.3%, including the share of sunflower increased from 8.4% to 23.1%. The percentage of forage crops decreased from 26.0% in 2000 to 9.6% in 2010 and to 5.4% in 2021. it is proved that the use of nine different crops at the enterprise makes it possible to organize 7-full crop rotations with the best predecessors.

The analysis of the development of the feed production industry in PE "Agro-Novoselovka 2009" Novovodolazhsky district, Kharkiv region over the past 3 years is carried out. It is established that in 2021 the percentage of grain and leguminous crops was 69.6 %, technical – 26.2 %, fodder – 4.2 %. In 2022, their ratio did not change significantly and amounted to 72.2, 23.0 and 4.7%, respectively. the share of Feed area increased from 4.2% in 2021, to 4.7% in 2022 and to 7.5% in 2023, which is positive for feed production and animal feed supply.

It is determined that the company uses 7-field crop rotations and effective agrotechnical techniques in crop production, so that good predecessors are selected for agricultural crops. Corn for both silage and grain proved to be the most effective fodder crop in three years, under various conditions. However, the highest yield of crude protein per 1 ha was found in perennial grasses (alfalfa) for hay.

Keywords: feed production, climate, acreage, crop rotations, forage land, yield, feed units, productivity of 1 ha of area.

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ФАКТОРИ КОРМОВИРОБНИЦТВА В УМОВАХ ЗМІН КЛІМАТУ

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В статті досліджено сучасний стан і динаміку розвитку галузі кормовиробництва в Україні та розглянуто питання щодо впливу факторів змін клімату та форс-мажорного характеру на її розвиток.

Визначено, що зміни клімату відмічено, не тільки у всьому світі, а й в умовах України. Вони супроводжуються зростанням теплозабезпеченості та посушливості вегетаційного періоду, нерівномірним розподілом атмосферних опадів протягом року.

Встановлено, що за останні 20 років структура посівних площ сільськогосподарських культур суттєво змінилася. У 2000 році зернові складали 50,2 %, технічні 15,4%, кормові 26,0 %. Проте за ці роки суттєво піднялися ціни на зернові та технічні (окрім цукрового буряка) а поголів'я тварин суттєво скоротилося. Тому в загальній структурі посівних площ, площа технічних культур збільшилась у 2021 році до 32,3 %, в тому числі питома вага соняшника збільшилась з 8,4 до 23,1 %. Відсоток кормових культур зменшився з 26,0 % у 2000 р. до 9,6 % у 2010 р. і до 5,4 % у 2021 р. доведено, що застосування на підприємстві дев'яти різних культур дає можливість організовувати 7-польні сівообороти з найкращими попередниками.

Проведено аналіз розвитку галузі кормовиробництва у ПП «Агро-Новоселівка 2009» Нововодолазького району, Харківської області за останні 3 роки. Встановлено, що у 2021 році відсоток зернових і зернобобових культур складав 69,6 %, технічних – 26,2 %, кормових – 4,2 %. У 2022 році їх співвідношення суттєво не змінилося і склало відповідно 72,2; 23,0 та 4,7 %. частка площі кормових площ збільшувалась з 4,2 % у 2021 р., до 4,7 % у 2022 р. і до 7,5 % у 2023 р., що є позитивним для кормовиробництва і забезпечення тварин кормами.

Визначено, що на підприємстві застосовують 7-польні сівозміни та ефективні агротехнічні прийоми в рослинництві, завдяки чому для сільськогосподарських культур підібрані добрі попередники. Найбільш ефективною кормовою культурою за три роки, при різних умовах, виявила себе кукурудза як на силос так і на зерно. Проте найбільший вихід сирого протеїну з 1 га встановлено у багаторічних трав (люцерна) на сіно.

Ключові слова: кормовиробництво, клімат, посівні площі, сівозміни, кормові угіддя, урожайність, кормові одиниці, продуктивність 1 га площі.

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Introduction. The basis for the development of the livestock industry is the feed production industry, which is formed from different types of feed depending on the livestock industry. In realizing the productive potential of farm animals, feed is crucial, since their share in the production of livestock products is about 60 %.

The development of feed production according to V. F. Petrichenko (2022) should be aimed, in particular, at increasing the specific share of high-yielding varieties of corn, perennial legumes and grasses, annual legumes adapted to various conditions, resistant to pathogens, environmental stresses, with increased symbiotic activity; increasing the yield of forage crops and rational use of hayfields and pastures.

An important prerequisite for the development of animal husbandry in agricultural formations of various forms of ownership is the creation of a strong feed base in each farm. The importance of the feed industry is due to the fact that it is the basis for the growth of livestock and increasing its productivity, and this in turn determines the growth rate and level of production of livestock products, since feed accounts for 68-73% in the cost of livestock production. However, in recent years, the deficit of feed protein is 25-30%, which requires a new approach and significant changes in the formation of the feed base (Ambrosov V. Ya., 2009).

Feed production is a multifaceted but imperfect industry in technical and technological terms, which requires both the use of innovative technologies, achievements of modern science and technology, and significant investments, attracting material and labor resources. The peculiarity of feed production is its belonging to all three spheres of the agro – industrial complex - (feed is the means of production (objects of labor) of livestock products); direct production (cultivation, harvesting and storage of feed); industries of processing agricultural products (production of mixed feed, feed from plant waste).

An important issue of agricultural production is the efficiency of feed production, because this industry is located at the "junction" of two branches of Agriculture: Animal Husbandry and crop production. Feed production is a multifaceted but imperfect industry in technical and technological terms, which requires both the use of innovative technologies, achievements of modern science and technology, and significant investments, attracting material and labor resources (Ambrosov V. Ya., 2009).

Important scientific developments on forage crop cultivation technologies were carried out by well-known scientists of Ukraine A. A. Babich, V. F. Petrichenko, M. I. Bakhmat, G. I. Demidas, V. G. Kurgak, A.V. Korneychuk, I. I. Senik., Yu. a. Veklenko and others. However, many issues of this issue are still insufficiently studied.

Climate change has been observed not only all over the world, but also in the conditions of Ukraine. As V. A. Balabukh Notes (2022), they are accompanied by an increase in Heat Supply and aridity of the growing season, uneven distribution of precipitation throughout the year, a shift in the long-term timing of meteorological calendar phenomena, and so on.

At the same time, according to Ambrosov V. Ya. (2009), the production of feed both in quantity and quality does not meet the needs of animal husbandry, which is a consequence of low efficiency of feed use, their overspending, high feed capacity of a unit of production. In this regard, the problem of innovative development of feed production and improving the efficiency of animal husbandry on this basis requires a detailed study, taking into account theoretical, methodological and practical aspects. Also, in the context of global warming and a decrease in the level of summer precipitation, there is an increasing need to study the intensification of forage crops and provide livestock with high-quality feed.

The purpose of this article was to establish the influence of agroclimatic features and technological factors on the intensity of forage crop cultivation in the conditions of eastern Ukraine.

Materials and methods. The object of the study is the process of functioning of the feed production industry, both in Ukraine as a whole and in a separate enterprise, in the context of global climate change.

The theoretical and methodological basis of the research is a set of methods, approaches, principles of scientific knowledge, fundamental provisions of agrobiology of forage crops, scientific developments on the formation of effective feed production and justification of directions for improving the efficiency of forage crops in the context of climate change.

The article uses the method of analysis and synthesis, abstract-logical (formation of theoretical generalizations and conclusions), economic-mathematical, statistical, comparison and generalization. The theoretical and informational base of the research consists of the works of domestic scientists, analytical reviews of experts, as well as statistical information on the dynamics of the formation of the feed production industry.

Additional surveys were carried out in the state of emergency "Agro-Novoselovka 2009" in 2021, 2022 and 2023 on the sown areas of fodder, grain and industrial crops. The effectiveness of fodder and grain crops under various agroclimatic conditions was studied.

Research results. According to the World Meteorological Organization (WMO, 2019), since the 80s of the last century, every decade has been warmer than the previous one and this trend will continue in the future. The annual global temperature in 2019 was 1.1°C higher than the average for 1850-1900).

Today's warming trend is of particular importance, as much of it is the result of human activity since the mid-twentieth century and has been occurring at a rate unprecedented for decades to millennia.

Global climate change over time has coincided with population growth over the past century, leading to a significant increase in food demand. The United Nations predicts that the world's population will reach 9.7 billion by 2050, 10.8 billion by 2080, and 11.2 billion by 2100. While these projections do suggest a slowdown in global population growth, Africa and South Asia are expected to grow significantly and steadily: by 2100, these two regions may well be home to a population of 9 billion out of the projected 11 billion people on the planet. Driven by these important demographic forces, demand for products is expected to increase significantly, especially in Africa and South Asia (FAO, 2018). As noted by William Chutney (2019), climate change can cause food shortages and, as a result, hunger in the world. According to UN FAO (2018) estimates in 2017, more than 820 million people, about one in nine of the population, are still malnourished.

Climate changes on a global scale have also affected the territory of Ukraine. Since the beginning of the 80s and still in Ukraine, there has been a rather rapid trend towards an increase in the average annual air temperature. So, according to the Ukrainian hydrometeorological center, if the change in annual temperature in the northern hemisphere of the Earth for 50 years has not yet reached 1 °C, then in Ukraine it has increased by 1.4 °C. Over the past 20 years, the average monthly temperature in winter has increased by 2–3 °C, which has led to a reduction in winter by almost a month. This warming extends from south to North (Adamenko T. I., 2023).

The average air temperature increased in the north-eastern and south-eastern sub–regions of Ukraine by 2.7-2.8 °C, in the north – western–by 1.1-1.7°C. In accordance with the increase in air temperature, the number of frosty days decreased by about 5-10%.

Humidity increased by 10-25%. Smaller changes are observed in relation to precipitation: for them, the trend values are outside the 90% significance level. In Crimea, the trends of temperature and humidity changes were the same as for the mainland regions, but their absolute value was less, which is probably due to the influence of the Black Sea. Every 10 years in the regions of Ukraine, on average, there is an increase in temperature by 0.3–0.4 °C, that is, over 30 years – by 1 °C. According to scientists of the Institute of Botany of the National Academy of Sciences of Ukraine, this can lead to a shift of natural zones by 160 km (Lyakhu D. S., 2024).

According to the Ukrainian hydrometeorological center (2019), warming will continue until 2030. the temperature will rise by an average of 0.2–0.3 °C. At this rate of warming in Ukraine in 2030, it will be possible to grow crops in the southern regions only if the irrigation system is restored, since droughts will constantly recur. By 2050, a 2 °C increase in temperature could lead to desertification in the southern regions. These changes significantly affect the development of feed production and the provision of feed to farm animals.

Due to a significant reduction in the acreage under forage crops in recent years, the dynamics of forage crop production in Ukraine is generally negative (Table 1).

Table 1

Area of agricultural crops in Ukraine, mln. Ga

Indicators	2000	2005	2010	2015	2021
Agricultural crops, total	27,1	26,0	26,9	26,9	28,6
Grain crops and legumes	13,6	15,0	15,1	14,7	16,0
including wheat	5,31	6,18	6,13	6,69	6,91
corn	1,36	1,71	2,71	4,12	5,52
Technical crops	4,18	5,26	7,29	8,35	9,24
including sugar beet, thousand hectares	856	652	501	237	227
sunflower	2,94	3,74	4,57	5,11	6,62
Fodder crops	7,06	3,73	2,60	1,99	1,53
including feed corn	1,92	0,77	0,47	0,31	0,21

Thus, in Ukraine there is a trend inherent in countries with highly developed agriculture, where the increase in crop production is achieved through more intensive use of land resources, which ensures an increase in crop yields.

Over the past 20 years, the structure of sown areas of agricultural crops has changed significantly. Their total area increased by 1.5 million square meters. ha or by 5.5 %. The area under grain crops increased by 2.4 million hectares. ha or 17.6 %, wheat by 1.6 million. ha (30.1 %), corn – from 1.36 to 5.52 million hectares. ha or 4.0 times. Over the years, the area of industrial crops has increased 2.2 times. However, the area sown with sugar beet decreased from 856 to 227 thousand hectares, or 3.7 times, and sunflower – increased by 3.7 million. ha or 2.3 times. Over the years, the number of farm animals has almost halved. Accordingly, the area of forage crops sown decreased from 7.0 to 1.53 million hectares. ha or 4.8 times, including the area of fodder corn decreased from 1.9 million hectares. ha up to 0.21 or 9 times.

Table 2

Structure of acreage in Ukraine, %

Indicators	2000	2005	2010	2015	2021
Rural culture-household	100,0	100,0	100,0	100,0	100,0
Grain crops and legumes	50,2	57,6	56,0	54,8	56,0
Including wheat	39,0	41,2	40,7	45,4	43,2
corn	10,0	11,4	18,0	28,0	34,5
Technical crops	15,4	20,2	27,1	31,0	32,3
Including sugar beet, thousand hectares	20,4	12,4	6,9	2,8	2,5
sunflower	70,3	71,2	62,7	61,1	71,6
Fodder crops	26,0	14,4	9,6	7,4	5,4
Feed corn	27,2	20,7	18,2	15,5	13,9

Under the influence of market conditions, external and internal food situations, the ratio of acreage in Ukraine has changed significantly in recent years. That is, there have been significant changes in the structure of acreage not in favor of crop rotations and the feed production industry. Most Ukrainian scientists believe that in the structure of sown areas, fodder should be 50-60 % (Sprynchuk N. A., Voronetskaya I. S., 2022, Shapoval I. S.; Veklenko Yu.a., Kravchenko V. P., 2024).

In 2000, cereals accounted for 50.2%, technical 15.4%, and fodder 26.0% of arable land. However, over the years, prices for cereals and technical crops (except sugar beet) have significantly increased, and the number of animals has significantly decreased. Therefore, in the total structure of sown areas, the area of industrial crops increased in 2021 to 32.3%, including the share of sunflower increased from 8.4% to 23.1%. The percentage of forage crops decreased from 26.0% in 2000 to 9.6% in 2010 and to 5.4% in 2021.

As noted by leading scientists of the Institute of feed and Agriculture of Podillia of the National Academy of Agrarian Sciences of Ukraine Sprynchuk N. A. (2022), Korniychuk O. V., Petrichenko I. V. (2022), over the past decades in the field of animal husbandry of the leading countries of the world have undergone extremely high rates of transformation, called "revolution in animal husbandry". This was accompanied by significant technological innovations and structural changes in this sector. At the same time, millions of rural residents continue to keep animals within traditional production systems, which helps to ensure the livelihood and food security of the state, to a large extent inherent in the modern Ukrainian Village.

Does this trend of rapid reduction of forage land correspond to the modern needs of animal husbandry, especially cattle breeding? This question can only be answered by analyzing the structure and productivity of the feed wedge in a particular enterprise.

An example of a modern medium-sized agricultural enterprise is the private enterprise "Agro-Novoselovka 2009" of Novovodolazhsky district, Kharkiv region. It is located south-east of the Regional Center on the border of forest-steppe and steppe natural zones and has a meat and grain specialization. The total land area of the enterprise over the past three years has been unchanged at about 3 thousand hectares, and the area of crops increased from 2553 hectares in 2021 to 2753 hectares in 2022, in 2023 the area of agricultural land was 2888.4 hectares, hayfields 29.9, pastures 105.5 hectares. The number of cattle increased in three years from 564 to 713 heads, or 26.4%, while the

number of pigs had a steady tendency to increase from 2,875 to 6,634 heads, or 2.3 times.

The company uses scientifically verified and tested 7-field crop rotations and effective agrotechnical techniques in crop production, thanks to which the genetic potential of varieties and hybrids of agricultural crops is realized.

Crop rotation is quite a powerful tool that allows you to solve many problems, both agronomic and economic. In modern agriculture, with the deepening of the processes of specialization and concentration of production, the role of crop rotations increases (Senik I. I., 2020). Neither fertilizers and irrigation, nor pesticides used in the cultivation of agricultural crops, do not allow you to completely get rid of weeds, pests and diseases. Moreover, the better the land is fertilized and irrigated, the more favorable conditions are created for the development of weeds and diseases.

Of course, not all cultures are demanding of their rapid return to the same field. For example, corn, millet, buckwheat can be grown even for several years in a row without significantly reducing the yield. But repeated sowing of peas, sugar beet, wheat, barley, oats can have a very negative impact on the harvest. But they respond well to proper placement in crop rotation and the choice of a predecessor.

The use of nine different crops at the enterprise makes it possible to select the best and best predecessors (Table 3).

Table 3

Indicators of crop production development of PE "Agro-Novoselovka 2009"

Field crops	2021			2022			2023		
	ha	c	%	ha	c	%	ha	c	%
Cereals and legumes, total	1778	83427,7	69,6	1936	77567	72,2	1867	82199	67,6
including winter wheat	849	34228,3	33,3	1029	42970,6	38,4	567	24927,6	20,6
spring wheat	-	-	-	214	7146,6	8,0	468	13886,8	17,0
winter barley	-	-	-	10	522,6	0,4	106	2549,4	3,9
spring barley	277	8092,8	10,8	223	6221,8	8,3	132	4485,2	4,8
corn for grain	460	37812,6	18,0	351	17533,6	13,1	411	30475,8	14,9
peas	192	3294,0	7,5	109	3171,8	4,1	183	5874,8	6,6
Technical crops, total	669	9981,7	26,2	617	12543,9	23,0	679	12120,1	24,6
including sunflower	669	9981,7	26,2	617	12543,9	23,0	647	11886,7	23,5
mustard							32	233,4	1,2
Feed-total	106	31165,9	4,2	127	29567,7	4,7	207	49223,8	7,5
including corn for silage and green feed	83	30521,9	3,3	127	29567,7	4,7	207	49223,8	7,5
annual herbs	-			-	-		-	-	-
perennial grasses for hay	23	644	0,9	36	1872	0,7	56	3808	0,9
Total crops	2553		100	2706	-	100	2819	-	100

The total area of Zeon and leguminous crops sown over the past three years has not changed significantly, in 2022 it increased by 8.9 %, in 2023 only by 5.0% compared to 2021. however, it should be noted that the area of winter wheat sown in 2022 increased by 180 hectares or 25% compared to 2021. And this is quite logical, because in the spring of 2022, due to Russian military aggression, it was extremely difficult or even impossible to conduct a spring sowing campaign (this includes direct military operations near the farm, and a lack of fuel, fertilizers, herbicides, etc.). In this regard, in 2023, the area of winter wheat sown decreased by almost 2 times compared to 2022, while the area of spring wheat, on the contrary, increased by 2 times and reached 416 hectares. The area sown with spring barley decreased in 2022 by 54 hectares, in 2023 – by 145 hectares or 2 times. The sown area of industrial crops, namely sunflower, in 2022 decreased by 52 hectares (8.4 %), in 2023– increased by 30 hectares. As for the area of forage crops, it had a positive trend for animal husbandry, that is, it increased in 2022 by 19 hectares, in 2023 by 101 hectares or 2 times compared to 2021. The area of corn increased 1.5 times in 2022, and 2.5 times in 2023, and the area of perennial grasses per hay (alfalfa) increased 1.5 and 2.4 times, respectively. This made it possible to increase the harvesting of silage in 2023 by 1.6 times and hay in 2022 by 3 times , in 2023 by 6 times.

Analyzing the structure of the area of crops, it should be noted that in 2021 the percentage of grain and leguminous crops was 69.6%, technical – 26.2%, fodder – 4.2 %. In 2022, their ratio did not change significantly and amounted to 72.2, 23.0 and 4.7%, respectively. In 2023, the same trend was observed, with the exception of a significant increase in the share of forage land by 90%, and the ratio was 67.8, 24.7 and 7.5 %. The share of winter wheat has changed significantly. So, in 2021 it was 33.3%, in 2022 it was 33.3%. it increased to 38.4%, and in 2023, due to the impact of force majeure factors of the war and the inability to hold the autumn sowing of 2022, it decreased by 2 times to 20.6%. The percentage of spring wheat, on the contrary, doubled from 8.0% to 17.0%. In the structure of crops, spring barley halved, from 10.8% in 2021 to 4.8% in 2023. it should be noted a positive trend of a steady decrease in the share of industrial crops, namely sunflower, from 26.2% in 2021 to 23.0% in 2022, and to 24.7% in 2023. on the contrary, the share of fodder areas increased from 4.2% in 2021, to 4.7% in 2023. 2022 and up to 7.5% in 2023, which is positive for feed production and providing animals with feed.

So, thanks to this ratio of areas of individual crops and 7-field crop rotation, all crops were provided with excellent predecessors. So, winter wheat was sown after legumes (peas), perennial legumes and corn for silage. However, their area for the entire area of wheat sowing was not enough, so for the second part, the predecessor was corn for grain with manure application at the rate of 10 t/ha. It should be noted that the state of emergency "Agro-Novoselovka 2009" contains about 700 heads of cattle, which annually produces about 4500-5000 tons of manure for crop production. However, it is not possible to make the 10t/ha of manure indicated in the technological maps for all crops, since the company has only 2,800 hectares of crops. Therefore, manure is applied under undesirable precursors in order to enrich the soil with nitrogen.

Corn does not belong to crops that are very demanding of their predecessors, it grows best after winter crops, legumes, perennial grasses, buckwheat. The best precursors for alfalfa are winter and early spring crops, such as wheat, barley, and oats. They leave the soil in a clean phytosanitary state, which contributes to better rooting of alfalfa (Kamenshchuk B. D., 2020).

It should be noted that a negative factor is the presence of 23-26% of sunflower seeds in the structure of the company's acreage. That is, it indicates that the Sunflower returns to its field every four years. There is one very important rule: sunflower seeds, in crop rotation, must be returned to their previous place no earlier than six to eight years

(Butko V. Ya., 2014).

Therefore, it is necessary to place sunflower seeds in crop rotation after crops that usually leave a little nitrogen in the soil. Such precursors can be winter cereals. Excess nitrogen can cause overgrowth and delayed maturation of sunflower seeds, which is highly undesirable. As a rule, it is optimal for sunflower in the crop rotation to allocate one field in such a way that it is returned to this field no earlier than in 4-5 years, and if there is winter rapeseed and sugar beet in the crop rotation – in 7-8 years. Thanks to this, it is possible to almost completely avoid the defeat of crops by diseases and pests.

But if you grow sunflower seeds every 2-3 years, a variety of parasites (broomrape, white and gray rot, powdery mildew, etc.) spread significantly. This leads to a decrease in yield and deterioration of seed quality. Corn (both for silage and grain), winter wheat, spring ears, and legumes do not use the moisture of deep soil horizons, so these crops are the best as precursors for sunflower (Petrenko S. D., 2021).

The advantage of sunflower as a precursor for other crops is determined by the climatic conditions during its cultivation. In regions where there is a large amount of moisture, it is considered a fairly good precursor for winter wheat, as well as for other winter cereals. This plant permeates the soil with its powerful roots, which creates favorable conditions for the next crop to assimilate a large volume of soil. In the field, sunflower leaves approximately 7 t / ha of dry organic mass of plant residues, they must be crushed and embedded in the soil so that the next crop can use nutrients. Sunflower plant residues contain a significant amount of magnesium and potassium, which is why subsequent crops, in most cases, do without potash fertilizers, but instead, after sunflower, almost completely exhausted reserves of nutrients (especially nitrogen) and moisture reserves (Petrenko S. D., 2021).

The most important criterion for evaluating fodder and grain crops is their feed value based on 1 ha (table. 4). The most effective crop in terms of nutrient yield in all years was corn for grain. The company uses a high-yielding early-maturing hybrid of corn Kharkiv 195 MV. For its sowing, the required rate of manure of 10 tons and mineral fertilizers $N_{60}P_{60}K_{90}$ was applied per hectare. Thus, in 2021, with a yield of 82.2 c/ha, the yield of feed units from 1 ha was 106.0 C, and crude protein 731.6 C. In second place in terms of the yield of feed units was corn for silage (99.3 C) with a yield of 367.7 c/ha of green mass.

Table 4

Nutritional indicators of grain and fodder crops crop production PE "Agro-Novoselovka 2009", C

Crops	2021			2022			2023		
	c/ha	f.u/ha	cr.pr/ha	c/ha	f.u/ha	cr.pr/ha	c/ha	f.u/ha	cr.pr/ha
winter wheat	40,3	47,6	487,9	41,8	49,3	428,9	44,0	51,9	451,4
spring wheat	-	-	-	33,4	39,4	342,7	29,7	35,0	304,7
winter barley	-	-	-	52,3	62,8	538,7	24,1	28,9	248,2
spring barley	29,2	33,6	280,3	27,9	32,1	267,8	34,0	39,1	326,4
corn for grain	82,2	106,0	731,6	50,0	64,5	445,0	74,2	95,7	660,4
peas	17,2	20,5	326,8	29,1	34,6	552,9	32,1	38,2	609,9
corn for silage and feed	367,7	99,3	786,9	232,8	62,9	498,2	237,8	64,2	508,9
perennial grasses for hay	58,0	29,6	696,0	52,0	26,5	624,0	68,0	34,7	816,0

The third place was taken by winter wheat (47.6 centners per unit/ha) and spring barley – 33.6 centners per unit/ha. The last place was occupied by legumes, perennial grasses for hay (alfalfa) – 29.6 C and peas – 20.5 C. In terms of crude protein yield (SP), corn for silage – 786.9 and for grain-731.6 C K/Ha was in the first place. In second place were perennial grasses for hay 696.0 C with a yield of 58.0 c/ha. The third place was taken by winter wheat (487.9 C). And on the last one – peas (326.8 C) and spring barley (280.3 C).

In 2022, crop productivity was affected by many force majeure factors of martial law and objective factors affecting climate change. However, due to the noted factors of influence, no manure was applied for spring crops, and mineral fertilizers were applied only in the form of top dressing at the level of N30P30K30. The yield of winter wheat and barley sown in autumn 2021 turned out to be quite high due to favorable weather factors in winter, and amounted to 41.8 and 52.3 C/ha, respectively. However, critically difficult spring and summer cultivation and top dressing of plants led to a decrease in the yield of spring and fodder crops. Thus, the yield of spring wheat decreased to 33.4 C/ha, corn for grain to 50.0 c/ha (by 64.0%), corn for silage to 232.8 c/ha (by 58.2%), perennial grasses for hay to 52.0 c/ha (by 11.5 %). Accordingly, the yield of nutrients from 1 ha of crops also decreased, which led to changes in the ratio between individual crops in terms of their efficiency. Thus, the first places in terms of the yield of feed units per hectare of area were traditionally occupied by corn for grain (64.5 C) and silage (62.9 C) and spring barley (62.8 C). The second place was taken by winter wheat – 49.3 C, the third – spring wheat (39.4 C) and peas (34.6 C), and the last place was taken by perennial grasses for hay 26.5 C K/ha. The highest yield of crude protein per hectare, on the contrary, was observed in perennial grasses for hay 624.0 C, in second place due to their high yield were peas (552.9 C) and spring barley (538.7 C). The third place was taken by corn for silage (498.2 C) and grain (445.0 C). And in last place were spring wheat (342.7 C) and barley (267.8 C).

In 2023, 10 tons of manure and mineral fertilizers $N_{60}P_{60}K_{90}$ were applied for grain crops. However, crop production in eastern Ukraine was significantly affected by the weather conditions of the summer period, such as insufficient precipitation and elevated temperatures. It should be noted that most crops have increased their yields, including winter wheat (by 2.2 centners/ha), spring barley (by 6.1 centners/ha), and perennial grasses (by 16 centners/ha), peas – by 3 centners/ha and corn for silage (by 5 centners/ha). On the contrary, spring wheat decreased its yield by 3.7 c/ha, winter barley by 2.2 times to 24.1 C/ha. Despite different weather conditions and the level of fertilizer application, as in all years, the highest yield of feed units per hectare was 95.7 C. in second place was corn for silage – 64.5 C, followed by winter wheat – 51.9 C, then spring barley, wheat and peas. The lowest yield of feed units was obtained from perennial grasses for hay 34.7 C, and crude protein per hectare of area – on the contrary, the largest 816 C. in second place were corn for grain (660 C) and peas (609 C), corn for silage 509 C. the lowest amount of crude protein per hectare was determined for spring wheat and barley grains.

As a result, we note that the most effective forage crop for three years, under various conditions, even under adverse weather conditions, was the hybrid corn Kharkiv 195 MV, both for silage and grain.

Discussion. The analysis showed that over the past 20 years, the structure of sown areas of agricultural crops has changed significantly. Thus, the area of grain and leguminous crops increased slightly by 17.6%, and the area of sunflower by 2.3 times, and in the structure of crops from 11.0% in 2020 to 23.2% in 2021. in contrast, the area of fodder crops decreased during this period from 26.0% to 5.4%, which is a negative factor for the development of animal husbandry. Scientists, based on monitoring the state

of development of feed production in Ukraine, claim that in the field of crop production in the future there is a negative trend towards reducing the area under forage crops (Ambrosov V. Ya., 2009). However, according to N. A. Sprynchuk (2020), there is a tendency to increase crop production in Ukraine due to more intensive use of land resources and an increase in crop yields.

In recent years, Ukraine has seen an increase in the average annual temperature and a sharp increase in uneven precipitation throughout the year. As a result, the frequency of repeated droughts and floods has more than doubled (Petrichenko V. F., 2022). These changes significantly affect the development of feed production and the provision of feed to farm animals.

Analysis of the average size (2816 ha) of the agricultural enterprise PE "Agro-Novoselovka 2009" of Novovodolazhsky district, Kharkiv region confirmed the general state trend of changes in the structure of sown areas. So, over the past three years, the share of grain and leguminous crops was 67-72%, sunflower – 23.6-26.2%, fodder increased from 4.2% to 7.5 %. This made it possible to increase silage harvesting in 2023 by 61.3% and hay harvesting by 6 times compared to 2021.

The company uses 7-full crop rotations and effective agrotechnical techniques in crop production, so that good predecessors are selected for agricultural crops. Unfortunately, the company has disadvantages due to the discrepancy between the areas of the best predecessors and the rotation of crops, because the best predecessors are chosen for more economically profitable crops, such as winter wheat, corn for grain and sunflower. Another significant disadvantage is that the company does not have pure or black vapors, which are the best precursors for winter wheat..

The most effective forage crop for three years under various conditions, corn for both silage and grain, perennial grasses for hay (alfalfa) provided the lowest yield of feed units per hectare, and crude protein, on the contrary, the largest amount. And in the structure of acreage, they occupied less than one percent. However, according to Petrichenko V. F. (2022) and Petrichenko I. V. (2013), perennial legumes in the structure of sown areas of forage crops should occupy 50-55% and their characteristic feature is that they can grow on various types of soils, including low-fertile and eroded.

When selecting perennial grasses for sowing, it is necessary to pay attention to their yield and feed value. In studies by Kulik M. et al. (2024), millet (56.0 t/ha) and perennial sorghum (53.6 t/ha) had the highest biomass yield in forest-steppe conditions. However, it should be noted that grass mixtures of three or four components have an advantage over single-component ones. According to the NSC "Institute of Agriculture of the National Academy of Sciences", the highest yield was provided by alfalfa-runoff mixture, namely (on average for four years): single-species alfalfa sowing – 7.84 t / ha; alfalfa + eastern fescue-9.32 t / ha; alfalfa + gryastitsa combined-8.82 t/ha; alfalfa + stoeless runoff – 10.3 t / ha of dry matter (Kukharchuk P. I., Slyusar S. M., 2017).

The most common grass mixtures, as in the western and northern parts of Ukraine as a whole, were and are mixtures of oats with legumes, most often with spring vetch (Hetman N. Ya., 2023, Lekhman A.V. et al., 2023). This is confirmed by V. F. Petrichenko (2022), who notes that it is advisable to grow two-and three-component legume-cereal-cabbage mixtures using oats with cabbage or legumes, which in the structure of crops of annual fodder crops should occupy at least 65-70 %.

In turn, N. Ya.Getman and L. M. Burko (2023) add that in the conditions of the central forest-steppe of Ukraine, alfalfa sown for three years of life and 2 years of use is able to fix 735 kg/ha of nitrogen from the air, enriching the soil by 598 kg/ha with nitrogen. During spring coverless sowing in two slopes, alfalfa captures 173 kg/ha of nitrogen from the air, leaving it in the soil 148 kg/ha. Also, the results of the study of the

nitrogen-fixing ability of alfalfa and increasing soil nitrogen saturation are confirmed by Olifirovich V. F., Veklenko Yu.a. (2022). Didur I. M., Shevchuk V. V. (2020) prove that legume-rhizobial systems capture 40 to 300 kg of nitrogen per 1 ha of crop each year from the atmosphere.

Studies of Rudavskaya N. M. and Oyster Mushroom V. V. (2018) found that on average for two years of using the grass stand, the highest yield (9.0 t/ha of dry weight) was provided by legume-cereal grass mixture, fertilized normally N60P60K90 with seed treatment with rhizobophyte and polymyxobacterin biologics.

Summing up the above, we can make suggestions for improving crop rotations in general, and feed production in particular. The company needs to introduce mandatory pure or black pairs in the amount of 10-15% of the sown area into the structure of sown areas, in conditions of climate change, increase the feed wedge to the desired minimum 10-15%, especially in conditions of climate change, it is desirable to increase the area of sowing perennial grasses inside the feed areas, as well as use two-or three-component (legume-cereal) feed mixtures with the proportion of legume grasses at the level of 50-60 %.

Conclusions: Based on the conducted studies to establish the influence of agroclimatic features on the intensity of forage crop cultivation, the following conclusions can be drawn:

1. Research has established that over the past 20 years in Ukraine, the structure of sown areas of agricultural crops has changed significantly. Their total area increased by 1.5 million square meters. ha or by 5.5 %. The area under grain crops increased by 2.4 million hectares. ha or 17.6 %, wheat by 1.6 million.ha (30.1 %), corn – from 1.36 to 5.52 million hectares. ha or 4.0 times. Over the years, the area of industrial crops has increased 2.2 times. However, the area under sugar beet cultivation decreased from 856 to 227 thousand hectares, or 3.7 times, and the area under sunflower increased by 3.7 million hectares. ha or 2.3 times.

2. It is proved that in the structure of acreage in Ukraine in 2000, cereals accounted for 50.2 %, technical 15.4%, fodder 26.0%. However, over the years, prices for cereals and technical crops (except sugar beet) have significantly increased, and the number of animals has significantly decreased. Therefore, in the total structure of sown areas, the area of industrial crops increased in 2021 to 32.3%, including the share of sunflower increased from 8.4% to 23.1%. The percentage of forage crops decreased from 26.0% in 2000 to 9.6% in 2010 and to 5.4% in 2021.

3. The analysis of the state of feed production in PE "Agro-Novoselovka 2009" Novovodolazhsky district, Kharkiv region over the past three years is carried out. It is established that the company uses scientifically verified and tested 7-field crop rotations and effective agrotechnical techniques in crop production, thanks to which the genetic potential of varieties and hybrids of agricultural crops is realized.

4. Analyzing the structure of acreage in the enterprise, it was found that the area of grain and industrial crops has practically not changed over the past 3 years, and the area of fodder crops has increased from 4.2% to 7.5%. However, sunflower seeds have a critical amount (23-26% of the total area sown) for the formation of optimal crop rotations.

5. Analysis of crop productivity showed that corn for both silage and grain proved to be the most effective forage crop in three years, under various conditions, even under adverse weather conditions. And in terms of raw protein yield, perennial grasses for hay were the most effective.

6. To effectively overcome the negative factors of changes in climatic conditions and the structure of acreage, the company is proposed to introduce mandatory pure or

black pairs in the amount of 10-15% of the acreage into the structure of acreage, in the conditions of climate change, increase the feed wedge to the desired 10-15%, and inside the fodder areas it is desirable to increase the area of sowing perennial grasses, as well as use two-or three-component (legume-cereal) feed mixtures with the proportion of legume grasses at the level of 50-60%.

References

Adamenko, T.I. (2023). Transformation of the structure of agricultural production in the face of climate change. [Transformatsiya struktury vyrobnytstva sil′s′kohospodars′koyi produktsiyi v umovakh zmin klima]. Scientific Collection «InterConf». 34(159): with the Proceedings of the 4th International Scientific and Practical Conference «Concepts for the Development of Society's Scientific Potential». available at**:** https://archive.interconf.center/index.php/2709-4685/article/view/3896 (in Ukrainian)

Ambrosov, V.Ya. (2009). Collection of scientific works. T.1. Economics of feed production and animal husbandry. .[Tendentsiyi zminy chastoty ta intensyvnosti ekstremal′nykh hidrometeorolohichnykh yavyshch na terytoriyi Donets′koyi oblasti]. Kh.: NSC "*Institute of Agrarian Economics*" 412 c. (in Ukrainian)

Balabukh, V.O. (2022). Trends in the frequency and intensity of extreme hydrometeorological phenomena in the territory of Donetsk region. available at**:** http://eco.com.ua/content/tendenciyi-zmini-chastoti-ta-intensivnosti-ekstremalnih-gidrometeorologichnih-yavishch-na. (in Ukrainian)

Butko, V.A. (2023). The influence of climatic conditions on the development of agricultural crops. [Vplyv klimatychnykh umov na rozvytok sil′s′kohospodars′kykh kul′tur]. *Agronomic articles*.№ 3. C. 44-50. available at**:** https://himagro.com.ua/vpliv-klimatichnix-umov-na-rozvitok-silskogospodarskix-kultur (in Ukrainian)

Voloshchuk, O.P., Voloshchuk, I.S., Hlyva, V.V., Pashchak, M.O. (2021). Weather conditions as a factor influencing the growth and development of corn plants in the western forest-steppe of Ukraine. [Pohodni umovy yak chynnyk vplyvu na rist i rozvytok roslyn kukurudzy v zakhidnomu Lisostepu Ukrayini]. Sciences of Europe. № 71. C. 3-8. available at**:** https://cyberleninka.ru/article/n/pogodni-umovi-yak-chinnik-vplivu-na-rist-i-rozvitok-roslin-kukurudzi-v-zahidnomu-lisostepu-ukrayini/viewer (in Ukrainian)

Hetman, N.Ya., Burko, L.M., Svystunova, I.V. (2023). Productivity of alfalfa seed in organic production of plant raw materials under climate change conditions. Scientific Reports of the National University of Life Resources and Environmental Sciences of Ukraine. No. 2. P. 86-90 http://nbuv.gov.ua/UJRN/Nd_2023_2_9 (in Ukrainian)

Global climate change – current views and trends (Ukrainian Hydrometeorological Center). available at**:** http://meteo.gov.ua/ua/33837. (in Ukrainian)

Didur, I.M., Shevchuk, V.V. (2020). Increasing soil fertility as a result of the accumulation of biological nitrogen by legumes. [Pidvyshchennya rodyuchosti hruntu v rezul′tati nakopychennya biolohichnoho azotu bobovymy kul′turamy]. *Agriculture and Forestry*. No. 16. P. 50-62. DOI: 10.37128/2707-202016 (in Ukrainian)

Kamenshchuk, B.D. (2020). Ways to increase the efficiency of growing corn for grain. [Shlyakhy pidvyshchennya efektyvnosti vyroshchuvannya kukurudzy na zerno]. *Feed and feed production*. No.89. P. 85-92. DOI: 10.31073/kormovyrobnytstvo202089-08 (in Ukrainian)

Kvitko, G.P., Getman N.Ya. (2013). Nitrogen-fixing capacity and soil enrichment with nitrogen depending on the years of life of alfalfa in the conditions of the Forest-Steppe. [Azotfiksuyucha spromozhnist′ ta zbahachennya gruntu azotom zalezhno vid rokiv zhyttya lyutserny posivnoyi v umovah Lisostepy]. *Feed and feed production.* Issue.51. P. 54–57. DOI: https://doi.org/ 10.31027/kormovyrobnytstvo201354-57 (in Ukrainian)

Kulyk M.I., Pryshlyak N.V., Demin D.G., Pisarenko D.O. (2024). Plant raw materials of energy crops: yield, biomass volume and its quality. [Roslynna syrovyna enerhetychnykh kul′tur: vrozhaynist′, obsyah biomasy ta yiyi yakist′. Kormy i kormovyrobnytstvo]. *Feed and feed production*. No. 98. P. 85-92. DOI: 10.31073/kormovyrobnytstvo202497-09. (in Ukrainian)

Kukharchuk, P.I., Slyusar, S.M., Artyushenko, O.O. (2017).Alfalfa yield in single-species crops and mixtures with cereals. [Urozhaynist′ lyutserny v odnovydovykh posivakh i sumishkakh zi zlakamy]. Collection of scientific works of the National Research Center *"Institute of Agriculture of the Ukrainian Academy of Sciences"*. Kyiv: "EKMO". Issue. 1. P. 125-131. available at**:** http://nbuv.gov.ua/UJRN/znpzeml_2007_1_26 (in Ukrainian)

Lyahu, D.S. (2024). Adaptation of agriculture to climate change: challenges and strategies. [Adaptatsiya sil′s′koho hospodarstva do klimatychnykh zmin: vyklyky ta stratehiyi.]. VI International Scientific and Practical Conference "The Impact of Climate Change on the Spatial Development of the Earth's Territories: Consequences and Solutions" (June 10, 2024). KhDAU. P. 30-36. available at**:** https://www.ksau.kherson.ua/files/konferencii/2024/09/mater_28_06_24.pdf (in Ukrainian)

Mashchak, Ya.I., Rudavska, N.M. (2021). The influence of fertilizers and biological products on the productivity of legume-cereal grass mixture. [Vplyv udobrennya i biopreparativ na produktyvnist′ bobovo-zlakovoyi travosumishky]. *Feed and feed production.* Issue. 70. P. 76–79. DOI: https://doi.org/ 10.31048/kormovyrobnytstvo202170-7679 (in Ukrainian)

Olifirovych, V.F., Veklenko, Yu.A., Chinchyk, O.S. (2022). Dynamics of the species composition of alfalfa-cereal agrophytocenosis depending on the technological methods of cultivation. [Dynamika vydovoho skladu lyutserno-zlakovoho ahrofitotsenozu zalezhno vid tekhnolohichnykh pryyomiv vyroshchuvaannya.]. *Feed and feed production.* Issue 94. P. 67-74. DOI: 10.31073/kormovyrobnytstvo202294-07 (in Ukrainian)

Petrenko, S.D., Petrenko, O.V. (2021). Fodder production. [Kormovyrobnytstvo]. K. 452 p. available at**:** https://agroscience.com.ua/sites/default/files/library/files/user585/kormovyrobnyctvo.pdf (in Ukrainian)

Petrichenko, V.F. (2022). Crop production: new technologies for growing field crops. Lviv. 2022. 806 c. available at**:** https://profbook.com.ua/roslynnitstvo.html?srsltid=https://profbook.com.ua/roslynnitstvo.html?srsltid= (in Ukrainian)

Petrychenko, V.F., Korniychuk, O.V., Babych, A.O., Bugayov, V.D., Kulyk, M.F. (2017). Concept of feed production development in Ukraine for the period until 2025. [Kontseptsiya rozvytku kormovyrobnytstva v Ukrayini na period do 2025 roky]. Vinnytsia. 12 p. (in Ukrainian)

Petrychenko, I.V. (2013). Factors and determinants of the development of the feed industry in Ukraine. [Faktory i determinanty rozvytku haluzi kormovyrobnytstva v Ukrayini Faktory i determinanty rozvytku haluzi kormovyrobnytstva v

Ukrayini]. *Innovative economy*. Ternopil. No 11 (49). P. 27-33. available at**:** http://library.wunu.edu.ua/libsearch/DocDescription?doc_id=315314 (in Ukrainian)

Senyk, I.I. (2020). Forage productivity of winter forage agrophytocenoses depending on elements of cultivation technology. [Kormova produktyvnist′ ozymykh kormovykh ahrofitotsenoziv zalezhno vid elementiv tekhnolohiyi vyroshchuvanna]. *Podolskyi visnyk. Kamianets-Podilskyi*. Issue 32. P. 68-72. DOI: https://doi.org/ 10.37406/2706-9052-2020-1-8. (in Ukrainian)

Sprynchuk, N.A. (2022). Algorithm for determining optimal volumes of commercial feed production for a farm. [Alhorytm vyznachennya optymal′nykh ob″yemiv tovarnoho kormovyrobnytstva dlya fermers′koho hospodars]. *Agrosvit.*. No 4. P. 76-82. DOI: 10.32702/2306-6792.2020.4.76 (in Ukrainian)

Sprynchuk, N.A., Voronetska, I.S., Korniychuk, O.O., Petrychenko, I.I. (2022). Climate crisis and features of modernization of field feed production in developed countries of the world. [Klimatychna kryza ta osoblyvosti modernizatsiyi pol′ovoho kormovyrobnytstva v rozvynenykh krayinakh svitu]. *Feed and feed production*. Issue 94. P. 105-112. DOI: 10.31073/kormovyrobnytstvo202294-11

Statistical collection "Agriculture of Ukraine" for 2021. Electronic resource. available at**:** http://www.ukrstat.gov.ua/ (in Ukrainian) (in Ukrainian)

Shapoval, I.S.; Veklenko, Yu.A., Kravchenko, V.P., Yarmilko, S.A., Yaremych, L.V. (2024). The effectiveness of fodder crop rotations under different fertilization systems in the conditions of the Left-Bank Forest-Steppe. [Efektyvnist′ kormovykh sivozmin za riznykh system udobrennya v umovakh livoberezhnoho Lisostepu.]. *Feed and Feed Production*. Issue 97. P. 95-105. DOI:10.31073/kormovyrobnytstvo202497-10 (in Ukrainian)

FAO. 2018. The future of food and agriculture – Alternative pathways to 2050. Rome. 224 pp. Licence. available at**:** https://knowledge4policy.ec.europa.eu/publication/future-food-agriculture-alternative-pathways-2050_en

Chutney, W. (2019). The Impact of Global Climate Change on World Hunger February 27. available at**:** https://weatheringrisk.org/sites/default/files/document/UNSC_Food_Insecurity_Report_0.pdf

WMO confirms 2019 as second hottest year on recordhttps. available at**:** https://public.wmo.int/en/media/press-release/wmo-confirms-2019-second-hottest-year-record.

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PROSPECTS FOR ALTERNATIVE USE SHEEP WOOL

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This article contains the results of the authors ' review of the publicly available literature devoted to the problem of alternative (except for traditional textile) use of sheep wool in various areas of production. It reflects the decline in total wool production and purchase prices for it, as well as outlines the relevance for the development of the sheep industry of finding alternative ways to use it. It is shown that due to its specific physical and technical properties, wool is increasingly used as an environmentally friendly material for the manufacture of composite new building materials-heat and noise – insulating materials, plaster, bricks, concrete. It is also used as mulch and agrofibre substitutes in agricultural technologies for growing grain, vegetable crops, berries and sugar beet, as well as in Horticulture, Forestry and landscape gardening. At the same time, in new materials and technological processes made with the use of wool, heat and sound insulation and mechanical properties (building materials) are significantly improved, yield (crops) increases and the temperature and biological regime of the soil, its saturation with nitrogen, carbon and some minerals improves. Wool, as a natural product that is subject to the action of biodegradation processes, contributes to the greening of production and reducing its energy intensity.

Keywords: alternative use, sheep wool, building materials, energy and eco - efficiency.

ПЕРСПЕКТИВИ АЛЬТЕРНАТИВНОГО ВИКОРИСТАННЯ ОВЕЧОЇ ВОВНИ

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Дана стаття містить результати виконаного авторами огляду наявної у відкритому доступі літератури, що присвячена проблемі альтернативного (крім традиційного текстильного) використання овечої вовни в різних сферах виробництва. В ній відображено зниження загального виробництва вовни та закупівельних цін на неї, а також окреслено актуальність для розвитку галузі вівчарства пошуку альтернативних шляхів її використання. Показано, що завдяки своїм специфічним фізико-технічним властивостям, вовна все більше використовується в якості екологічно чистого матеріалу для виготовлення композиційних нових будівельних матеріалів – тепло- та шумоізолюючих матеріалів, штукатурки, цегли, бетону. Також знаходить застосування в якості мульчі та замінників агроволокна в агротехнологіях вирощування зернових, овочевих культур, ягід і цукрових буряків та в садівництві, лісовому і садово-ландшафтному господарстві. При цьому, у нових матеріалах та технологічних процесах, виконаних з використанням вовни, значно покращуються тепло- і звукоізоляціні та механічні властивості (будівельні матеріали), збільшується врожайність (сільськогосподарські культури) та покращується температурно-вологічний режим грунту, його насиченість азотом, вуглецем та деякими мінеральними речовинами. Вовна ж, як натуральний продукт, що підпадає дії процесів біодергадації, сприяє екологізації виробництва та зниженню його енергоємності.

Ключові слова: альтернативне використання, вовна овець, будівельні матеріали, енерго- та екоефективність.

Introduction. Sheep wool is a unique renewable natural product, which is a substitute raw material resource for the development of the textile industry. Low thermal conductivity, roll capacity, hygroscopicity, elasticity and and strength firmness – this is a complex of physical and technical properties that is inherent only in natural wool fibers. Due to these properties, sheep wool has acquired a variety of uses. In the structure of its world production, fine wool accounts for 40-45%, semi – fine wool – 25-30%, semi-coarse and coarse wool-30-35%. At the same time, 2/3 of its total volume is used for the manufacture of clothing, and 1/3 – for carpet, non-woven (felted) products and technical purposes. Total wool production in 2023 in the world amounted to 1 million tons. 746 thousand tons, and in the dynamics over the past 10 years, there is a decrease in this indicator by 305 thousand tons, or by 14.9 % (according to FAO statistics, 2023).

This state of production is primarily due to a decrease in purchase prices for unwashed and washed sheep wool, due to competition in the market from cheaper synthetic fibers. Over a ten-year period, these prices in one of the leading countries producing high – quality sheep wool – New Zealand-decreased by almost 1.6 times (from 3.1 to 1.97 thousand $/ton), in Europe-by 2.5-5 times (Hungary, Poland, Germany). The price per ton of wool in Ukraine also decreased by 2.1 times. This situation encourages producers to change the specialization of the sheep industry to produce lamb and Lamb, as well as milk and other products. In this regard, there is an increasing interest in breeding sheep breeds that are characterized by a reduced ability to form runes and, as a rule, produce low-quality wool, which has a low purchase price and is not in demand in the textile industry. Along with this, the search for ways to reduce the cost of production of sheep products by reducing energy consumption when creating sheep breeding facilities has become relevant.

Authors (Berge, B., 2001; Korjenic A., Klarić S. et al., 2015) in their experimental studies give the main directions for improving the efficiency of the livestock complex. The researchers emphasize that sheep storage facilities don't have to be complex or expensive. Therefore, preference is given to the reconstruction of existing livestock premises in the direction of improving the technological efficiency and ergonomics of the main production processes, as well as ensuring appropriate microclimate conditions and comfortable keeping of animals. As an alternative to capital rooms for keeping sheep, light structures and awning shelters are considered. This is not only about reducing energy consumption and loss, but also about reducing the cost of construction or reconstruction through the use of natural and locally available materials, including the products of the sheep themselves, namely their wool. An important condition for creating livestock production facilities is their environmental friendliness – minimal impact on the deterioration of air and water quality, as well as soil cleanliness.

Against the background of the above - mentioned depreciation of wool, in the system of measures to improve the efficiency of sheep production, in addition to finding ways to reduce capital and current costs, there is also an urgent issue-expanding the range of its application in various fields. Therefore, the purpose of this publication was to study ways of alternative (except for the textile industry) use of sheep wool in other industries.

Materials and methods. The object of research was the directions and trends, as well as the effectiveness of using sheep wool in various production areas. The research was carried out by accumulating, analyzing and summarizing the data available in open publications on the efficiency and prospects of using sheep wool in various production areas. Systematization of primary information was carried out using bibliographic and source analysis.

Research results. In the context of preventing global environmental challenges, improving the ecology of building materials plays an important role. This problem is most widely covered in the book" The Ecology of Building Materials " (Berge B.,2001). From the point of view of ensuring sustainable development, it shows the feasibility of wider use of local environmentally friendly raw materials for the production of building materials, which are easily processed and restored. These include wood, straw, biological fibers, and sheep wool. At the same time, it is important that technologies with a low level of energy consumption are used in the process of processing it. According to the author, solving this problem requires scientific and intersectoral interaction. Therefore, manufacturers ' interest in obtaining cheap new construction and thermal insulation materials of organic nature is spreading in the world.

According to korjenic A., Klarić I., Hadžić A. and Korjenic S. (2015) the construction industry accounts for up to 40% of CO_2 emissions and is characterized by

high energy intensity of production processes. Therefore, according to them, research aimed at finding alternative renewable natural building materials can significantly affect the reduction of these indicators.

In European countries, the importance of solving the problem of reducing environmental challenges associated with building materials is emphasized by the relevant directive - 2010/31/EC of the European Parliament and the European Council (of 19 May 2010). It sets out the requirements that the building energy efficiency certificate must contain information on the actual impact of heating and cooling on the building's energy needs, as well as on primary energy consumption and carbon dioxide emissions.

The use of wool waste as a new raw material in the construction industry, according to Torring T., Sandvik K. (2000) is an excellent opportunity to create a closed economy model, as it reduces the need for non-renewable resources, reduces the carbon footprint, and is a resource-saving method for producing industrial components, which can also play a significant role in energy conservation and reducing environmental pollution.

Now, in order to effectively use wool as waste from agricultural production and its use in other production areas, Midolo G., Valenti F. (2024), an organizational model based on the Geographic Information System (GIS) is proposed for determining the location and amount of waste (sheep wool) and sustainable management of the use of these raw materials as a potential component of green building.

So, from the above-mentioned reports, it can be stated that a possible area of widespread alternative use of wool is the construction industry.

Among the promising areas for improving the energy efficiency of buildings, first of all, the issue of improving their thermal insulation characteristics through the use of new insulation materials is considered. So, Korjenic A.; Petránek, V. et al. (2011) note that thermal insulators made of natural materials are likely to become an acceptable alternative to widely used boards made of various artificial materials (mineral wool, polystyrene or polyurethane), since they are quite competitive in their technical and technological characteristics. So Zach J., Korjenic A., Petránek V. et al. (2012) propose to study sheep wool as an alternative thermal insulation biomaterial with good thermal and acoustic properties to improve building comfort and reduce carbon emissions. The use of this natural material in construction can also be useful for preserving human health.

The advantages of wool as a heat insulator are more clearly emphasized in the message of Doroudiani S. et Omidian H. (2010) that expanded polystyrene, which is now one of the most common thermal insulation materials in construction, poses serious problems for environmental safety and human health, as it has an extremely high tendency to burn and emits toxic fumes.

According to Korjenic A., Klarić I., Hadžić A. and Korjenic S. (2015), sheep wool, evaluated by hygrothermal modeling and ecological balance methods compared to mineral wool and calcium silicate, provides better physical, energy and environmental characteristics of the thermal insulation material. At the same time , only 5.4 kg of CO_2 emissions are generated per 1 M3 in the production of insulation from sheep wool, while for mineral wool this figure exceeds 135 kg.

Ahmed A., Qayoum A., Qayoom Mir F. (2019), also consider sheep and goat wool and horsehair (Mane) as possible environmentally and cost-effective insulation materials. At the same time, they note the best hygroscopic properties of sheep and goat hair over horsehair, which is due to the different structure and thickness of The compared fibers. Goat hair was the lowest in terms of thermal conductivity among the compared materials.

The results of tests of sheep wool indicate excellent performance of the material not only in the development of construction thermal insulation elements, but also for sound insulation.

According to Borlea (Mureşan) S. I., Tiuc A.-E., Nemeş O., Vermeşan H., Vasile O.(2020) materials obtained by simple hot pressing of wool are characterized by high sound – absorbing properties with significant acoustic absorption coefficients-more than 0.7 for the frequency range of 800 ÷ 3150 Hz, and according to these characteristics, sheep wool has comparable sound-absorbing properties to mineral wool or polyurethane foam.

According to Parlato M.C.M. and Porto S.M.C. (2020), the use of sheep wool as an alternative to insulation materials such as fiberglass, rock wool, polyurethane foam, polystyrene provides significant benefits for sustainable development, reducing production costs for the production of these materials and reducing environmental pollution.

Research by Polish scientists (Kicińska-Jakubowska A. et al., 2023) prove the high efficiency of multifunctional sound and thermal insulation materials from waste natural protein and lignocellulose fibers in accordance with the principles of bioeconomics. As natural materials, they studied the low-quality wool of local coarse-wool sheep and a mixture of Bast fiber waste formed as a result of the manufacture of ropes and ropes, which were connected in various proportions by needle punching in the form of technical mats. An increase in the proportion of wool over Bast fibers helped to improve the thermal insulation properties of products, while a large proportion of Bast fibers gave products better barrier sound protection properties.

Also a number of scientists (Vasina M. et al., 2024) indicates the possibility of using sheep wool in polymer composites in the manufacture of materials with noise - and vibration-absorbing and electrical insulation properties. According to their data, the addition of wool fibers to epoxy, polyurethane and polyester resins from 3 to 5% by weight significantly improved their physical qualities.

In addition to the traditional use of wool for the manufacture of Textiles, Starkova O., Sabalina A, Voikiva V. and Osite A. (2022) emphasize the possibilities of its application in the process of reinforcing polymer composites for 3D printing.

Pederneiras C.M., Veiga R. and Brito J. (2019) focus on the feasibility of using sheep wool for reinforcing mortars and, in particular, to increase the resistance of plasters to cracking. According to their data, the use of cement and cement-lime mixture as binders with the inclusion of 10% and 20% (by volume) wool fibers with a length of 1.5 cm and 3.0 cm increased the plasticity of mortars, and improved their mechanical properties.

See Also Atbir A., Khabbazi A., Cherkaoui M. et al. (2023), investigating a new thermal insulation material - a porous plaster reinforced with a multilayer thread base made of sheep wool, note an increase in flexural strength in this material from 30% to 74% with a decrease in compressive strength from 48% to 71%. At the same time, the density of the material decreased with increasing length of wool fibers.

In search of sustainable and environmentally friendly insulation materials, Urdanpilleta M., Leceta I., Martín-Garín A. et al. (2025) conducted a study of biocomposites from sheep's wool included in a polymer matrix that bound soy protein by sublimation drying with 7, 10, 15 and 20% by weight of wool. To protect these biocomposites from insects, coffee grounds were added to the formulation. The new insulation material was not inferior in thermal conductivity to other natural analogues and showed high protective properties against keratophagous insects, especially against carpet beetles.

In addition to the above-mentioned building materials, sheep wool is also used in the production of concrete.

Thus, the positive effect on the physical and mechanical properties of concrete and sheep wool composites is noted in their publications Fantilli A. P., Sicardi S., Dotti F. (2015) and Dénes O., Florea I., Manea D.L. (2019).

Research By Alyousef R., Alabduljabbar H., Mohammadhosseini H. et al. (2020) and Alyousef R. , Mohammadhosseini H., Ebid A.A.K., Alabduljabbar H. (2022) also found that adding up to 6% of sheep wool fibers with a length of 70 mm in the production of fiber concrete reduced the shrinkage rates of fresh concrete, while reducing its compressive strength. However, with a certain period of time, the tensile and flexural strength of fiber concrete increased, which increased its ductility and ability to absorb energy. The microstructure of concrete was characterized by a decrease in the size of cavities. In addition, the addition of 2 % (in volume fractions) of fibers to Portland cement reduced the sorption coefficient of the concrete composite and the depth of penetration of chlorides into the mixtures.

Research By Abdul Awal A.S.M., Mohammadhosseini H. (2016). they prove that not only natural sheep wool, but also carpet thread waste together with palm oil ash can be effectively used in the production of concrete. Its technical characteristics-high tensile and flexural strength (increased ductility), lower porosity, higher energy absorption capacity ensures not only high quality as a building material, but also increases the environmental cleanliness of production.

In addition to facing heat and noise insulation materials and plasters, concrete production, natural sheep wool finds its application in the manufacture of bricks.

El Wardi F.Z., Ladouy S., Khabbazi A. et al. (2021) report that in the process of manufacturing unburned clay bricks, light composite materials with lower thermal conductivity and higher compressive and flexural strength were obtained by adding used cork pellets and quicklime and sheep wool fibers to it in certain proportions than bricks made using traditional technology. Clay-cork blocks reinforced with 30% lime content and 2% sheep wool fibers had the highest thermomechanical indicators. They had a low volume density of 583 kg/m3 and were characterized by increased thermal and mechanical qualities.

Similar changes in the physical and mechanical properties of bricks made of clay of various compositions were noted in their research by Galán-Marín C., Rivera-Gómez C., Petric-Gray J. (2010). According to them, sheep wool added in optimal proportions to its composition minimized shrinkage, reduced curing time and increased the compressive and flexural strength of the finished product.

Parlato M.C.M., Cuomo M., Porto S.M.C.(2022). it is also proposed to use sheep wool fibers as reinforcing components to clay in the manufacture of bricks. At the same time, they note an increase in tensile strength, ductility, impact resistance, viscosity and a decrease in shrinkage during brick drying.

According to Atbir A., Taibi M., Aouan B. et al. (2023), unburned solid bricks based on white and red clay, made with the addition of sheep wool, have fairly high thermal and mechanical characteristics and are characterized by a light specific gravity. At the same time, the positive effect of wool admixture was significant on the mechanical behavior of the developed materials after 90 days - in terms of flexural strength from 18 to 56% for white clay and from 8 to 29% for red clay. These mechanical characteristics were accompanied by an increase in thermal conductivity from 4 to 41% for white and from 6 to 39% for ore clay fractions.

Wool, due to its unique physical and technical properties, also finds its application as components of mulch in the cultivation of various agricultural crops. Thanks to it, it is

possible to control thermoregulatory processes and soil moisture, as well as enrich it with nitrogen and other substances formed during the biodegradation of wool.

In this regard, Camilli F., Focacci M., Dal Prà A. et al. (2025) note that sheep wool has great potential for use in agricultural production, as it has a high content of nitrogen, carbon, sulfur and is characterized by a high ability to absorb water and retain moisture. These properties help preserve the carbon content of the soil and increase its fertility, as well as reduce the risk of water pollution due to the gradual decomposition and release of nitrogen.

According to the research results of Gabryś T., Fryczkowska B. (2022), the use of sheep's wool by mixing it evenly with the soil when growing indoor plants Chlorophytum comosum better provided them with water and increased root system growth. At the same time, individual fibers dispersed in the soil decomposed better than fibers that were arranged in a layer at the bottom of the pot.

In the publication Broda J., Gawłowski A., Rom M. et al. (2024) reported a comparative evaluation of the use of mountain sheep wool, straw, and bark as mulch in strawberry cultivation. They experimentally proved the best thermoregulatory properties of wool compared to other materials. Wool mulch minimized fluctuations between day and night soil temperatures (fluctuations did not exceed 2-3 ° C in hot weather, which is almost five times less than in the control area and twice as much as in straw and bark mulch. In addition, the wool retained more moisture after precipitation (the mass of water retained by the wool is several times higher than its own mass). Wool, as a substance for mulch, had a much longer period of biological decomposition than bark and especially straw, and therefore can be used for a longer period. These advantages of wool contributed to an increase in Strawberry yield.

By the same researchers (Broda, J., Gawlowski, A., Rom, M., & Kobiela-Mendrek, K., 2023) it was found that during the growing season, nitrogen compounds gradually enter the soil, and the nitrogen content in it clearly correlates with an increase in the processes of dergadation of sheep wool introduced into it. The released wool nitrogen had a positive effect on wheat growth at different stages of its development, which was observed in the form of increased tillering, an increase in the height of stems and provided an increase in yield.

In the publication Taskin M. B. (2024) it is reported that the introduction of sheep wool and wool hydrolysate into the soil contributed to an increase in the yield of both stem mass (by 40.2% and 52.1%) and sugar beet root crops (by 4.59% and 7.61%). At the same time, an increase in the concentration of nitrogen, calcium, magnesium and phosphorus in the stems of plants was found, both with the use of wool and its hydrolysate, and the sugar content of beets from the specified technological method of their cultivation did not worsen.

Studying the interaction in the plant – soil –micro-organisms chain, Juhos K., Papdi E., Kovács F. et al. (2023) when growing bell peppers under laboratory conditions, it was found that the effect of sheep's wool content in various soils (sand, loam, peat) on biomass yields was significantly higher with less irrigation than when using agrofibre and straw. The microbiological activity of the soil was also higher than the introduction of wool, due to its better water retention capacity, which positively affected the yield of pepper.

Similar studies on pepper cultivation (Capsicum annuum L.) was also performed on soils of different composition – Sandy with a low content of organic substances and coarse loams with a high moisture capacity, Papdi E., Veres A., Kovács F., Juhos K. (2022). They found that all irrigated areas where wool mulch was used were characterized

by a higher carbon content in the soil, and loamy areas – and beta-glucosidase activity. The use of wool mulch contributed to the formation of a higher yield.

To study the characteristics of tree growth and development, as well as the yield and quality of gitea m plum fruits.A., Borza I.M., Domuta C.G. et al. (2024) conducted an experiment on the use of sheep wool mulch and conditioner for soil containing corn starch. They found an increase against the control of plum yield by 27% in areas where wool mulch was used and by 37% where a combination of wool and soil conditioner was used, against the background of an increase to 48.9% by weight and from 5 to 19% by fruit size.

As an alternative to vegetable mats made from coconut fibers, Herfort S., Pflanz K., Larsen M.-S. et al. (2023) evaluated similar products with the inclusion of different amounts of sheep wool in their composition when growing perennials for garden and landscape design. Their studies revealed a 21-38-fold higher content of nitrogen in the products and 1.5 - 1.8 – fold higher content of water. Perennial plants that were grown using combined coconut-wool mats were characterized by better development and yield.

In addition to the use of wool as a mulch for the soil in the cultivation of agricultural crops, it also finds its application in forestry.

The effectiveness of using a repellent made from sheep's wool to protect young forest stands from damage to the tops of their shoots by deer was tested in Bernacka H studies., Święcicka, N., Naworska N. (2015). In forest stands that were not covered with wool repellent, the area of damage caused by deer increased more than 4 times over three years of the experiment. In this regard, the researchers concluded that it is advisable to use sheep's wool to protect forest stands, as an alternative to their treatment with harmful chemicals.

Another area of application of sheep wool, according to Szatkowski P., Tadla A., Flis Z. et al. (2022), is the production of thermally insulated, eco-friendly packaging, which is characterized by complete biological theft. The experimental packaging produced by them by injection molding was not inferior in thermal insulation characteristics to its expanded polystyrene analogues, and the introduction of wool fibers accelerated the process of its biodegradation in laboratory-modeled natural conditions.

Discussion. Analysis of the state of wool production and the dynamics of purchase prices for it in the world and in Ukraine indicates a decrease in both the total volume of its production and demand for it. This is one of the reasons for finding alternative production areas for this natural, renewable product than the textile industry. Research by many scientists also shows increased attention to the problem associated with the use of sheep's wool waste to solve environmental problems (Torring T., Sandvik K.,2000; Parlato M. C.M. and Porto S.M.C. , 2020; Parlato M.C.M., Porto S.M.C., Valenti F., 2022; Juhos K., Papdi E., Kovács F. et al., 2023; Midolo G., Valenti F., 2024).

According to a number of publications, there is an increase in research aimed at evaluating the effectiveness of using sheep's wool in the manufacture of new building materials – heat and noise insulation, plaster, brick, concrete (El Wardi F.Z., Ladouy S., Khabbazi A. et al., 2021; Alyousef R., Alabduljabbar H., Mohammadhosseini H. et al., 2020 and Alyousef R. , Mohammadhosseini H., Ebid A.A.K., Alabduljabbar H., 2022) , as well as studies related to the use of wool in agricultural technologies and forestry in the cultivation of various crops (Camilli F., Focacci M., Dal Prà A. et al., 2025).

At the same time, Corscadden, K. W., Biggs, J. N., & Stiles, D. K. (2014) note that the choice of materials in production has the nature of trade-offs between their characteristics, properties, Environmental Impact, Sustainability, availability, and cost-effectiveness. The desire to integrate more bio-products and natural and renewable resources primarily in the construction industry is now associated with sheep wool.

This approach, according to Jannat N., Hussien A., Abdullah B. (2020), reduces energy consumption in construction and other industries, and promotes efficient waste management.

Currently, major projects are already being implemented to create industrial enterprises aimed at solving environmental problems related to the use of wool. So, according to Al-Malah K.M., Al-Khalafat M.R., Al-Zayadeen N.A. et al. (2025) the construction of a processing plant in Jordan for more than 6.5 thousand tons of wool per year (production of 114.8 thousand m3 of insulation boards) is a profitable project with a return on investment within one year.

Conclusions.

1. sheep wool, in addition to its traditional use in textile production, is finding an increasingly wide alternative application in various production areas.

2. due to its unique physical and technical properties and the ability to biodergadate, its use is widespread in the construction and agricultural sectors, in the production of packaging materials, and in the manufacture of products using 3 D technologies.

3. for many countries of the world and Ukraine, the development of modern technologies and assessment of the economic and environmental efficiency of alternative applications of sheep wool in the field of manufacturing new materials, construction, agricultural production, forestry is a promising urgent task.

References

Abdul Awal A.S.M., Mohammadhosseini H. (2016). Green concrete production incorporating waste carpet fiber and palm oil fuel ash. *Journal of Cleaner Production.* Vol. 137, P.157-166. https://doi.org/10.1016/j.jclepro.2016.06.162

Ahmed A., Qayoum A., Qayoom Mir F. (2019) . Investigation of the thermal behavior of the natural insulation materials for low temperature regions. *Journal of Building Engineering.* Vol. 26, 100849 https://doi.org/10.1016/j.jobe.2019.100849

Al-Malah K.M., AlKhalafat M.R., Al-Zayadeen N.A., Al-Mfalfal A.S., Alma'atah B.M. and Alkhamis T.M. (2025). The Feasibility of a Proposed Plant Design of Sheep Wool Insulation Material in Jordan to Eliminate the Negative Environmental Impact of Wasted Sheep Wool. *Journal of Environmental Protection*, 16, 130-147. https://doi.org/10.4236/jep.2025.162007

Alyousef R., Alabduljabbar H., Mohammadhosseini H. , Mohamed A.M, Siddika A., Alrshoudi F., Alaskar A. (2020). Utilization of sheep wool as potential fibrous materials in the production of concrete composites. *Journal of Building Engineering*. Vol. 30, 101216. https://doi.org/10.1016/j.jobe.2020.101216

Alyousef R., Mohammadhosseini H., Ebid A.A.K, Alabduljabbar H. (2022). An Integrated Approach to Using Sheep Wool as a Fibrous Material for Enhancing Strength and Transport Properties of Concrete Composites. Materials (Basel). 15(5):1638. https://doi.org/10.3390/ma15051638

Atbir A., Taibi M., Aouan, B., Khabbazi A., Ansari O., Cherkaoui M., Cherradi T. (2023). Physicochemical and thermomechanical performances study for Timahdite sheep wool fibers application in the building's insulation. *Scientific Reports* , 13, 5038 https://doi.org/10.1038/s41598-023-31516-9

Atbir A., Khabbazi A., Cherkaoui M.,Ibaaz K., El Wardi F.Z., Chebli S. (2023). Improvement of thermomechanical properties of porous plaster reinforced with a

network of morocco sheep wool skeletons for energy efficiency. *building and environment.* vol. 234, 110171 https://doi.org/10.1016/j.buildenv.2023.110171

Berge B. The Ecology of Building Materials; Architectural Press Linacre House: Oxford, UK, 2001. https://api.pageplace.de/preview/DT0400.9781136434624_A23846429/preview-9781136434624_A23846429.pdf [from 21.03.2025]

Bernacka, H., Święcicka, N., Naworska, N. (2015). Application of sheep wool in preventing damage caused by deer in young forest plantations. Acta Sci. Pol. Zootechnica, 14(4), 5–14. https://asp.zut.edu.pl/pdf/asp-2015-14-4-335.pdf [from 21.03.2025]

Borlea (Mureşan) S. I., Tiuc A.-E., Nemeş O., Vermeşan H., Vasile O. (2020). Innovative Use of Sheep Wool for Obtaining Materials with Improved Sound-Absorbing Properties. *Materials***,** 13(3), 694 https://doi.org/10.3390/ma13030694

Broda, J., Gawlowski, A., Rom, M., & Kobiela-Mendrek, K. (2023). Utilisation of waste wool from mountain sheep as fertiliser in winter wheat cultivation. *Journal of Natural Fibers*, 20(2),2200047. https://doi.org/10.1080/15440478.2023.2200047

Broda J., Gawłowski A., Rom M., Kukulski T., Kobiela-Mendrek K. (2024). Thermoregulation and Soil Moisture Management in Strawberry Cultivation Mulched with Sheep Wool**.** *Applied Sciences***,** *14*(23),10884; https://doi.org/10.3390/app142310884

Camilli F., Focacci M., Dal Prà A., Bortolu S., Ugolini F., Vagnoni E. and Duce P.(2025). Turning waste wool into a circular resource: a review of eco-innovative applications in agriculture. *Agronomy***,** *15*(2), 446; https://doi.org/10.3390/agronomy15020446

Corscadden K. W., Biggs J. N., & Stiles D. K. (2014). Sheep's wool insulation: A sustainable alternative use for a renewable resource?. *Resources, Conservation and Recycling*, *86*, 9-15. https://doi.org/10.1016/j.resconrec.2014.01.004

Dénes O., Florea I., Manea D.L. (2019). Utilization of Sheep Wool as a Building Material. *Procedia Manufacturing*.Vol. 32. 236-241. https://doi.org/10.1016/j.promfg.2019.02.208

Directive 2010/31/EU of the European Parliament and of the Council of 19 May 2010 on the Energy Performance of Buildings. Available online: http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri= CELEX: 32010L0031 &from=EN

Doroudiani S., Omidian H. (2010). Environmental, health and safety concerns of decorative mouldings made of expanded polystyrene in buildings. Build. *Environ.* 45, 647–654. https://doi.org/10.1016/j.buildenv.2009.08.004

El Wardi F.Z., Ladouy S., Khabbazi A., Ibaaz K., Asmae Khaldoun A.(2021). Unfired Clay-Cork Granules Bricks Reinforced with Natural Stabilizers: Thermomechanical Characteristics Assessment. *Civil Engineering Journal*. Vol. 7, No. 12. 2068-2082. http://dx.doi.org/10.28991/cej-2021-03091778

Fantilli A.P., Sicardi S., Dotti F. (2015). The use of wool as fibre-reinforcement in cement-based mortar. In: Proceedings of the 1st International Conference on Bio-based Building Materials, Clermont-Ferrand, France, ICBBM; 2015, pp. 341-346. file:///C:/Users/user/Downloads/1693-RUGC%20Manuscript-6039-1-10-20200505%20(2).pdf [from 21.03.2025]

Gabryś T., Fryczkowska B. (2022). Using Sheep's Wool as an Additive to the Growing Medium and its Impact on Plant Development on the Example of Chlorophytum comosum. *Journal of Ecological Engineering* 2022, 23(6), 205–212. https://doi.org/10.12911/22998993/148220

Galán-Marín C., Rivera-Gómez C., Petric-Gray J. (2010). Effect of animal fibres reinforcement on stabilized earth mechanical properties. *Journal Of Biobased Materials And Bioenergy*, vol. 4, n 2, pp. 121-128. https://doi.org/10.1166/jbmb.2010.1076

Gitea M.A., Borza I.M., Domuta C.G., Gitea D., Rosan C.A., Vicas S.I., Pasca M.B. (2024). A Sustainable Approach Based on Sheep Wool Mulch and Soil Conditioner for Prunus domestica (Stanley Variety) Trees Aimed at Increasing Fruit Quality and Productivity in Drought Conditions. *Sustainability***,** *16*(17), 7287; https://doi.org/10.3390/su16177287

Herfort S., Pflanz K., Larsen M.-S., Mertschun T. and Grüneberg H. (2023). Influence of Sheep's Wool Vegetation Mats on the Plant Growth of Perennials. *Horticulturae* 2023, ***9***(3), 384; https://doi.org/10.3390/horticulturae9030384

Jannat N., Hussien A., Abdullah B., Cotgrave A. (2020) Application of agro and non-agro waste materials for unfired earth blocks construction: A review. Construction and Building Materials**.** Vol. 254**,** 119346. https://doi.org/10.1016/j.conbuildmat.2020.119346

Juhos K., Papdi E., Kovács F., Vasileiadis V. P., Veres A. (2023).The Effect of Wool Mulch on Plant Development in the Context of the Physical and Biological Conditions in Soil. *Plants***,** *12*(3), 684; https://doi.org/10.3390/plants12030684

Kicińska-Jakubowska A., Broda J., Zimniewska M., Bączek M., Mańkowski J. (2023).Effect of Blend Composition on Barrier Properties of Insulating Mats Produced from Local Wool and Waste Bast Fibres. *Materials.*16(1), 459; https://doi.org/10.3390/ma16010459

Korjenic A., Klarić S., Hadžić A. and Korjenic S. (2015). Sheep Wool as a Construction Material for Energy Efficiency Improvement. *Energies*, 8(6), 5765-5781; https://doi.org/10.3390/en8065765

Korjenic A., Petránek V., Zach J., & Hroudová J. (2011). Development and performance evaluation of natural thermal-insulation materials composed of renewable resources. *Energy and Buildings*, 43(9), 2518-2523. https://doi.org/10.1016/j.enbuild.2011.06.012

Livestock and crop products. FAOSTAT, 2023 https://www.fao.org/faostat/ru/#data/QCL [цитовано 21.03.2025]

Midolo G., Valenti F. (2024). Sheep Wool Waste Availability for Potential Sustainable Re-Use and Valorization: A GIS-Based Model. *Agriculture* 14(6):872 https://doi.org/10.3390/agriculture14060872

Papdi E., Veres A., Kovács F., Juhos K. (2022). How different mulch materials regulate soil moisture and microbiological activity? *Journal of Central European Green Innovation Talajbiológia különszám*, 26–38. https://doi.org/10.33038/jcegi.3560

Parlato M.C.M., Cuomo M., Porto S.M.C.(2022). Natural fibers reinforcement for earthen building components: Mechanical performances of a low quality sheep wool ("Valle del Belice" sheep). *Construction and Building Materials*. Vol. 326, (4), 126855 https://doi.org/10.1016/j.conbuildmat.2022.126855

Parlato M.C.M. and Porto S. M.C. (2020). Organized Framework of Main Possible Applications of Sheep Wool Fibers in Building Components. *Sustainability* 2020, *12*(3), 761; https://doi.org/10.3390/su12030761

Pederneiras C.M., Veiga R. and Brito J. (2019). Rendering Mortars Reinforced with Natural Sheep's Wool Fibers. *Materials* 2019, *12*(22), 3648; https://doi.org/10.3390/ma12223648

Starkova O., Sabalina A., Voikiva V. and Osite A. (2022). Environmental Effects on Strength and Failure Strain Distributions of Sheep Wool Fibers. *Polymers***,** *14*(13), 2651; https://doi.org/10.3390/polym14132651

Szatkowski P., Tadla A., Flis Z., Szatkowska M., Suchorowiec K., Molik E. (2022). Production of biodegradable packaging with sheep wool fibres for medical applications and assessment of the biodegradation process. Animal science and genetics/ Published by the Polish Society of Animal Production, vol. 18, no 3, 57-67 https://doi.org/10.5604/01.3001.0016.0365

Taskin M. B.(2024). Reducing Mineral Fertilizer Usage: Utilizing Sheep Wool and Alkaline Hydrolysate for Enhanced Sugar Beet Cultivation. *Sugar Tech*, 26(6):1653- 1664. https://doi.org/10.1007/s12355-024-01426-9

Torring T., Sandvik K. Management of demolition waste-using life cycle assessment methodologies. In Proceedings of the International Symposium of Integrated Life-Cycle Design of Materials and Structures, Helsinki, Finland, 22–24 May 2000; pp. 522–526. https://doi.org/10.1617/235158029X.096

Urdanpilleta M., Leceta I., Martín-Garín A., Millán-García J.A., Guerrero P., Caba K. (2025). Valorized sheep wool biocomposites towards a more sustainable building sector: Thermal insulation, sound absorption, and resistance against insects. *Developments in the Built Environment*, Vol. 21, 100608 https://doi.org/10.1016/j.dibe.2025.100608

Vasina M., Straznicky P., Hrbacek P., Rusnakova S., Bosak O., and Kubliha M. (2024). Investigation of Physical Properties of Polymer Composites Filled with Sheep Wool. *Polymers***,** 16(5), 690; https://doi.org/10.3390/polym16050690

Zach J., Korjenic A., Petránek V., Hroudova J., Bednar T. (2012). Performance evaluation and research of alternative thermal insulations based on sheep wool. *Energy Build*. 49, 246–253. https://doi.org/10.1016/j.enbuild.2012.02.014

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STRESS RESISTANCE OF PIGGLES OF DANISH ORIGIN AND THEIR PRODUCTIVITY

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The article presents the results of scientific research on determining the level of stress resistance in piglets of Danish origin, weaned from sows at the age of 28 days. According to the results of the variation series of the live weight test of piglets, the modal class included the II group of weakly responsive animals, which had a stress resistance criterion within ±0.67 sigma, relative to the individual arithmetic mean values of the sample for this indicator; the III group of highly responsive animals - by -0.67 sigma in the direction of decrease and the I group of resistant animals - by +0.67 sigma in the direction of increase. The obtained experimental data indicate that within the normalized distribution of the total livestock, the maximum number of young animals was found to be weakly responsive (group II) to the stress factor (51 animals or 56.7%), the resistant type (group I) is characteristic of a group of animals of 28 animals or 31.1%, while 11 animals or 12.2% were included in the group of highly responsive piglets (group III).

Analyzing the results of the studies, it can be stated that the growth energy of young animals of group M^+ for the period from weaning from mothers to the end of the growing-up period was quite high, compared to peers of groups M^0 and M^- by 62.0 g or 13.9% ($p<0.05$) and 87 g or 20.6% ($p<0.01$). At the age of 6 months, the growth rate of the young animals of the M^+ group began to increase again and they exceeded the piglets from the M^0 group by 44.0 g or 5.6% ($p<0.05$) and individuals from the M^- group by 81.0 g or 10.7% ($p<0.01$). The improvement of the adaptive capacity to the stress factor in the M^+ group of piglets was also accompanied by a significant increase in the average daily gain over the entire growing period by 39.0 g or 6.3% ($p<0.05$) and 67.0 g or 11.3% ($p<0.01$) compared to the young animals from the other groups.

Subsequently, the evaluated young animals were distributed by types of higher nervous activity. The highest level of strength of nervous processes, balance of excitation and inhibition processes, and their mobility were observed in piglets that had a strong balanced mobile type of nervous activity with a high statistical difference of $p<0.001$ in all cases of comparison with the group identified as weak.

Keywords: stress resistance, growth, piglets, weaning, productivity.

СТРЕСОСТІЙКІСТЬ ПОРОСЯТ ДАНСЬКОГО ПОХОДЖЕННЯ ТА ЇХ ПРОДУКТИВНІСТЬ

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У статті викладено результати наукових досліджень з визначення рівня стресостійкості у поросят данського походження, відлучених від свиноматок у віці 28 діб. За результатами даних варіаційних рядів залікового тестування живої маси поросят до модального класу віднесли ІІ групу слабореагуючих тварин, що мали критерій стресостійкості в межах ±0,67 сигми, відносно індивідуальних середніх арифметичних значень вибірки за цим показником; до ІІІ групи сильнореагуючих – на -0,67 сигми у бік зменшення і до І групи стійких – на +0,67 сигми у бік збільшення. Одержані дані експерименту свідчать про те, що в межах нормованого розподілу загального поголів'я, максимальну кількість молодняку виявлено слабореагуючим (ІІ група) на дію стрес-чинника (51 голову або 56,7 %), стійкий тип (І група) властивий групі тварин із 28 голів або 31,1 %, тоді як до групи сильнореагуючих поросят (ІІІ групи) зарахували 11 голів або 12,2 %.

Аналізуючи результати досліджень можна констатувати, що енергія росту молодняку групи M^+ за період від відлучення від матерів і до кінця періоду дорощування виявилася достатньо високою, порівняно з ровесниками груп M^0 і M^- на 62,0 г або 13,9 % ($p<0,05$) та 87 г або 20,6 % ($p<0,01$). У віці 6 місяців інтенсивність росту молодняку групи M^+ розпочала знову збільшуватися і вони переважали поросят із групи M^0 вже на 44,0 г або 5,6 % ($p<0,05$) та особин із групи M^- – на 81,0 г або 10,7 % ($p<0,01$). Покращення адаптаційної здатності до стрес-чинника в групи поросят M^+ супроводжувалося й вірогідним збільшенням величин середньодобового приросту в цілому за період вирощування на 39,0 г або 6,3 % ($p<0,05$) і 67,0 г або 11,3 % ($p<0,01$) щодо молодняку з решти груп.

У подальшому оцінений молодняк розподілили за типами вищої нервової діяльності. Найвищий рівень сили нервових процесів, врівноваженість процесів збудження і гальмування, їх рухливість відмічався у поросят, які мали сильний врівноважений рухливий тип нервової діяльності за високої статистичної різниці $p<0,001$ в усіх випадках порівняння до групи, виділених як слабких.

Ключові слова: стресостійкість, приріст, поросята, відлучення, продуктивність.

Introduction. Before the start of the full-scale war in Ukraine, the breeding of new specialized breeds of pigs of foreign selection, which are characterized by increased metabolism in the body, high fattening and meat qualities, which are realized under certain technologies of keeping and feeding systems of animals (Hao Y. et al., 2021; Ramirez B.C. et al., 2022; Chaly O.I. et al., 2023). Extensive work has been carried out on the development of methodological principles for their assessment and further

improvement (Karpovsky V.I. et al., 2012). However, as practice has proven, animals with high genetic potential for productivity are the most sensitive to the effects of negative external stressors (O'Connor E.A. et al., 2010; Čobanović N. et al., 2020). The response of the pig body to stress is reflected in productivity losses (Munsterhjelm C. et al., 2010; Campbell J. et al., 2013; Sutherland M.A. et al., 2014; Bankole T. et al., 2024), reduced reproductive capacity (Mayorga E.J. et al., 2020), weakened natural resistance (Stovbetska L. et al., 2021), behavioral disorders (Gonzalez-Rivas P.A. et al., 2020), deterioration of product quality indicators (Gonzalez-Rivas P.A. et al., 2020; Serviento A.M. et al., 2020), leads to a significant increase in livestock waste (Poroshynska O.A. et al., 2020), and, as a result, these farms suffer significant economic losses.

In addition to the main technological stresses in pig farming, military operations can be a powerful stress factor in certain regions of Ukraine in recent years. The negative consequences of war-induced stress have been repeatedly exacerbated in farms located close to the epicenter of active hostilities and engaged in raising young animals - one of the most vulnerable and less protected groups in industrial pork production technology, especially during the weaning period, which is reflected, first of all, in the intensity of their growth (White H.M. et al., 2008; Dokmanovic M. et al., 2017). In this context, the assessment of stress provoked by weaning piglets from sows comes to the fore (da Fonseca de Oliveira A.C. et al., 2019; Lange A. et al., 2020). It is worth noting that during the "weaning crisis" period, piglets experience uneven growth rates, which is directly related to the impact of other technological stress factors and the corresponding reaction of animals to them (Skaperda Z. Et 2019; Vyslotska L. et al., 2021). Of course, when growing young animals, more and more attention should be paid to the selection of stress-resistant animals in order to obtain competitive products and constantly develop new methods and improve production technologies to reduce the harmful effects of stress on animals. However, these issues require thorough scientific generalization. Therefore, the unresolved nature of a number of problems, the urgency of which has increased in the conditions of modern threats, determined the relevance and decisively influenced the relevance of the work carried out.

The purpose of the research is to determine the level of stress resistance and productivity of piglets of Danish origin under the influence of the stress factor "weaning crisis".

Materials and methods. The scientific and economic experiment was conducted in the production conditions of the PP AF "Svitanok" of the Novovodolazha district of the Kharkiv region. Stress resistance was determined using the method of V.A. Kovalenko et al., improved by O. M. Tsereniuk during the "weaning crisis" period, which lasted 15 days. The total duration of the experiment was 182 days.

To organize the experiment, a technological group of 90 heads was selected from the total population of suckling piglets. The principle of normalized deviation was used to distribute the piglets into experimental groups. The young were weaned at the age of 28 days. The piglets were weighed individually: at weaning, 4, 9 and 15 days after it, then the stress resistance criterion was calculated based on the sum of the differences in the absolute values of live weight in these reference periods and the livestock was divided into groups.

As a result of processing the data of variation series of the test of absolute live weight gains of piglets 15 days after weaning them from sows, the modal distribution class was assigned to group II of weakly reacting animals (M^0), which had a stress resistance criterion within ±0.67 sigma; to group III of strongly reacting animals (minus variant M^-) – by –0.67 sigma in the direction of decrease, and to group I of resistant animals (plus variant M^+) – by +0.67 sigma in the direction of increase.

Animals of all experimental groups were kept in the same room according to generally accepted technology. The conditions of care for piglets were the same and met sanitary and zootechnical requirements. Production premises were vacated each time in the case of regular sanitary treatment and the need to fill the next technological cycle with young animals, while adhering to the principle of "everything is empty, everything is occupied".

The feeding rations of the experimental young animals were prepared in accordance with detailed norms and adjusted for the growing periods, taking into account the age, growth intensity, chemical composition and nutritional value of the feed. Group feeding, twice a day: morning and evening, drinking - ad libitum, with free access to drinkers. For feeding, standard compound feeds were used, manufactured in accordance with a specific production period. Compound feeds for experimental animals were fed in the form of dry feed.

Live weight was determined by the results of individual weighing of animals in the morning before feeding at weaning, at the age of 4; 9; 15; 61; 122 and 182 days, with a measurement accuracy of ±0.1 kg. Based on the weighings, the total gain for the period and the average daily gain in live weight were calculated.

As an additional test for assessing the stress resistance of piglets, the parameters of the higher nervous system were determined based on the results of the analysis of the processes of formation, excitation, inhibition of motor-food conditioned reflexes when changing the stereotype of feeding circumstances (using powdered milk-enriched compound feed) and the speed of developing an orienting reaction (Karpovsky, V.I., 2012). The reaction of young animals to changes in technological conditions was expressed in conventional units - from one to four. The study was conducted over five consecutive days, with a time expenditure of 20 minutes for each animal. For a more objective distribution of experimental animals into groups by type of higher nervous activity, in the same individuals it was determined once more with an interval of 2 days after the main testing under similar conditions of the experiment. After comparing the ethological observation data obtained in this way, the young animals were assigned to one of four groups, according to the types of higher nervous activity. The first group included piglets with a strong balanced mobile type of higher nervous activity, the second - strong balanced inert, the third - strong unbalanced and the fourth group - with a weak type.

The obtained results of experimental studies were processed biometrically, by methods of variational statistics using electronic spreadsheets of a Pentium/4 personal computer and a package of modern applied licensed programs Statistika, SPSS 15.0, MS Excel spreadsheets, 2003. The difference between the values of the compared indicators of the experimental groups was considered probable when achieving three levels of probability *$p<0.05$; **$p<0.01$; ***$p<0.001$.

Research results: When evaluating the results of the individual distribution of the experimental livestock according to the stress resistance criterion, it is worth pointing out its significant variability depending on the group (Table 1).

In particular, within the normalized distribution of the total livestock, the maximum number of young animals was found to be weakly responsive (M^0) to the stress factor (51 animals or 56.7%), the resistant type (M^+) was characteristic of a group of animals consisting of 28 animals or 31.1%, while 11 animals or 12.2% were included in the group of highly responsive piglets (M^-).

Table 1

Distribution of experimental young animals by stress resistance

Group	Distribution of piglets by stress resistance criterion	
	heads	%
I – M^+ (resistant)	28	31,1
II – M^0 (weakly reactive)	51	56,7
III – M^- (strongly reactive)	11	12,2
Total	90	100,0

Young animals of Danish origin with different stress tolerance showed quite high fattening rates during the first six months of rearing, however, in certain age periods, the variability of their values between groups was directly due to the unequal rate of response to weaning from mothers (Table 2).

Table 2

Changes in live weight of young animals of different types of stress resistance, kg ($\bar{x} \pm S_{\bar{x}}$)

Indicator	Group		
	I – M^+ (resistant)	II – M^0 (weakly reactive)	III – M^- (strongly reactive)
Live weight of piglets, kg:			
at weaning	7,84±0,32	7,56±0,18	7,80±0,38
on the 4th day after weaning	8,62±0,52	7,84±0,22	7,67±0,41
on the 9th day after weaning	9,93±0,55*/*	8,37±0,34	8,21±0,65
on the 15th day after weaning	12,52±0,63*/**	9,72±1,14	8,58±0,94
at the age 2 months	24,11±0,72*/**	21,86±0,57	21,34±0,69
at the age 4 months	58,72±1,28*/**	55,24±0,91	53,41±1,46
at the age 6 months	109,74±2,51*/**	103,55±2,12*	99,49±1,54

*Note. *p<0.05; **p<0.01 – probability of difference in relation to the distribution group M^0 and M^-*

As expected, piglets that were assigned to the M^+ group were heavier than their peers from the M^0 and M^- groups. In particular, on the fourth day after weaning, their advantage over the latter was 9.9 and 12.4%. A characteristic feature of changes in live weight during this period is its increase in young pigs M^+, while in the group of piglets M^- the studied indicator even decreased. At the same time, quantitative statistically significant differences in live weight between young pigs of these groups began to appear from the 9th day after weaning by 18.6% ($p<0.05$) and 21.0% ($p<0.05$), respectively, reaching maximum values at the age of 15 days - by 28.8% ($p<0.05$) and 45.9% ($p<0.01$) and 13.3% in favor of young pigs resistant to stress. It is noteworthy that at the age of 6 months the difference between the groups significantly decreased and amounted to only 6.0% ($p<0.05$) and 10.3% ($p<0.01$). The decrease in live weight in animals M^-, compared with peers from other groups, is a natural result of their weaker adaptation to the stress factor.

Similar advantages in terms of live weight are also characteristic of young animals assigned to the M^0 group compared to peers from the M^- group. In particular, the average indicator of their live weight on the 15th day after weaning from their mothers exceeded

the latter by 13.3%, while in the subsequent months of growing the difference between these groups decreased to 4.1% at the age of 6 months, although it was statistically significant ($p<0.05$).

The absolute live weight gains of piglets, taking into account the distribution group by stress resistance during rearing, also differed: from 15 days after weaning and until reaching 2 months of age, the live weight gain of animals in the M^+ group was 11.6 kg or 1.9 times, in the M^0 group - 21.1 kg or 2.2 times, and in the $M^–$ group - only 12.8 kg or 2.5 times. In general, from weaning to 6 months of age, the total weight gain of young animals assigned to the M^+ group increased by 101.9 kg or 14.0 times, in the M^0 group - by 96.0 kg or 13.7 times, and in the $M^–$ group - by 91.7 kg or 12.8 times.

Weaning of piglets from mothers also affected the growth energy of young animals. In terms of average daily live weight gains in piglets of all groups, there is a natural age-related increase in the period from weaning to reaching the age of 6 months. It is noteworthy that the general trend regarding the nature of their formation depending on the distribution group and live weight parameters of young animals is preserved (Table 3).

Table 3

Dynamics of average daily live weight gains of young animals of different stress resistance, ($\bar{x} \pm S_{\bar{x}}$)

Indicator	Group		
	I – M^+ (resistant)	**II – M^0 (weakly responsive)**	**III – $M^–$ (strongly responsive)**
Average daily gain for the period, g:			
from weaning to 2 months of age	509±14,24*/***	447±19,61	422±20,08
from 2 to 4 months of age	567±15,95*	547±12,27	526±11,34
from 4 to 6 months of age	836±18,79*/**	792±9,68*	755±14,82
from weaning to 6 months of age	662±13,53*/**	623±10,37	595±14,76
Feed consumption per 1 kg of live weight gain for 6 months of the experiment, EKO	3,93	4,67	4,84

*Note. *p<0.05; **p<0.01; ***p<0.001 – probability of difference in relation to the distribution group M^0 and $M^–$*

Considering the results presented in the table, it can be stated that the growth energy of the young animals of the M^+ group for the period from weaning from mothers to the end of the growing-up period was quite high, compared with the peers of the M^0 and $M^–$ groups by 62.0 g or 13.9% ($p<0.05$) and 87 g or 20.6% ($p<0.01$). Therefore, the effect of stress on the piglets' organism during this period is maximum. In the period from 2 to 4 months of age, the average daily gains in piglets of all groups further increase, but the pace of this process slows down to minimum values and the difference between the young animals of the M^+ and M^0 groups has only the character of a tendency to increase and is 20.0 g or 3.7%, while between the piglets of the M^+ and $M^–$ groups it is at the level of 41 g or 7.8% and acquires the lowest level of statistical significance ($p<0.05$). At the age of 6 months, the growth rate of the young animals of the M^+ group began to increase again and they exceeded the piglets from the M^0 group by 44.0 g or 5.6% ($p<0.05$) and individuals from the $M^–$ group by 81.0 g or 10.7% ($p<0.01$). The improvement of the adaptive capacity to the stress factor in the M^+ group of piglets was also accompanied by

a significant increase in the average daily gain over the entire growing period by 39.0 g or 6.3% ($p<0.05$) and 67.0 g or 11.3% ($p<0.01$) compared to the young animals from the other groups.

Piglets of group M^0, although inferior to their peers assigned to group M^+ during the growing period, also differed from their peers of group M^- in terms of average daily live weight gains: at 2 months – by 25.0 g or 5.9%, at 4 months – by 21.0 g or 4.0%, at 6 months – by 37.0 g or 4.9% ($p<0.05$) and during the growing period – by 28.0 g or 4.7%.

Against the background of the assessment of young animals of different types of stress resistance, a clear manifestation of sexual dimorphism was established, and, as a result, boars, regardless of their normalized distribution into groups, naturally exceeded gilts in terms of growth intensity from weaning to 6 months of age by an average of 5.8%.

Most domestic and foreign experts believe that the level of stress resistance of pigs kept in large industrial complexes directly depends on the typological features of higher nervous activity. That is, they must be resistant to technological stresses, have a quick and adequate reaction to external stimuli, and belong to a strong balanced mobile type of higher nervous activity. Such animals are distinguished by higher growth intensity and digestibility coefficients of feed nutrients, they have a faster process of forming meat productivity and improving product quality.

For an in-depth justification of this position, we determined the typological features of higher nervous activity in piglets of different types of stress resistance when transferring them to the finishing shop. Piglets, which are characterized by a strong balanced mobile type of higher nervous activity, during the observation period behaved confidently, calmly moved along the passage of the room to the feeder, immediately found it and from the first or second attempt began to consume compound feed without paying attention to the experimenter, almost did not react to the conditioned stimulus and did not show aggression. Young pigs with a strong balanced inert type were somewhat more cautious, but also calmly moved along the passage of the room, approached the feeder later (from the second to the fourth attempt), consumed compound feed willingly (from the first or second attempt), also almost did not react to the conditioned stimulus and the experimenter and did not show aggression. Animals with a strong unbalanced type behaved completely differently: they moved quickly along the passage to the feeder, carefully watching the experimenter, consumed the compound feed excitedly and reluctantly immediately approaching the feeder or on the third attempt, some individuals pushed or gnawed it during feeding, reacted strongly to the conditioned stimulus with a shudder. Piglets with a weak type moved slowly and timidly along the passage of the room with a characteristic sound of danger or stopped altogether, some of them had to be urged, they approached the feeder on the fourth to sixth attempt, consumed the compound feed cautiously, rather reluctantly and not immediately (on the third to fifth attempt), carefully watched the experimenter, reacted quite excitedly to the conditioned stimulus, refusing the feed and moving away from the feeder, sometimes urination or defecation was noted in them.

Based on the assessment, piglets of different stress resistance were distributed by types of higher nervous activity in the following ratio (Table 4).

The results of visual observations show that 34 heads (37.8%) of the total number of piglets have a strong balanced mobile type, 28 heads (31.1%) - strong balanced inert, 9 heads (10.0%) - strong unbalanced and 19 heads (21.1%) - weak.

Table 4

Distribution of experimental young animals by types of higher nervous activity

Type of higher nervous activity	Distribution of experimental young animals by types of higher nervous activity	
	heads	%
Strong balanced mobile	34	37,8
Strong balanced inert	28	31,1
Strong unbalanced	9	10,0
Weak	19	21,1
Total	90	100,0

As part of the study of conditioned reflex activity, the following were directly taken into account: the strength of nervous processes, the balance of excitation and inhibition processes, the mobility of nervous processes (Table 5).

Table 5

Typological parameters of higher nervous activity of young animals, mind. units, ($\overline{X} \pm S_{\bar{x}}$)

Type of higher nervous activity	Level of nervous processes		
	strength	balance	mobility
Strong balanced mobile	3,76±0,17***	3,88±0,09***	3,93±0,15***
Strong balanced inert	3,11±0,08***	2,92±0,06***	2,57±0,19***
Strong unbalanced	1,84±0,12*	1,68±0,14	1,82±0,28*
Weak	1,34±0,20	1,49±0,13	1,09±0,16

*Note. *p<0.05; ***p<0.001 – probability of difference in relation to the group of weak young animals*

Analysis of the features of individual assessment of young animals indicates a different norm of their reaction to weaning from sows. The highest level of strength of nervous processes, balance of excitation and inhibition processes, their mobility was observed in piglets that had a strong balanced mobile type of nervous activity with a high statistical difference p<0.001 in all cases of comparison to the group identified as weak and amounted to an average of 3.86 conventional units.

In animals with a strong balanced inert type, the values of the studied indicators are higher, respectively by 1.77; 1.43 and 1.48 conventional units against young animals with a weak type of higher nervous activity, with the same high level of statistical significance (p<0.001 in all cases of comparison). Piglets with strong unbalanced and weak types of nervous activity were characterized by almost the same value of these indicators. However, in the group of young animals classified as strong unbalanced type, they were higher by 0.50 (p<0.05); 0.19 and 0.73 (p<0.05) conventional units. In the group of animals with weak type of nervous activity, the values of the studied indicators were minimal and equaled on average 1.31 conventional units.

At the same time, young animals with strong balanced mobile type of higher nervous activity are characterized by increased mobility and balance and reduced strength of nervous processes. Piglets with strong balanced inert type, on the contrary, were characterized by higher strength of nervous processes and their balance, but lower their mobility. In representatives of the strong unbalanced type, the processes of balance of excitation and inhibition were weaker, while their strength and mobility were higher.

Nervous processes in piglets with a weak type have the least mobility, and the balance of excitation and inhibition processes prevails over their strength. Therefore, as the reactivity of piglets decreases, both the strength of nervous processes, the balance of excitation and inhibition processes, and their mobility naturally decrease.

The strength of the stress factor "weaning crisis" depended, first of all, on the closeness of the connection between stress resistance and types of higher nervous activity of piglets, determined under the influence of another stress factor - transfer to the growing-up shop (Table 6).

Table 6

The relationship between types of stress resistance and higher nervous activity in the experimental young, %

Group	Type of higher nervous activity							
	strong balanced mobile		strong, balanced inert		strong, unbalanced		weak	
	heads	%	heads	%	heads	%	heads	%
I – M^+ (stable)	19	67,9	9	32,1	–	–	–	–
II – M^0 (weakly reactive)	15	29,4	19	37,3	7	13,7	10	19,6
III – M^- (strongly reactive)	–	–	–	–	2	18,2	9	81,8

Comparison of the data presented in the table gives grounds to state that among the stable animals, 19 animals (67.9%) were registered with a strong balanced mobile type of higher nervous activity and 9 animals (32.1%) with a strong balanced inert type, while piglets with strong unbalanced and weak types of nervous activity were not noted among these animals. Among the animals that react poorly to stress, 15 animals (29.4%) were registered with a strong balanced mobile type, 19 animals (37.3%) with a strong balanced inert type, 7 animals (13.7%) with a strong unbalanced type and 10 animals (19.6%) with weak types. Strongly reactive young animals were represented by 2 heads (18.2%) with strong unbalanced and 9 heads (81.8%) with weak types of nervous activity.

Scientific practice has proven, and numerous studies have confirmed, that the nature of the adaptive reaction of animals to the action of any stress factors is reflected in their productivity. The results of determining the live weight of piglets of different types of nervous activity are presented in Table. 7.

The obtained data give reason to believe that among the total number of experimental piglets of different stress resistance, individuals classified as strong balanced mobile and strong balanced inert types of higher nervous activity grew more intensively. Moreover, the advantage in increasing live weight occurred at all stages of their growth. While these differences between young animals with strong unbalanced and weak types are not significant. In particular, at 2 months of age, the difference in live weight between these animals was greater, respectively, by an average of 2.5 and 3.9 kg or 11.6 and 19.0% ($p<0.01$) and 0.7 and 2.1 kg or 8.1 and 10.0% in favor of the first two groups. Subsequently, the difference between the distribution groups by this indicator increased. In particular, at 4 months of age, when the second phase of growing came to an end, piglets with a strong balanced mobile type of higher nervous activity exceeded their peers who had a strong balanced type by 3.7 kg or 1.2%, a strong unbalanced type by 5.2 kg or 9.5%, and a weak type by 7.5 kg or 14.5% ($p<0.01$). In turn, the advantage

in terms of increasing live weight on their side at 6 months of age increased and amounted to 9.1 kg or 8.9%; 11.9 kg or 11.9% and 14.3 kg or 14.7%, respectively ($p<0.05$).

Table 7

Formation of live weight of young animals of different types of nervous activity, kg ($\bar{x} \pm S_{\bar{x}}$)

Indicator	Type of nervous activity			
	strong balanced mobile (n = 34)	strong, balanced inert (n = 28)	strong, unbalanced (n = 9)	weak (n = 19)
Live weight of piglets, kg at the age of:				
2 months	24,35±0,90**	22,51±0,58	21,82±1,03	20,46±0,86
4 months	59,69±2,02**	56,03±1,58	54,51±1,73	52,15±1,42
6 months	111,74±5,94*	102,60±4,09	99,82±2,94	97,41±3,97

*Note. *p<0.05; **p<0.01 – probability of difference in relation to the group of weak young animals*

Quite a clear variability in growth during these age periods of growth was also characteristic of the young animals included in the group with a strong balanced inert type of higher nervous activity. Analysis of changes in their live weight at 2 months of age proves that they, while inferior in this indicator to their peers with a strong balanced mobile type of higher nervous activity, had a stable, but at the same trend, advantage over representatives of the group with a strong unbalanced type by 0.7 kg or 3.2% and young animals of a weak type by - 2.1 kg or 10.0%. While ontogenetic differences in live weight values between these distribution groups at 4 months of age were 1.5 kg or 2.8% and 3.9 kg or 7.4%, respectively. During the final phase of fattening, the process of increasing live weight intensified and the difference between these groups increased to 2.8 and 5.2 kg or 2.8 and 5.3%, respectively.

In contrast, among peers who had strong unbalanced and weak types of nervous activity, the age difference in fluctuations in live weight was insignificant: its increase ranged only from 1.4 kg to 2.4 kg or from 2.4% to 6.6%.

During the period of growing up and fattening, the total increase in live weight of young animals, which were classified as a group with a strong balanced mobile type of higher nervous activity, increased by 87.4 kg or 4.6 times, strong balanced inert - by 80.1 kg or 4.6 times, strong balanced mobile - by 78.0 kg or 4.6 times, weak - by 77.0 kg or 4.8 times.

The growth intensity of experimental piglets can be judged by the values of changes in average daily live weight gains (Table 8).

In the process of processing the results, it was established that the growth rates of young animals of different groups increased as their age increased. Against this background, piglets with a strong balanced mobile type of higher nervous activity were significantly better in all periods of growing. Evaluating the average daily gains in live weight in the age dynamics of changes, their natural increase was recorded in the period from 2 to 4 months from 29.0 g to 59.0 g or from 5.3% to 11.3% ($p<0.01$). Their high growth rate was maintained during the fattening period from 90 g to 160.0 g or from 11.8% to 23.1 ($p<0.001$). The leading position in terms of the manifestation of higher adaptive capacity to the action of a stress factor in piglets classified as a group of a strong unbalanced type of nervous activity during this period was also strengthened in relation to young animals, which are characterized by a strong unbalanced type and the difference

in the studied indicator between them also became statistically significant (p<0.01). In the age interval from 2 to 6 months, an increase in growth intensity in these animals was recorded within the range from 60.0 g to 85.0 g or from 9.4% to 13.5%. Despite the superiority of piglets with a strong balanced mobile type of nervous activity over the rest of the groups in terms of average daily live weight gains, statistically significant significance for this indicator was registered only for young animals with a strong unbalanced and weak types (p<0.05 in both cases of comparison).

Table 8

The level of average daily live weight gains of young animals of different types of nervous activity, ($\bar{x} \pm S_{\bar{x}}$)

Indicator	Type of nervous activity			
	strong balanced mobile (n = 34)	strong, balanced inert (n = 28)	strong, unbalanced (n = 9)	weak (n = 19)
Average daily gain for the period, g:				
from 2 to 4 months of age	579±11,75**	550±12,14	536±19,22	520±14,63
from 4 to 6 months of age	853±12,51**/***	763±16,84	743±26,77	693±27,90
from 2 to 6 months of age	716±19,48*/*	656±23,73	639±22,51	631±28,95

*Note. *p<0.05; **p<0.01; ***p<0.001 – probability of difference in relation to the group of strong unbalanced and weak young animals*

Piglets with a strong balanced inert type of higher nervous activity also stood out with higher average daily live weight gains compared to animals characterized by a strong unbalanced type: during growing by 14.0 g or 2.6% and during fattening - by 20.0 g or 2.7%. This also caused a general increase in the level of average daily live weight gains during the research by 17.0 g or 2.7%. In young animals with a strong unbalanced type of nervous activity, the growth intensity increased not only compared to animals with a strong unbalanced type, but also compared to individuals of a weak type. In particular, in the period from 2 to 4 months of age, the level of average daily gains in them increased by 30.0 g or 5.8%, from 4 to 6 months of age - by 70.0 or 10.1%, and for the total period of research - by 25.0 g or 4.0%. Since the intergroup differences in the growth energy of young animals in these age periods are insignificant, they turned out to be unreliable when statistically processing the results.

The relationship between the type of nervous activity and the intensity of growth in piglets with a strong unbalanced type was expressed to the smallest extent in relation to peers with a weak type and the difference found between them was: from 2 to 4 months of age 16.0 g or 3.1%, from 4 to 6 months of age – 50.0 g or 7.2% and from 2 to 6 months of age – 8.0 g or 1.3%, however, it did not acquire statistical significance between them.

Discussion. With the introduction of intensive pork production technologies, the problem of stress phenomena, especially at an early age of piglets, is becoming increasingly acute (Poroshynska O.A. et al., 2020; Lange A. et al., 2020). Piglets after weaning from their mothers react quite strongly to changes in environmental conditions and get used to new ones after a certain time. Their ability to adapt is closely related to changes in productive qualities (Munsterhjelm C. et al., 2010; Campbell J. et al., 2013; Sutherland M.A. et al., 2014; Bankole T. et al., 2024). In this case, stress is considered as an adaptation syndrome and occurs when the body's protective functions are mobilized to the action of external stressors (Gonzalez-Rivas P.A. et al., 2020; Serviento A.M. et al.,

2020). However, a generalization of domestic literature sources in which studies have been initiated on assessing the stress resistance of piglets in the early period of postnatal growth indicates insufficient attention by scientists, who only fragmentarily reveal the essence of this phenomenon, which in turn encourages the experimental development of appropriate techniques.

Within the framework of the conducted research, the peculiarities of the formation of live weight and growth intensity of young pigs of Danish origin were detailed depending on stress resistance and types of higher nervous activity, assessed during weaning and reaching 6 months of age. It was found that the growth energy of young pigs of the M^+ group for the period from weaning from mothers to the end of the growing-up period was quite high, compared with peers of groups M^0 and $M^–$ by 62.0 g or 13.9% ($p<0.05$) and 87 g or 20.6% ($p<0.01$). In the period from 2 to 4 months of age, piglets of all groups further increase in average daily gains, however, the pace of this process slows down to minimal values and the difference between young pigs of groups M^+ and M^0 has only the character of a tendency to increase. At the age of 6 months, the growth rate of the young pigs of the M^+ group began to increase again and they exceeded the piglets from the M^0 group by 44.0 g or 5.6% ($p<0.05$) and individuals from the $M^–$ group by 81.0 g or 10.7% ($p<0.01$). The improvement of the adaptive capacity to the stress factor in the M^+ group of piglets was also accompanied by a significant increase in the average daily gain over the entire growing period by 39.0 g or 6.3% ($p<0.05$) and 67.0 g or 11.3% ($p<0.01$) compared to the young pigs from the other groups. The obtained data extend the published results (White H.M., et al., 2008; Sutherland M.A. et al., 2014; Stovbetska L. et al., 2021).

When distributing piglets taking into account the types of higher nervous activity and using the motor-food method, it was found that 34 heads (37.8%) of piglets from the total number have a strong balanced mobile type, 28 heads (31.1%) - a strong balanced weighted inert, 9 heads (10.0%) – strong unbalanced and 19 heads (21.1%) – weak. Analysis of the features of individual assessment of young animals indicates a different rate of their response to weaning from sows. Piglets classified as resistant are characterized by an almost maximum yield of individuals of a strong balanced mobile type of higher nervous activity, which react less to transfer to the finishing shop with a decrease in productivity and have a higher adaptive suitability for industrial pork production technology. The highest level of strength of nervous processes, balance of excitation and inhibition processes, and their mobility were observed in piglets that had a strong balanced mobile type of nervous activity with a high statistical difference of $p<0.001$ in all cases of comparison with the group identified as weak. A similar picture of changes in eating behavior in pigs in connection with technological stresses is reported (Danchuk O.V. et al., 2020).

Therefore, the conducted studies do not exhaust the entire depth of the problem, but within the framework of its initial implementation, farm specialists should take into account not only their stress resistance, but also the types of higher nervous activity for the purpose of directed management of the weaning process of piglets.

Conclusions:

1. Weaning piglets from their mothers is a heavy functional load on their body, which is accompanied by growth retardation. The most susceptible to the action of the stress factor were young animals classified as group $M^–$.

2. The level of stress resistance of young animals is directly related to the typological features of higher nervous activity. Piglets classified as resistant are characterized by an approximate maximum yield of individuals of a strong balanced mobile type of higher nervous activity, which react less to transfer to the growing-up shop

with a decrease in productivity and have a higher adaptive suitability for industrial pork production technology.

References

Bankole, T.O., Adebiyi, O.A., Ewuola, E.O., Oluyemi, A.A., Abiola, O.J. & Adebiyi, F.G. (2024). Effect of weaning regimes on growth performance, stress and behavioural responses in weanling pigs. *Slovak Journal of Animal Science,* 57 (3), 12–21. https://doi.org/10.36547/sjas.888

Campbell, J.M., Crenshaw, J.D. & Javier, P. (2013). The biological stress of early weaned piglets. *Journal of Animal Science and Biotechnology,* 4 (1), 19–27. https://doi.org/10.1186/2049-1891-4-19

Danchuk, O.V., Broshkov, V.I., Karpovsky, V.I., Bobrytska, M.I., Tsvivlikhovsky, M.I., Tomchuk, V.A., Trokoz, V.O., & Kovalchuk, I.I. (2020). Types of higher nervous activity in pigs: characteristics of behavior and effects of technological stress. *Neurophysiology*, 52, 358–366. https://doi.org/s11062-021-09892-7

da Fonseca de Oliveira, A.C., Vanelli, K., Sotomaior, C.S., Weber, S.H., & Costa, L.B. (2019). Impacts on performance of growing-finishing pigs under heat stress conditions: a meta-analysis. Veterinary Research Communications, 43 (1), 37–43. https://doi.org/10.1007/s11259-018-9741-1

Dybkjær L. (1992). Determination of behavioral indicators of «stress» in piglets weaned at an early age. *Applied Animal Behaviour Science.* 35 (2). 135–147. https://doi.org/10.1016/0168-1591(92)90004-U

Gonzalez-Rivas, P.A., Chauhan, S.S., Ha, M., Fegan, N., Dunshea, F.R., & Warner R.D. (2020). Effects of heat stress on animal physiology, metabolism, and meat quality: A review. *Meat Science*, 162, 108025. https://doi.org/10.1016/j.meatsci.2019.108025

Chalyi, O.I., Nahornyi S.A. (2023). Osoblyvosti vyroshchuvannia porosiat u postembrionalnyi period [Features of raising piglets in the post-embryonic period]. *materialy Mizhnararodnoi naukovo-praktychnoi konferentsii «Suchasni tendentsii rozvytku haluzi tvarynnytstva: svitovyi ta natsionalnyi vymiry»*, Poltava. https://doi.org/10.37143/Conf-1-2023. (in Ukrainian).

Hao, Y., Xing, M., & Gu X. (2021). Research progress on oxidative stress and its nutritional regulation strategies in pigs. *Animals (Basel)*, 11 (5), 1384–3205. https://doi.org/10.3390/ani11051384

Karpovskyi, V.I., Trokoz, V.O., Kryvoruchko, D.I. (2012). Metodyka vyznachennia typiv vyshchoi nervovoi diialnosti svynei u vyrobnychykh umovakh [Methodology for determining the types of higher nervous activity of pigs in production conditions]. *Naukovo-tekhnichnyi biuleten Instytutu biolohii tvaryn ta derzhavnoho naukovo-doslidnoho kontrolnoho instytutu vetpreparativ ta kormovykh dobavok*, 13, 1/2. 105–108. (In Ukrainian).

Čobanović, N., Stajković, S., Blagojević, B., Betić, N.,Dimitrijević, M., Vasilev, D., & Karabasil, N. (2020). The effects of season on health, welfare, and carcass and meat quality of slaughter pigs. *International Journal of Biometeorology,* 64 (11), 1899–1909. https://doi.org/10.1007/s00484-020-01977-y

Lange, A., Gentz, M., Hahne, M., Lambertz, C., Gauly, M., Burfeind, O., & Traulsen, I. (2020). Effects of different farrowing and rearing systems on post-weaning stress in piglets. *Agriculture. 10* (6), 230–243. https://doi.org/10.3390/agriculture1006023

Mayorga, E.J., Ross, J.W., Keating, A.F., Rhoads, R.P., & Baumgard, L.H. (2020). Biology of heat stress; thenexus between intestinal hyperpermeability and swine

reproduction. *Theriogenology,* 154, 73–83. https://doi.org/10.1016/j.theriogenology.2020.05.023

Munsterhjelm, C., Valros, A., Heinonen, M., Hälli, O., Siljander-Rasi, H., & Peltoniemi, O.A.T. (2010). Environmental enrichment in early life affects cortisol patterns in growing pigs. *Animal,* 4 (2), 242–249. https://doi.org/10.1017/S1751731109990814

O'Connor, E.A., Parker, M.O., McLeman, M.A., Demmers,T.G., Lowe, J.C., Cui, L., Davey, E.L., Owen, R.C., Wathes, C.M., & Abeyesinghe, S.M. (2010). The impact of chronic environmental stressors on growing pigs, Sus scrofa (Part 1): stress physiology,and play behaviour. *Animal,* 4 (11), 1899–1909. https://doi.org/10.1017/S1751731110001072

Patent na korysnu model № 97393 Ukraina, MPK A01K 67/02 Sposib vidboru stresostiikoho remontnoho molodniaku [Method of selecting stress-resistant repair young stock]. Tsereniuk, O.M.; Instytut tvarynnytstva NAAN. № u 201411118; zaiavl. 13.10.2014; opubl. 10.03.2015. Biul. № 5. 3 s. (In Ukrainian).

Poroshynska, O.A., Shmaiun, S.S., Nishchemenko, M.P., Stovbetska, L.S., Yemelianenko, A.A., Kozii, V.I. (2020). Vplyv stresovykh chynnykiv na adaptyvni ta povedinkovi reaktsii u svynomatok i porosiat [The influence of stress factors on adaptive and behavioral responses in sows and piglets]. *Naukovyi visnyk veterynarnoi medytsyny*, 2. 110–121. https://doi.org/10.33245/2310-4902-2020-160-2-110-121. (In Ukrainian).

Ramirez, B.C., Hayes, M.D., C.F.S. Condotta, I. & Leonard, S.M. (2022). Impact of housing environment and management on pre-/post-weaning piglet productivity, *Journal of Animal Science,* 100 (6), 142–154. https://doi.org/10.1093/jas/skac142

Serviento, A.M., Lebret, B., & Renaudeau, D. (2020). Chronic prenatal heat stress alters growth, carcass composition, and physiological response of growing pigs subjected to postnatal heat stress. *Journal of Animal Science*, 98 (5), 161–174. https://doi.org/10.1093/jas/skaa161

Skaperda, Z., Veskoukis, A.S., & Kouretas, D. (2019). Farm animal welfare, productivity and meat quality: Interrelation with redox status regulation and antioxidant supplementation as a nutritional intervention (Review). *World Academy of Sciences Journal,* 1 (4), 177–183. https://doi.org/10.3892/wasj.2019.19

Stovbetska, L., Poroshinska, O., Nischemenko, M., Shmayun, S., Emelyanenko, A., & Koziy, V. (2021). Effect of stress on performance and physiological functions in pigs. *Scientific messenger of Lviv national university of veterinary medicine and biotechnologies. Series: Veterinary sciences,* 23 (102), 14–23. https://doi.org/10.32718/nvlvet10203. (In Ukrainian).

Sutherland, M.A., Backus, B.L., & Mc. Glone, J.J. (2014). Effects of transport at weaning on the behavior, physiology and performance of pigs. *Animals (Basel),* 4 (4), 657–669. https://doi.org/10.3390/ani4040657

Vyslotska, L., Gutyj, B., Kozenko, O., Khalak, V., Chornyj, M., Martyshuk, T., Krempa, N., Vozna, O., & Todoriuk, V. (2021). System of antioxidant protection of the body of piglets under the action of feed additive «Sylimevit». *Scientific messenger of LNU of veterinary medicine and biotechnologies. Series: Veterinary Sciences,* 23 (104), 10–17. https://doi.org/10.32718/nvlvet10402. (In Ukrainian).

White, H.M., Richert, B.T., Schinckel, A.P., Burgess, J.R., Donkin, S.S., & Latour, M.A. (2008). Effects of stress on growth performance and baconquality in grow-finish pigs housed at two densities. *Journal of Animal Science,* 86 (8), 1789–1798. https://doi.org/10.2527/jas.2007-0801

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ASSESSMENT OF GENETIC DIVERSITY IN THE POPULATION OF CHAROLAIS CATTLE OF UKRAINIAN SELECTION USING MICROSATELLITE MARKERS

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The article presents the results of a study of the genetic-population structure in a herd of beef-type Charolais breed cows of Ukrainian selection (SEEF "Hontarivka", Kharkiv region). Genetic variation analysis in the experimental group of animals was performed using 10 microsatellite loci recommended by FAO-ISAG: ETH225, BM2113, ETH3, BM1818, BM1824, ILSTS006, INRA023, TAGLA053, TAGLA122, ETH10. The amplification products were separated in native polyacrylamide gels. All the loci studied were polymorphic. The number of detected alleles per locus ranged from 2 (ETH10) to 10 (TGLA053) (an average of 5 alleles per locus), the size of which ranged from 117 bp (ETH3) to 307 bp (ILSTS006). The vast majority of the studied loci belong to informative and valuable markers (PIC > 0.5). The most polymorphic loci were TGLA053 (PIC =0.81) and INRA023 (PIC =0.72). The main population and genetic parameters for the studied loci are calculated. The highest values of heterozygosity (H_e) and effective allele count (n_e) were inherent in the loci TGLA053 (H_e =0.82, n_e =5.7) and INRA023 (H_e =0.73, n_e =3.8). The minimum values of observed heterozygosity are set for loci ETH10 (H_o =0.21) and TGLA122 (H_o =0.44).

Most microsatellite loci are characterized by an equilibrium state between actual and expected genotype frequency indicators, and a likely deviation in the form of heterozygote deficiency was established only for the TGLA122 locus (F_{is} = 0.29; $p < 0.05$).

Changes in the genetic structure of the experimental population of Charolais cattle in comparison with data from previous years and populations from other regions of the world are analyzed. These results indicate a significant narrowing of genetic variability in the domestic Charolais population. This can have further negative consequences and requires replenishment of allelic diversity and control of genetic processes in breeding work using DNA markers.

Keywords: microsatellites, polymorphism, population, Charolais, allele, genotype, heterozygosity

ОЦІНКА ГЕНЕТИЧНОГО РІЗНОМАНІТТЯ В ПОПУЛЯЦІЇ КОРІВ ШАРОЛЕЗЬКОЇ ПОРОДИ УКРАЇНСЬКОЇ СЕЛЕКЦІЇ ЗА ВИКОРИСТАННЯ МІКРОСАТЕЛІТНИХ МАРКЕРІВ

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У статті наведені результати дослідження генетико-популяційної структури в стаді корів м'ясного напряму продуктивності шаролезької породи української селекції (ДПДГ «Гонтарівка», Харківської області). Аналіз генетичної мінливості в дослідній групі тварин проводили з використанням 10 мікросателітних локусів рекомендованих FAO-ISAG: ETH225, BM2113, ETH3, BM1818, BM1824, ILSTS006, INRA023, TAGLA053, TAGLA122, ETH10. Продукти ампліфікації розділяли в нативних поліакриламідних гелях. Всі досліджені локуси виявились поліморфними. Кількість виявлених алелів на локус коливалась від 2 (ETH10) до 10 (TGLA053) (у середньому 5 алелів на локус), розмір яких знаходився в межах від 117 п.н. (ETH3) – до 307 п.н. (ILSTS006). Переважна більшість досліджених локусів належить до інформативно-цінних маркерів (PIC > 0,5). Найбільш поліморфними виявилися локуси TGLA053 (PIC=0,81) та INRA023 (PIC=0,72). Розраховано основні популяційно-генетичні параметри за досліджуваними локусами. Найвищі значення показників гетерозиготності (He) і ефективної кількості алелів (n_e) були властиві локусам TGLA053 (H_e=0,82, n_e=5,7) та INRA023 (H_e=0,73, n_e=3,8). Мінімальні значення фактичної гетерозиготності встановлені для локусів ETH10 (H_o=0,21) і TGLA122 (H_o=0,44).

Для більшості мікросателітних локусів властивим є рівноважний стан між фактичними і очікуваними показниками частот генотипів, Вірогідне відхилення у вигляді дефіциту гетерозигот встановлено лише для локусу TGLA122 (F_{is} = 0,29; $p < 0,05$).

Проаналізовано зміни в генетичній структурі дослідної популяції корів породи шароле порівняно з даними попередніх років та популяцій з інших регіонів світу. Отримані результати свідчать про суттєве звуження генетичної мінливості у вітчизняній популяції шароле. Це може мати в подальшому негативні наслідки та потребує поповнення алельного різноманіття та контролю генетичних процесів в селекційно-племінній роботі за використання ДНК-маркерів.

Ключові слова: мікросателіти, поліморфізм, популяція, шароле, алель, генотип, гетерозиготність

Introduction. Knowledge of the genetic diversity of cattle (cattle) is an important step for effective management of genetic resources in agriculture. Assessment of the genetic diversity of animals is carried out using molecular markers. Microsatellites are the most involved DNA markers for this type of research. Microsatellites (short tandem repeats, STR) are widely used for animal identification and genetic examination of origin, scientific support for breeding work, determination of the level of consolidation of created groups and the degree of genetic differentiation of populations (Debrauwere H. et al., 1997; Senan S. et al., 2014). Microsatellites can be used as a fairly subtle and effective tool for studying genetic variability, which allows monitoring processes, in particular, in artificially reproducible populations of farm animals (Shel'ov, 2017; Al-Jubori & Senkal,

2023) (Shel'ov, 2015; Mishra S. et al., 2017; Zhao J. et al., 2017).

To assess the biodiversity and genetic research of cattle, the International Society for animal genetics (ISAG) and the Food and Agriculture Organization of the United Nations (FAO) have proposed microsatellite panels and recommendations for their use in scientific research (ISAG/FAO, 2004; FAO, 2011). ISAG recommends 12 STR loci (TGLA53, TGLA122, TGLA126, TGLA227, ETH3, ETH10, ETH225, SPS115, INRA23, BM2113, BM1824, BM1818) based on dinucleotide repeats for testing and confirming paternity (Koskinen, 2006). According to the FAO recommendations, 30 microsatellite loci were identified for cattle (FAO, 2011).

The study of allelic polymorphism of cattle microsatellite loci began in the late 1990s. This development coincided with the introduction of specialized equipment, namely DNA analyzers such as Abi377, Abi3110, and their more modern versions. These analyzers enabled accurate fragment analysis of amplified segments. As a result, deviations in determining the number and size of microsatellite alleles were minimized. However, most domestic scientists of the NAAS system are deprived of the possibility of using such equipment and, at best, send samples for analysis abroad. An example of such works is the study of microsatellite variability of native cattle breeds of domestic selection, such as Ukrainian Gray (Shkavro et al., 2010), lebedinskaya (Shkavro et al.; 2018, Ladyka et al., 2019), Krasnaya stepnaya (Kramarenko et al., 2018), Southern Meat (Snegin et al., 2019, Kramarenko, 2019), Buffaloes (Dzitsiuk et al., 2020). Given the great desire to engage in research in this area, we searched for alternative methodological approaches. This possibility was found based on the use of native gel electrophoresis and detailed analysis of the conformational structure of the DNA molecules that are formed (Kulibaba & Liashko, 2016).

The paper presented for consideration is a continuation of the study of microsatellite polymorphism of cattle populations in the Kharkiv region (Liashko et al., 2024) and is devoted to the study of the genetic variability of one of the breeds of meat productivity – the Charolais breed. The versatility to adapt to a wide range of farming conditions has made Charolais a widely used cattle meat breed in the world. In Ukraine, populations of the Charolais breed were mainly concentrated in the experimental farm "Gontareva" of the Institute of animal husbandry of the National Academy of Sciences, LLC "Rachanskoe" of the Zhytomyr region and LLC "Agricor holding" of the Chernihiv region (Petrenko et al., 2016).

Purpose of the study. To assess the genetic diversity in the Charolais cattle herd *SEEF "Hontarivka"* (Kharkiv region), which is a specialized breeding farm for breeding this breed. To analyze the changes that occurred during the reproduction of the local Charolais population on the basis of reproductive and breeding management, as well as local isolation, data on the genetic diversity of the Charolais breed of the same farm, made in 2010, will be used (Shkavro et al.) and from other regions of the world (Sifuentes-Rincón et al., 2007; Putnova et al., 2011).

Materials and methods. The object of research was the number of cows of the meat direction of productivity of the sharolezh breed of Ukrainian selection, which was kept in the breeding reproducer of the State Enterprise Experimental Farm "Hontarivka" (Volchansky district, Kharkiv region) in 2018. The sample consisted of 30 individuals. DNA isolation was performed from hair follicles using the NeoPrep DNA reagent kit (Lab Neogene P.C., Ukraine).

10 microsatellite loci from the FAO-ISAG list were used to study microsatellite variability: ETH225, BM2113, ETH3, BM1818, BM1824, ILSTS006, INRA023, TAGLA053, TAGLA122, ETH10.

Amplification of fragments of the studied loci was performed using the Amply-4

thermal cycler (Biocom, Russia) according to the appropriate program: 1 cycle – denaturation 94°C 3 min; 35 cycles – denaturation 94°C 20 s, annealing 30 s (56-62 °C depending on the locus), elongation 72°C 55 S; 1 cycle – final elongation 72 °C 10 min. The volume of the reaction mixture was 10 μL, which included 5 μL of Mastermix (2×buffer with 4 mM $MgCl_2$, 0.4 mM DNTP mixture and 0.5 units. DreamTaq DNA polymerase (Thermo Scientific)), 2.5 μL of 1 pM primer and 2.5 μL of DNA Matrix.

Amplification products were separated in native polyacrylamide gels of various concentrations (5-8 %). Gel staining was performed using ethidium bromide (visualization was performed in the ultraviolet spectrum) or silver nitrate. Genotyping of individuals based on a set of microsatellite markers was performed according to the method of assessing the conformational structure of DNA under the conditions of native PAAG electrophoresis based on the use of available equipment (Kulibaba & Liashko, 2016). This makes it possible to accurately determine the allelic spectrum (number of alleles) of the studied SSR loci. The disadvantage of this approach is certain inaccuracies in determining the size of amplified fragments.

The fragment size was determined using molecular weight markers pUC19 and O'RangeRuler 20 bp (Thermo Scientific, USA). Molecular weights of amplification products were calculated using the GelAnalyzer program (version 2010a freeware). Control of the software calculation of the size of electrophoregram fragments was performed using a millimeter ruler on a monitor screen.

Based on the data obtained, genotype and allele frequencies, actual (H_o) and expected (H_e) heterozygosity, effective allele count (ne), Wright fixation index (F_{is}) were calculated, and The Hardy-Weinberg genotype distribution was checked using the GenAlEx 6.503 add-on integrated in Excel (Peakall &Smouse, 2012). https://biology-assets.anu.edu.au/GenAlEx/Download.html).

Research results. Based on the results of the conducted studies, it was found that all microsatellite loci that were used in experimental animal populations are polymorphic (the proportion of polymorphic loci was 100 %). The number of detected alleles ranged from 2 (ETH10) to 10 (TGLA053) per locus. The analysis of the obtained results of genotyping of individuals allowed us to identify a total of 50 alleles for 10 microsatellite loci (an average of 5 alleles per locus), the size of which ranged from 117 bp (ETH3) to 307 bp (ILSTS006) (table 1).

Table 1

Allele frequencies in the study population at 10 loci

locus	Allele frequency (allele (bp)-frequency)									
ETH10	**216**-0,89					**218**-0,11				
BM1824	**190**-0,15			**194**-0,46			**196**-0,39			
BM1818	**266**-0,08		**268**-0,63			**276**-0,19			**278**-0,10	
ILSTS006	**291**-0,11		**295**-0,28			**301**-0,11			**307**-0,50	
BM2113	**125**-0,22		**127**-0,09		**135**-0,17		**137**-0,50		**139**-0,02	
ETH3	**117**-0,65		**119**-0,10		**121**-0,04		**125**-0,17		**127**-0,04	
TGLA122	**148**-0,56		**152**-0,03		**156**-0,17		**160**-0,17		**172**-0,07	
ETH225	**140**-0,02		**144**-0,05		**146**-0,02		**150**-0,22		**152**-0,60	**154**-0,09
INRA023	**199**-0,04		**203**-0,18		**205**-0,40		**211**-0,04		**215**-0,25	**219**-0,09
TGLA53	**154**-0,11	**158**-0,04	**160**-0,07	**168**-0,06	**170**-0,03	**174**-0,17	**178**-0,04	**180**-0,33	**182**-0,11	**190**-0,04

The highest level of polymorphism in the study population was found at the TGLA53 locus (10 alleles). However, the frequency of occurrence of only 2 alleles of this locus was 0.17 and 0.33, and for 6 out of 10 alleles, the frequency value was in the range of 4-7%, which affected the value of the effective number of alleles (n_e = 5.68 (57) (table. 2).

Table 2

Main genetic and population indicators in the experimental group of cattle by microsatellite markers

Locus	Indicators						
	N_a	n_e	H_o	H_e	F_{is}	χ^2	PIC
ETH225	6	2,38	0,60	0,58	-0,03	0.071	0,46
BM2113	5	2,98	0,63	0,66	0,05	0.124	0,62
ETH3	5	2,15	0,51	0,54	0,06	0.185	0,50
BM1818	4	2,23	0,57	0,55	-0,04	0.079	0,51
BM1824	3	2,59	0,58	0,61	0,05	0.145	0,54
ILSTS006	4	2,84	0,62	0,65	0,05	0.128	0,63
INRA023	6	3,76	0,75	0,73	-0,03	0.045	0,72
TGLA53	10	5,68	0,79	0,82	0,04	0.08	0,81
TGLA122	5	2,65	0,44	0,62	0,29	5.06*	0,58
ETH10	2	1,24	0,21	0,20	-0,05	0.15	0,18
Average	5,0	2,85	0,57	0,60	0,04	0.61	0,56
Error	0.68	0.37	0.05	0.05	0.03	1.56	0.05

*Notes: Na - number of alleles, n_e – effective number of alleles, Ho – observed heterozygosity, He – expected heterozygosity, F_{IS} – Wright fixation index, PIC - information polymorphism Index, * - P<0.05.*

For 6-allele loci, the most aligned in allele frequencies (0.17 to 0.06) and the maximum value of their effective number (n_e = 3.8 (63)) was locus INRA023. among loci with 5 alleles, the most uniform distribution of allele frequencies was observed for locus BM2113 (0.2 to 0.075), of which 60 % (n_e = 2.98) can be considered effective. Higher ne scores occurred for the less polymorphic loci ILSTS006 (2.84 and 71% of effective alleles) and BM1824 (2.59 and 86%, respectively) (Table 1). 2).

Analysis of the distribution of expected heterozygosity (H_e), as one of the main indicators of genetic variation in the population, revealed the average level of the studied trait for a set of microsatellite loci (H_e = 0.6 ± 0.05). High values of expected heterozygosity were established for loci TGLA053 (H_e = 0.82) and inra023 (H_e = 0.73). The lowest level of variability in the study population was observed at the two-allele ETH10 locus (H_e = 0.2), which coincides with the actual number of heterozygous individuals (H_o = 0.21) and corresponds to the normal Hardy-Weinberg distribution (χ^2 = 0.15; table. 2).

The vast majority of microsatellite loci are characterized by an equilibrium state between actual and expected indicators (χ^2 = 5.1; $p < 0.05$), which was caused by an excess of homozygous individuals (F_{is} = 0.29). The average value of the Wright fixation index gives reason to believe that an independent state of genotype distribution occurs in the experimental cow population (F_{is} = 0.04 ± 0.03; table. 2).

Based on the results of the conducted studies, it was found that most of the studied loci belong to informatively valuable markers (average PIC value=0.56 ± 0.05). The exception is loci ETH10 (PIC = 0.18) and ETH225 (PIC = 0.46).

The results of the analysis prove the possibility of using 9 out of 10 SSR markers

for certification, identification and confirmation of the origin of individual individuals within the studied cattle populations.

Discussion. To analyze the changes that occurred during the reproduction of the experimental Charolais population, we used data on the genetic diversity of the Charolais breed from the same farm, made in 2010 (Shkavro et al., 2010), as well as populations from other regions – Mexico (two populations, Sifuentes-Rincón et al., 2007) and the Czech Republic (Putnova et al., 2011). Table 3 shows data on indicators of genetic variability of Charolais populations (total number of alleles at the locus (Na) and expected heterozygosity (H_e)), the analysis of which allows us to draw certain conclusions regarding the assessment of genetic processes that occur in artificially reproduced animal populations.

Table 3

Indicators of genetic variation in Charolais populations from different regions

Locus	Population, variability indicators									
	Char1		Char2		Char3		Char4		Char5	
	N_a	H_e	N_a	H_e	N_a	H_e	N_a	H_e	N_a	H_e
ETH225	6	0,58	5	0.51	5	0.74	-	-	-	-
BM2113	5	0,66	5	0.67	7	0.82	8	0.80	9	0.78
ETH3	5	0,54	5	0.53	5	0.55	-	-	-	-
BM1818	4	0,55	-	-	7	0.78	-	-	-	-
BM1824	3	0,61	3	0.66	5	0.73	6	0.80	8	0.78
ILSTS006	4	0,65	-	-	10	0.88	-	-	-	-
INRA023	6	0,73	6	0.76	10	0.78	13	0.87	13	0.88
TGLA53	10	0,82	10	0.83	-	-	13	0.91	14	0.88
TGLA122	5	0,62	-	-	11	0.74	-	-	-	-
ETH10	2	0,20	2	0.19	4	0.15	3	0.48	5	0.40
TGLA226	-	-	3	0.53	5	0.58	-	-	-	-
TGLA227	-	-	6	0.81	9	0.84	-	-	-	-
SPS115	-	--	5	0.62	5	0.51	-	-	-	-

Notes. Char1-own data; Char2-Shkavro et al., 2010; Char3-Czech population, Putnova et al., 2011; Char4-Mexican (import from France), Char5 – Mexican (import from France , Great Britain and Ireland), Sifuentes-Rincón et al., 2007; Na-number of alleles, He-expected heterozygosity

It should be noted that it was not possible to analyze polymorphism for all the studied loci due to the fact that the authors used different microsatellite panels. 7 common loci were found to compare Char1-Char2, 9 for Char1-Char3, 5 for Char1-Char2 (Char3). In addition, it is incorrect to compare the size of alleles and their frequencies for the Ukrainian Charolais population (Char1-Char2), but comparing data obtained using DNA analyzers (Char2-Char5) is of interest.

Analysis of the genetic structure of the Ukrainian population of Charolais cattle during 2010-2018 (possibly another period of time in the absence of information about the year of birth of cattle of the experimental livestock) revealed the absence of significant changes in the main indicators. The total number of Char1-Char2 alleles was 37 and 36 (average 5.3 and 5.1, respectively), and the average level of expected heterozygosity (H_e) in both groups of cattle was 0.59. The absence of changes in the genetic variability of the experimental population may be due to the peculiarity of the reproduction scheme of this breed based on the use of a limited number of breeding bulls in the breeding core.

The results of assessing genetic variability in Charolais populations from other regions confirm our assumption. If we compare the data with the Czech population

(Char1-Char3, table.3), we have a significant advantage of Char3 both in the total number of alleles 64 versus 40 (Char1) (on average 7.1 and 4.4, respectively) and in the average level of heterozygosity (H_e^{Chag3} = 0.69; H_e^{Chag1} = 0.57).

For the Mexican population of Char4 (imported Charolais animals from France), the indicators of genetic variation at 5 common microsatellite loci have even higher values: N_a^{Char4} = 43 (medium 8.6) and H_e^{Chag4} = 0.77 compared to the Ukrainian population (N_a^{Char1} = 26 (5.2) and H_e^{Chag1}= 0.60). A similar trend is observed for another Mexican population of Char5, created on the basis of Charolaise animals imported from France, Great Britain and Ireland (N_a^{Char5} = 49 (average 9.8), H_e^{Chag5}= 0.74).

Let's consider how the qualitative composition of alleles for the studied loci changed in the Ukrainian population of the Charolais breed. We are talking about the frequencies of the most common alleles inherent in the original forms of French breeding (Table 4).

Table 4

The most common alleles in Charolais cow populations

Locus	Population			
	Char2	**Char3**	**Char4**	**Char5**
BM2113	131, 135, 137	131	131	133, 135
TGLA53	166, 170	-	151, 153, 157	155, 157, 159
INRA023	200, 206, 214	206	203, 205, 207	199,203, 205
BM1824	178, 182, 188	182	178, 182	180, 182, 184

Notes. Char2– Shkavro et al., 2010; Char3-Czech population, Putnova et al., 2011; Char4-Mexican (import from France), Char5 – Mexican (import from France , Great Britain and Ireland), Sifuentes-Rincón et al., 2007; lower underscore-allele with the highest frequency

According to the BM1824 locus, the 182 bp allele is the most common in all populations. However, out of 5-8 alleles of BM1824, 3 remained in the Ukrainian population. a similar situation is observed for loci INRA023 (high frequency of occurrence of the INRA023^{206bp} allele and a decrease in variability from 10-13 alleles to 6), BM2113 (predominance of the BM2113^{131bp} allele, a decrease in the number of alleles from 7-9 to 5). The lowest genetic variability among all populations was observed for ETH10. Of the 6 known alleles of this locus, two ETH10^{217bp} and ETH10^{219bp} remained in the Ukrainian population and completely lost alleles, which were most common in the populations of French origin Char4 (ETH10^{215bp}) and Char4 (ETH10^{211bp}), as well as two alleles detected in the Czech Charolais population (ETH10^{221bp} and ETH10^{223bp}).

A significant decrease in the variability of these loci may be due to the peculiarities of reproductive and breeding management in the domestic Charolais population, which, according to Shkavro et al. (2010), for a long time bred "by itself" due to the lack of "blood flow", which usually occurs when purchasing new breeding bulls and using them in the herd reproduction system.

Conclusions

1. Data from the analysis of 10 microsatellite DNA markers in the population of cattle of the meat direction of productivity of the Charolais breed of Ukrainian selection are obtained. The proportion of polymorphic loci was 100 %. The number of detected alleles per locus ranged from 2 (ETH10) to 10 (TGLA053) and averaged 5 alleles per locus.

2. The average level of genetic variability for the set of microsatellite loci in the experimental population was established (H_e=0.6±0.05). High values of expected heterozygosity are characteristic of loci TGLA053 (H_e = 0.82) and INRA023 (H_e = 0.73),

while low values are characteristic of locus ETH10 (H_e = 0.20).

3. Most microsatellite loci are characterized by an equilibrium state in the distribution of genotype frequencies. The deviation was found only at the TGLA122 locus (χ^2 = 5.1; $p < 0.05$) caused by the excess of homozygous individuals (F_{is} = 0.29).

4. Most of the studied loci belong to informatively valuable markers (average value PIC =0.56 ± 0.05;), indicating the possibility of their use for certification, identification and confirmation of origin in the experimental animal population.

5. Analysis of genetic changes that occurred during the reproduction of experimental populations of the Charolaise breed in comparison with data from other regions of the world indicates a narrowing of genetic variability, which in the future may have negative consequences and requires replenishment of allelic diversity and control of genetic processes in breeding work using DNA markers.

References

Debrauwere, H., Gendrel, C., Lechat, S. Dutreix, M. (1997). Differences and similarities various tandem repeat sequences: minisatellites and microsatellites, *Biochimie*, Vol, 79, P, 577–586. https://doi.org/10.1016/S0300-9084(97)82006-8.

Dzitsiuk, V., Guzevatiy, O., Lytvynenko, T., & Guzeev, Y. (2020). Genetic polymorphism of buffalo Bubalus bubalis bubalis by cytogenetic and molecular markers. *Agricultural Science and Practice*, 7(1), 24-31. https://doi.org/10.15407/agrisp7.01.024.

FAO. (2011). Molecular genetic characterization of animal genetic resources. *FAO animal production and health guidelines*, No, 9, Rome, Italy, URL: http://www.fao.org/docrep/014/i2413e/i2413e00.pdf.

FAO/ISAG. (2004). Secondary Guidelines. Measurement of Domestic Animal Diversity (MoDAD): New recommended microsatellite markers. URL: http://dad.fao.org/en/refer/library/guidelin/marker.pdf.

Koskinen, M.T. (2006). Development of STR assays for identification and forensic testing. In: Proceedings of the 30th International Conference on Animal Genetics, 2006, Porto Seguro, Brazil. Belo Horizonte, Brazil: CBRA, 2006. W582: 21. https://www.isag.us/Docs/2006ISAG_Proceedings.pdf.

Kramarenko, A. (2019). Genetic structure of the Southern meat cattle breed based on microsatellite markers. *Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series: Agricultural Sciences*, 21(91), 21-28. https://doi.org/10.32718/nvlvet-a9104.

Kramarenko, A. S., Gladyr, E. A., Kramarenko, S. S., Pidpala, T.V., Strikha, L.A., Zinovieva, N. A. (2018). Genetic diversity and bottleneck analysis of the Red Steppe cattle based on microsatellite markers. *Ukrainian Journal of Ecology*, Vol, 8 (2), P. 12–17. https://www.researchgate.net/publication/324068904

Kulibaba, R. A., Liashenko, Y. V. (2016). Influence of the PCR artifacts on the genotyping efficiency by the microsatellite loci using native polyacrylamide gel electrophoresis. *Cytology and Genetics*, Vol, 50, № 3, P, 162–167. DOI: 10.3103/S0095452716030087.

Ladyka, V.I., Khmelnychyi, L.M., Lyashenko, Y.V., Kulibaba, R.O. (2019). Analysis of the genetic structure of a population of Lebedyn cattle by microsatellite markers, *Regulatory Mechanisms in Biosystems*, Vol, 10 (1), P, 45–49, DOI:10,15421/021907

Liashenko, Y. V., Kulibaba, R. A., Marchuk, V. S, Kulibaba, S. V. (2024). The *Scientific and Technical Bulletin of the Institute of Animal Science NAAS of Ukraine*. №132. 87-99. DOI 10.32900/2312-8402-2024-132-87-99.

Mishra, S. P., Mishra, C., Mishra, D. P., Rosalin, B. P., Bhuyan, C. (2017). Application

of advanced molecular marker technique for improvement of animal: A critical review, *Journal of Entomology and Zoology Studies*, Vol, 5 (5), P, 1283–1295. https://www.researchgate.net/publication/320298591.

Peakall, R., Smouse, P.E. (2012). GenAIEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research – an update. *Bioinformatics*. 28 (19): 2537– 2539. doi: 10.1093/bioinformatics/bts460.

Petrenko, S.M., Nosevych, D.K., Tokar, Yu.I., Uhnivenko, A.M. (2016). Naukovi osnovy rozvytku miasnoho skotarstva v Ukraini. *K.: KOMPRYNT*,330 c. (in Ukrainian) https://nubip.edu.ua/sites/default/files/u249/naukovi_osnovi_rozvitku_myasnogo_skotarstva_v_ukrayini.pdf.

Putnova, L., Vrtkova, I., Srubarova, P., & Stehlik, L. (2011). Utilization of a 17 microsatellites set for bovine traceability in Czech cattle populations. *Iranian Journal of Applied Animal Science*, 1(1), 31–37. https://sanad.iau.ir/fa/Journal/ijas/DownloadFile/1023224.

Al-Jubori, S.M. & Senkal, R.H. (2023). Genetic Diversity And Productive Performance In Local And Imported Iraqi Cows Using Microsatellite Markers. *The Iraqi Journal Of Agricultural Sciences*, 54(6):1538-1547. Doi:10.36103/Ijas.V54i6.1854.

Senan, S., Kizhakayil, D., Sasikumar, B., Sheeja, T. (2014). Methods for development of microsatellite markers: an overview. *NotSciBiol*, Vol, 6 (1) P, 1–13. DOI: https://doi.org/10.15835/nsb619199.

Shel'ov, A. V. (2015). Polimorfizm mikrosatelitnykh lokusiv DNK u riznykh vydiv sil's'kohospodars'kykh tvaryn [Polymorphism of microsatellite DNA loci in different species of farm animals]. *Animal Breeding and Genetics*, 50, 183–190 (in Ukrainian). http://nbuv.gov.ua/UJRN/rgt_2015_50_28

Shel'ov, A. V., Kopylov, K. V., Kramarenko, S. S., & Kramarenko, O. S. (2017). Analysis of population-genetic processes in different cattle breeds by microsatellite loci of DNA. Agricultural Science and Practice, 4(1), 74–78. DOI: https://doi.org/10.15407/agrisp4.01.074

Shkavro, N. M., Radko, A., Slota, E., & Rossokha, V. I. (2010). Polimorfizm mikrosatelitnykh markeriv DNK dvokh porid velykoyi rohatoyi khudoby [Polymorphism of microsatellite DNA markers two breeds of cattle]. *Visnyk Kharkivs'koho Natsional'noho Universytetu imeni V. N. Karazina*. Seriya Biolohiya, 905(11), 120–126 http://nbuv.gov.ua/UJRN/VKhb_2010_905_11_19. (in Ukrainian).

Shkavro, N., Blyzniuk, O., Pomitun, I., & Babicz, M. (2018). Evaluation of the genetic structure and main productive traits of Lebedyn cattle based on genetic markers polymorphism. *Journal of Animal Science Biology and Bioeconomy*, 36(2), 17–26. DOI: 10.24326/jasbbx.2018.2.2.

Sifuentes-Rincón, A.M., Puentes-Montiel, H., & ParraBracamonte, G.M. (2007). Assessment of genetic structure in Mexican Charolais herds using microsatellite markers. *Electronic Journal of Biotechnology*, 10(4), 492–499. doi: 10.4067/S0717- 34582007000400002.

Snegin, E. A., Kramarenko, A. S., Snegina, E. A., & Kramarenko, S. S. (2019). Evaluation of genetic diversity and relationships among eight Russian and Ukrainian cattle breeds based on microsatellite markers. *Regulatory Mechanisms in Biosystems*, 10(4), 388-393. https://doi.org/10.15421/021958.

Zhao, J., Zhu, C., Xu, Z., Jiang, X., Yang, S., & Chen, A. (2017). Microsatellite markers for animal identification and meat traceability of six beef cattle breeds in the Chinese market. Food Control, 78, 469–475. doi:10.1016/j.foodcont.2017.03.017.

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FEASIBILITY OF USING PROBIOTICS FOR BEES

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The main mechanism of action of probiotics is to normalize the composition of the biological microflora of the gastrointestinal tract, that is, its colonization by competitive strains of bacteria-probionts, which carry out non-specific control over the number of conditionally pathogenic microflora by displacing it from the intestinal biocenosis, strengthening the barrier functions of the intestinal mucosa in bees, and also activates the synthesis of hemolymph cells, while stimulating digestion and strengthening the immune status of the body.

The article presents the results of studies of the antimicrobial properties of Bilact and Enteronormin preparations against putrefactive pathogens (Penibacillus larvae, Melisococcus pluton) in the laboratory. Their influence on the factors of non-specific resistance of bees is determined.. Experimental studies to verify antimicrobial properties were performed by Agar diffusion. The criterion for evaluating effectiveness was the size of the growth retardation zone (mm). To determine the effect of the drugs "Bilact" and "Enteronormin" on the body of adult bees, studies were conducted in bee colonies. Families of the I-th experimental group were added to the feed of the drug "Bilakt", the II-th group – the drug "Enteronormin", control – pure sugar syrup (1:2). Hemolymph was taken from bees before the experiment. During the experiment, hemolymph samples were taken after 7, 14, and 21 days and the activity of lysozyme, phagocytosis, and hemolymph bactericide were determined.

It was found that the experimental strains Penibacillus larvae and Melisococcus pluton were sensitive to both microbiological preparations: the growth retardation zone for crops even at the lowest concentration of 1.0 x 103 Kou/cm3 exceeded 15 mm. It was found that the preparations "Bilakt" and "Enterohormin" showed approximately the same effectiveness in relation to pathogens of bee brood rot. Both drugs at a maximum concentration of 1 x 109 Kou/cm3 caused growth retardation of both putrefactive pathogens in 24.9±0.71 mm and 24.5±0.51 mm (Bilact) and 23.8±0.8 mm and 24.2±0.84 mm (Enteronormin).

It was found that the activity of lysozyme in the hemolymph of bees of Group I was 1.4 times higher 7 days after feeding a mixture of sugar syrup with Bilact 21 days later – 1.7 times higher than in the control. The activity of lysozyme in the hemolymph of bees of Group II was 1.5 times higher 7 days after feeding the drug "Enteronormin", after 21 days – 1.7 times more than in the control. Lysozyme activity in the hemolymph of bees of groups I and II 7 days after the end of top dressing was increased by 43.8% and 45.7%, respectively, compared to the control.

Differences in the indicators of bactericidal activity of hemolymph indicators before and after the use of probiotics were revealed. The results obtained indicate that feeding the preparations "Bilakt" and "Enteronormin" contributes to an increase in the factors of non – specific resistance of the Bee body (lysozyme activity by 1.4, 1.7 times, phagocytosis by 1.1 times, bactericidal factor of hemolymph-by 2.0 times).

Keywords: bees, pathogens of bacterial diseases, probiotics, hemolymph, humoral and cellular immune factors.

ДОЦІЛЬНІСТЬ ЗАСТОСУВАННЯ ПРОБІОТИКІВ ДЛЯ БДЖІЛ

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Основний механізм дії пробіотиків полягає в нормалізації складу біологічної мікрофлори шлунково-кишкового тракту, тобто його заселенні конкурентоспроможними штамами бактерій-пробіонтів, які здійснюють неспецифічний контроль над чисельністю умовно-патогенної мікрофлори шляхом витіснення її з кишкового біоценозу, посиленні бар'єрних функцій слизової оболонки кишечнику у бджіл, а також активізує процеси синтезу клітин гемолімфи, при цьому стимулюється травлення та посилюється імунний статус організму.

У статті викладено результати досліджень антимікробних властивостей препаратів «Білакт» та «Ентеронормін» щодо збудників гнильців (Penibacillus larvae, Melisococcus pluton) у лабораторних умовах. Визначено вплив їх на фактори неспецифічної резистентності бджіл. Експериментальні дослідження щодо перевірки антимікробних властивостей проводили методом дифузії в агар. Критерієм оцінки ефективності була величина зони затримки росту (мм). Для визначення впливу препаратів «Білакт» та «Ентеронормін» на організм імаго бджіл дослідження проводили у бджолиних сім'ях. Сім'ям І-ї дослідної групи додавали в корм препарат «Білакт», ІІ-ї групи – препарат «Ентеронормін», контрольним – чистий цукровий сироп (1:2). У бджіл перед дослідом відбирали гемолімфу. В процесі досліду зразки гемолімфи відбирали через 7, 14, 21 добу та визначали активність лізоциму, фагоцитозу та бактерицидності гемолімфи.

Встановлено, що дослідні штами Penibacillus larvae та Melisococcus pluton виявились чутливими до обох мікробіологічних препаратів: зона затримки росту для культур навіть у найменшій концентрації 1,0 х 103 КОУ/см3 перевищила 15 мм. Встановлено, що препарати «Білакт» та «Ентерогормін» проявили приблизно однакову ефективність по відношенню до збудників гнильців розплоду бджіл. Обидва препарати у максимальній концентрації 1 х 109 КОУ/см3 викликали затримку росту обох збудників гнильців у 24,9±0,71 мм і 24,5±0,51 мм (Білакт) та 23,8±0,8 мм і 24,2±0,84 мм (Ентеронормін).

Встановлено, що активність лізоциму у гемолімфі бджіл І групи була більше в 1,4 рази через 7 діб після згодовування суміші цукрового сиропу з препаратом «Білакт» через 21 добу – в 1,7 рази більше ніж у контролі. Активність лізоциму у гемолімфі бджіл ІІ групи була більше в 1,5 рази через 7 діб після згодовування препарату «Ентеронормін», через 21 добу – в 1,7 рази більше ніж у контролі. Активність лізоциму у гемолімфі бджіл І та ІІ групи через 7 діб після закінчення підгодівлі була більше на 43,8 % та 45,7 % відповідно в порівнянні з контролем.

Виявлено відмінності показників бактерицидної активності показників гемолімфи до і після застосування пробіотиків. Отримані результати вказують на те, що згодовування препаратів «Білакт» та «Ентеронормін» сприяє підвищенню факторів неспецифічної резистентності організму бджіл (активності лізоциму в 1,4, 1,7 рази, фагоцитозу в 1,1 рази, бактерицидного фактору гемолімфи – в 2,0 рази).

Ключові слова: бджоли, збудники бактеріальних хвороб, пробіотики, гемолімфа, гуморальні та клітинні фактори імунітету.

The environment, honeybees and their microflora are the only ecological system that responds to any changes. In the normal physiological state of bees, the relationship between the body and microflora is generally symbiotic, which was formed and fixed in the process of evolutionary development. Nectar, pollen, and water enter the hive from the environment (DeGruttola, A. K. et al., 2016; Glenny, W. et al., 2017; Vagner de Alencar Arnaut de Toledo et al., 2020). However, it is important to know the diversity of microorganisms in the environment – hive biocenosis, especially due to the spread of pathogens that pollute bee products and the bee nest in the hive (fig. 1).

The figure schematically shows how parasites (varroa mites, Acarapis), protozoa (Nosema spp. Malpighamoeba mellificae), pathogenic viruses (black queen bee (BQCV), deformed wings (DWV), Kashmir Bee (KBV), sac-like brood (SBV), acute and chronic paralysis (ABPV, CBPV), Israel acute paralysis (IAPV) slow paralysis (SPV)) enter the middle of the nest and are distributed throughout it.

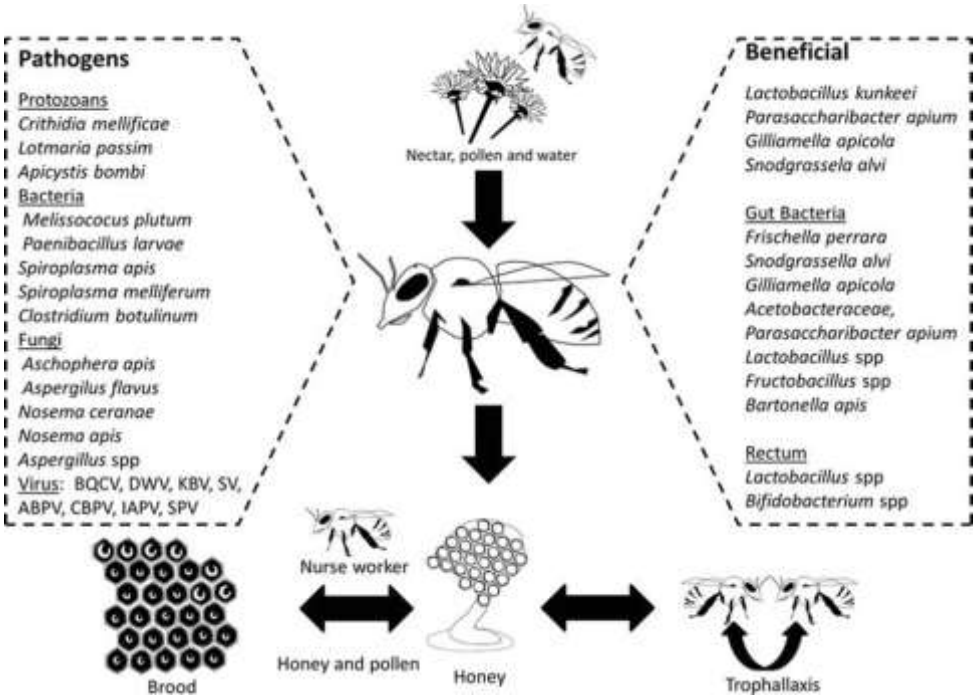

Fig. 1. **Pathogens and beneficial microorganisms in honeybees: one of the ways – bee food infection occurs through nectar, pollen (on flowers) and water collected by worker bees in the environment; the second – food is stored in the Hive and can be transmitted by trophalaxis between workers and brood; the third – is the consumption of infected honey and/or pollen from other hives by honeybees (based on materials: Honey Anaalysis. New Advances and Challennges. Edited by Vagner de Alencar Arnaut de Toledo, Emerson Dechechi Chambo (2020).**

There is also a niche of beneficial microbiota in the digestive tract (anterior, middle, and posterior intestines). Arrows indicate the transfer of microorganisms internally by hive bees by food route between individuals(brood and Imago), honey reserves, and parchment in the hive (Vagner De Alencar Arnaut de Toledo, 2020; Daisley, B., et al.2020).

Beneficial microorganisms can secretly remain in them, waiting for the moment when the environment becomes suitable for their development. Among the beneficial

bacteria found in bee products, we can mention some bacteria that act as prebiotics when ingested (Paytuví-Gallart, A. Et al., 2020; Tushak, S., 2018).

However, in modern conditions, insects are exposed to a whole range of adverse factors that affect the normal functioning of the main vital systems: on the one hand, the deterioration of the environmental situation, an increase in the number of stressful situations due to insufficient quantity and variety of pollen and honeybees, and on the other - the mass uncontrolled use of chemotherapeutic drugs, both for plants and bees. A significant level of microbial contamination of feed and environmental objects leads to advanced colonization of the insect gut by pathogenic microorganisms, which slows down and even prevents the formation of normal intestinal microflora (Piccini C. et al,, 2004; Kotsiumbas I. Ya. et al,, 2013, Kalinichenko S. V et al., 2013, Ogrodowczyk, A. M. et al., 2020).

The increase in output and growth rates of beekeeping products may be a natural consequence of the introduction of new technologies in the fields of microbiology and biochemistry, in particular in the correction of biocenoses of the gastrointestinal tract of bees-the artificial introduction of representatives of beneficial microflora into the hive. They are called probiotics, the main purpose of which is the formation of a metabolically active population of probiotic bacteria in the digestive tract, which contributes to a qualitative change in the composition of intestinal microflora and the displacement of pathogenic microorganisms. In addition, the use of probiotics promotes the synthesis of enzymes and other biologically active substances, in particular vitamins, bacteriocins, etc. (Kalinichenko S. V. et al, 2013).

Probiotics are living microorganisms that have a positive effect on bee health through anti-infective defense mechanisms, immunomodulatory effects, increased barrier functions, metabolic effects and positive effects on intestinal motility and function.

According to the form of release, probiotics are divided into two groups - liquid and dry. Dry probiotics are freeze-dried microorganisms that can be found in powder, capsules, or tablets. The shelf life of dry preparations is longer than that of liquid ones, in addition, they are less dependent on environmental conditions and, thus, do not require strict compliance with storage criteria, and are also much more convenient to transport. The disadvantage of dry probiotics is that during lyophilization, bacteria lose some of their useful properties, and after using the Drug, time is needed for the transition of bacteria from suspended animation to the active form and the onset of action (Kalinichenko S. V. et al, 2013).

Liquid probiotics are bacteria "with an active life position", that is, they retain all their valuable properties and begin to act immediately after entering the body. However, firstly, this requires strict compliance with the shelf life conditions, and secondly, the shelf life of these drugs is shorter than lyophilized analogues-no more than three months. Liquid probiotics can consist not only of bacteria that are in a physiologically active state, but also of a special nutrient medium that serves as a source of nutrition for them; additionally introduced ingredients that enhance the effectiveness of the drug (water - soluble vitamins, micro-and macronutrients, amino acids, etc.); metabolites (bacterial waste products).

The most dangerous infectious diseases of bee brood are: American (malignant) rot, the causative agent of which is the spore-forming Rod Penibacillus larvae larvae, European (benign) rot, the main causative agent of which is Melisococcus pluton and others. Parasitic diseases (varooz and acarapidosis), diseases that cause protozoa (Nosema spp., Malpighamoeba mellificae), as well as pathogenic mold fungi and yeast weaken the insect body. Rot pathogens do not have the same pathogenicity and virulence, which largely depends on their concentration: the higher it is, the more often outbreaks of the

disease occur and the more affected families (Glinski, Z et al., 2001; Piccini C. et al., 2004, Kutsan O. T. et al. 2013; Daisley, B., et al.2020;).

Factors of non-specific resistance of bees include cells of the hemolymph, body fat (cellular immunity), as well as antimicrobial fluids (humoral immunity) of the body. Hemolymph performs protective functions due to the presence of such cells and fluids in it. It is characteristic that the activity of cellular defense factors depends on humoral ones. The process of phagocytosis is largely activated by lysozyme, microcins and opsonine. They increase the rate of phagocytosis. Lysozyme, which is adsorbed on the mucopepid cell wall of the microorganism, breaks it down. As a result, the osmotic balance is disturbed and hydrolysis of the microbial cell begins (Glinski, Z et al., 2001).

The cellular basis of the general mechanisms of non – specific resistance of insects is the antimicrobial activity of the hemolymph, which is associated with the functions of its shaped elements-hemocytes, including phagocytes, which create prerequisites for activating the synthesis of antibacterial proteins. Morphological heterogeneity of hemocytes determines the variety of associated protective processes in the insect body. Changes in the number, ratio of shaped elements and biochemical parameters of bee hemolymph are noted both depending on the season, functional and age groups, and under the influence of pathogens of various diseases and the means of their elimination used (Szymaś B. et al., 2003; Larsen, A. et al., 2019; Danihlík, J. et al.,2018).

Purpose of research. To study the antimicrobial properties of probiotic drugs: "Bilaktu", which includes Lactobacillus spp. and Bifidobacterium; dry - "Enteronormin", containing Enterococcus spp in its composition., Lactobacillus spp., Bacillus spp. on pathogens of rot (Penibacillus larvae, Melisococcus pluton) in the laboratory. To study the effect of these drugs on humoral and cellular factors of non-specific resistance of bees: bactericidal and biochemical parameters of the hemolymph in general, lysozyme activity in particular, as well as the cellular composition of the hemolymph.

Materials and methods. Epizootic strains of pathogens of American rot – Penibacillus larvae, European rot – Melisococcus pluton were used in experiments, in the form of a suspension of 3-day cultures of vegetative cells in the amount of 1.0 × 106 PFU/cm^3, the sensitivity of pathogens of rot to drugs was determined: "Bilact" is a liquid composition of bacteria of the genus Lactobacillus spp. and Bifidobacterium; "Enteronormin" is a dry-form compound with Enterococcus spp., Lactobacillus spp., Bacillus spp.

The preparations were used in concentrations: 1.0 × 103, 1.0 × 106, 1.0 × 109 PFU/cm^3. Saline solution of 0.9% was used as a control. The study was performed by Agar diffusion (Labynskaya, A. S., 1978). Two petri dishes were used in parallel for each drug.

At the first stage, the lower layer of Agar was poured into sterile petri dishes (for the causative agent of American rot – Willis–HoBZ medium, for European rot – skull) in the amount of 10 cm^3, left for 30 minutes. for solidification. Three cylinders (10 mm high and 8 mm in diameter) were placed on the surface of the lower layer at a distance of 4 and 2.5 mm from each other and from the edge of the Cup, respectively. Then the top layer of selective Agar (for each pathogen) was poured in an amount of 10 cm3 into Perti cups, evenly distributed over the surface and allowed to harden. At the second stage, 1 cm^3 of suspensions of three–day penibacillus larvae culture was applied to the surface of Willis – HoBZ Agar, and Melisococcus pluton culture was applied to the surface of Skull Agar and evenly distributed over the agar surface. After 30 minutes. excess suspensions were removed by sucking out a Pasteur pipette, and the cylinders were removed with tweezers. Each well was numbered clockwise on the reverse side of the cup and 0.5 cm^3 of the test drug was added at the above concentrations (well volume). Petri dishes were

kept in a thermostat at a temperature of 37 (±1) °C for (48-120) hours. (depending on the type of pathogen). The growth retardation zone of the microorganism was determined in mm by measuring through the center of the well diameter. The experiment was repeated 3 times, the results were processed statistically.

The criterion for evaluating the effectiveness of a probiotic (quantitative indicator) was the size of the growth retardation zone: – up to 10 mm – the strain was considered insensitive; – from 11 mm to 15 mm – insensitive; – from 15 mm to 25 mm – sensitive; – more than 25 mm – highly sensitive.

To determine the effect of the drugs "Bilact" and "Enteronormin" on the body of adult bees, studies were conducted in bee colonies. For this purpose, two experimental and one control groups of bees were formed, ten families each. Families of the I-th experimental group were added to sugar syrup the drug "Bilakt", the II-th group – the drug "Enteronormin", the control group was fed pure sugar syrup (1:2). Hemolymph was taken from bees before the experiment. During the experiment, hemolymph samples were taken after 7, 14, 21 days and lysozyme activity, bactericidal and biochemical parameters of hemolymph, as well as the species and quantitative composition of hemolymph cells were determined.

The bactericidal activity of hemolymph was studied by diffusion into Agar. As test crops, pathogenic microorganisms for bees were used-pathogens of rot: American, European, and non – pathogenic – Escherichia coli. On the surface of agar, a culture of the microorganism at a concentration of 1 billion tons was introduced into a petri dish. cells / cm^3. The cups were kept in a thermostat at 37 °C for 2 hours. With a marker, holes with a diameter of (3-4) mm were made, into which hemolymph samples were introduced. The reaction results were recorded after 3, 6, 12, and 24 hours. (Labynskaya, A. S., 1978). Determination of lysozyme activity in the hemolymph was performed by turbidimetric method. Micrococcus lysodeikticus culture was prepared on phosphate buffer, and combined hemolymph samples were diluted 10 times with saline. Experimental and control samples were examined simultaneously. The amount of lysozyme in the hemolymph sample was calculated using the calibration curve in mcg/ml (Labynskaya, A. S., 1978).

Laboratory methods of research in biology were used to determine the biochemical parameters of protein metabolism in the hemolymph of bees (Vlizlo et al., 2012).

Smears were prepared from the hemolymph, fixed with methyl alcohol and stained with Azur-eosin. After washing and drying, the smears were microscopized at magnification (X900) using a Biolam microscope. 100 cells were counted in a single smear, assessing the morphological composition of bee hemolymph (Errapcaliu et al., 2009; Barakat et al., 2016).

Research results and discussion. It is known from the literature that probiotics are multifactorial therapeutic agents by their mechanism of action. They exhibit antagonistic activity against a wide range of pathogenic and opportunistic microorganisms, have a corrective effect on the biocenosis and stimulate reparative processes in the intestine, activate the body's defenses, improve metabolism, and affect non-specific factors of bee immunity (Brumfitt W. et al, 2002; Kalinichenko S. V. et al, 2013).

Analyzing our results, it should be noted that the experimental strains Penibacillus larvae and Melisococcus pluton were sensitive to both microbiological preparations: the growth retardation zone for crops even at the lowest concentration of 1.0 x 103 PFU /cm^3 exceeded 15 mm (table. 1).

Table 1

Sensitivity of probiotic rot pathogens (Agar diffusion method)

Drug name concentration,	Concentration, PFU / cm³	growth retardation zone, mm	
		P. larvae	***M. pluton***
«Bilakt»	$1,0 \times 10^3$	16,9±0,62	17,1±0,7
	$1,0 \times 10^6$	20,8±0,4	20,5±0,52
	$1,0 \times 10^9$	24,9±0,71	24,5±0,51
«Enteronormin»	$1,0 \times 10^3$	20,2±58	20,9±0,47
	$1,0 \times 10^6$	21,4±0,56	21,8±0,46
	$1,0 \times 10^9$	23,8±0,8	24,2±0,84
Control (physical solution)	0,9	–	–

Note: " – " - no growth retardation zone

Thus, when comparing the antimicrobial effect of the drugs "Bilakt" and "Enterohormin", it was found that they showed approximately the same effectiveness in relation to pathogens of bee brood rot. Both drugs at a maximum concentration of 1 x 109 PFU /cm^3 caused growth retardation of both putrefactive pathogens in 24.9±0.71 mm and 24.5±0.51 mm (Bilact) and 23.8±0.8 mm and 24.2±0.84 mm (Enteronormin), respectively.

Beneficial microorganisms increase the bactericidal effect of bee hemolymph due to the accumulation of lactic acid or the formation of a significant number of specific metabolic products (peptides, carbonyl compounds, hydrogen peroxide, etc.), which have antibacterial properties. Bactericidal and biochemical parameters of bee hemolymph were determined in samples taken before the start of the experiment and 21 days after the end of top dressing. The research results are shown in Table 2.

Table 2

Bactericidal and biochemical parameters of bee hemolymph

Bee groups / Day (n=10)		hemolymph parameters			
		Bactericidal factor, h.	**Total Protein, g / l**	**Total nitrogen, mg %**	**residual nitrogen, mg%**
Before feeding with probiotics		6	48,4±1,3	575,0±10,2	412,0±35,5
Top Dressing "Bilakt"	21	12	63,6±4,2	787,0±50,8	289,0±12,2
Top Dressing "Enteronormin"	21	12	65,0±3,1	808,0±56,3	268,9±12,2
Control (sugar syrup)	21	6	52,4±3,1	568,4±65,0	345,0±34,2

The study of the bactericidal factor of bee hemolymph showed that it did not differ in individuals who received top dressing with probiotics. Thus, it was found that hemolymph delayed the growth of cultures of pathogens of American rot for 12 hours and exceeded this indicator twice compared to the control, where sugar syrup was fed without any impurities and the initial indicator (before the experiment). The study of the bactericidal factor of bee hemolymph showed that it did not differ in individuals who received top dressing with probiotics.

Thus, it was found that hemolymph delayed the growth of cultures of pathogens of American rot for 12 hours and exceeded this indicator twice compared to the control, where sugar syrup was fed without any impurities and the initial indicator (before the experiment). Indicators of protein metabolism also indicate a positive effect of probiotics on the body of bees. A significant increase in the content of total protein and nitrogen in the hemolymph of bees fed probiotic preparations was established against the background of a decrease in the amount of residual nitrogen. On Day 21, the total protein index increased by 31.4% when feeding "Bilaktu" and 34.3 % – "Enteronormin", total nitrogen increased by 36.9%, respectively; and 40.5%, residual nitrogen decreased by 29.9% and 34.9%, respectively.

In the process of interaction of representatives of the beneficial flora, which was a component of probiotic agents and pathogenic microbes, which was located in the Bee cattery, the death and destruction of pathogen cells and parts of beneficial microorganisms occurred. At the same time, lysozyme was released from the bacterial wall, which enhanced the effect of its own from the hemolymph of bees.

When studying the effect of the drugs "Bilact" and "Enteronormin" on the body of bees, an increase in the activity of hemolymph lysozyme was found (table. 3).

Table 3

Activity of bee hemolymph lysozyme

Selection period, day	Group of bees. Lysozyme activity, mcg / ml(n=10)		
	I experienced «Bilakt»	II experimental “Enteronormin”	Control (sugar syrup)
Before the start of the experiment	36,4 ± 1,5		
7	51,9 ± 3,8[1)]	52,6±0,3[1)]	36,1 ± 1,4
14	54,2 ± 4,1[1)]	57,7 ± 0,8[1)]	36,4 ± 1,5
21	60,7 ± 4,1[1)]	62,7±1,2[1)]	36,2 ± 1,5

Note: 1) – the difference in results is likely compared to the results before the start of top dressing and the control group, $p<0.05$.

From the data in Table 3, it can be seen that the activity of lysozyme in the hemolymph of bees of groups I and II 7 days after the end of top dressing was higher by 43.8% and 45.7%, respectively, compared to the control.

After 21 days, this difference was 66.7% for the first group, 73.2% for the second group compared to the control group of bee families. Lysozyme activity in bees from Control families did not increase during the experiment.

The activity of certain types of hemocytes forms an immune response to the entry of a significant number of microorganisms into the insect body (Gábor et al., 2020). To interpret the ability of certain agents to have any effect on non-hemolymph of bees, it is important to fix the morphological (species) and quantitative composition of the latter's cells. The effect of probiotics "Bilakt" and "Enteronormin" in a field experiment on ten bee colonies was evaluated by manufacturing and microscopy of hemolymph smears: before the experiment; when using probiotics for feeding bees with sugar syrup after 7, 14, 21 days. Differentiation of hemocytes was based on their morphological and quantitative characteristics (Barakat et al., 2016; Jazlovitskaya et al., 2014; Richardson et al., 2018).

The results obtained indicate that feeding Bilact and Enteronormin preparations increases the factors of non – specific resistance of the Bee body (lysozyme activity by 1.4, 1.7 times, hemolymph bactericidal factor-by 2.0 times).

It is known that feeding bees with sugar syrup is a natural technological technique used during the period when there are no bribes in the environment and bees do not have enough food for the existence and development of families. In addition, it is sugar syrup that is most often the carrier of drugs that are used for preventive and curative purposes (Ptaszyńska et al., 2016; Saranchuk et al., 2021; Frizzera et al., 2020). The results of studying the quantitative and morphological characteristics of bee hemolymph cells on the Slave are shown in Table 4.

Table 4

Quantitative and morphological composition of hemocytes in bee hemolymph

Bee groups / day of experience (n=10)		Hemocyte classes				
		Proleuko-cytes	Phagocytes		Spherulo-cytes	Enocytoids
			neutrophilic	eosinophilic		
Before top dressing		14,46±1,33	30,62±0,29	22,0±0,79	25,93±1,2	3,0±0,37
Top Dressing "Bilakt"	7	15,00±2,69	33,0±5,26	24,2±1,08	25,8±4,38	3,0±0,73
	14	15,40±0,45	32,4±0,45	24,12±1,32	25,0±0,93	3,0±1,0
	21	15,62±0,82	31,25±1,45	23,2±4,18	26,00±1,38	3,0±0,35
Top Dressing "Enteronor min"	7	15,83±0,88	35,62±1,01	26,37±1,28	26,83±0,73	2,66±0,68
	14	16,20±1,88	35,0±2,89	25,60±3,05	27,60±2,14	3,0±0,94
	21	16,0±0,65	34,50±4,48	24,0±3,79	27,87±0,66	2,87±0,38
Control (sugar syrup)	7	14,50±2,49	31,20±1,71	22,33±0,85	24,0±3,68	3±0,65
	14	14,00±1,27	32,33±0,94	22,62±0,65	23,60±2,56	1,80±0,65
	21	13,62±0,98	33,53±1,45	23,06±2,22	22,75±0,76	1,62±0,29

When microscopy smears of individual hemolymph samples at the beginning of the experiment (before feeding the drugs), the quantitative indicators of hemolymph were: prodrug cells – 14.46%, phagocytes -56.59%, of which neutrophils – 33.53%, eosinophilic – 23.06%, spherulocytes – 25.93%, enocytoids – 3.0 %.

Comparison of the indicators of hemolymph smears that were examined before the start of feeding and after 21 days during feeding with sugar syrup indicates a decrease in proleukocytes by 0.84%, secretory cells (spherulocytes – 3.18, enocytoids – 1.38 %); an increase in phagocytic cells – neutrophils 2.91 %; which indicates the aging of bees and an increase in bacteria in their body, which, in turn, stimulate the growth of phagocytic function to maintain the body's homeostasis, eosinophils 1.06%, respectively.

Comparison of the results of microscopy of hemolymph smears of the control group on days 7, 14 and 21 with the groups receiving probiotics "Bilact" and "Enteronormin" showed a decrease in neutrophilic and eosinophilic phagocytes and an increase in proleukocytes and spherulocytes. This may indicate a positive trend in the hemolymph for the use of drugs and serve as confirmation of the immunostimulating and activating effect.

The dynamics of a decrease in the number of neutrophilic and eosinophilic phagocytes was also observed, especially in comparison with the control. A more pronounced decrease in phagocytic cells was observed in the group in which

Enteronormin was used. Such results indicate a high activity of probiotic drugs, the destruction of foreign substances in the hemolymph and the stabilization of cellular immunity. Analyzing the data, it should be noted that the drugs in sugar syrup have a more active effect on inactivating foreign inclusions and stabilizing immune processes.

Internal protection is provided by cellular and humoral factors. In (most) higher animals, two fluids circulate in the body: blood, which performs a respiratory function, and lymph, which performs mainly the function of delivering nutrients. The blood of insects differs significantly from the blood of higher organisms and therefore received a special name – hemolymph. It is the only tissue fluid in the body of insects. Like vertebrate blood, it consists of a liquid intercellular fluid – plasma and the cells that are in it – hemocytes. Unlike vertebrate blood, hemolymph does not contain cells that contain hemoglobin or other respiratory pigment. As a result, the hemolymph does not perform a respiratory function. All tissues and cells take the nutrients and other substances they need from the hemolymph and release metabolic products into it. Hemolymph transports digestive products from the intestinal walls to all organs, and waste products – to the excretory organs.

The composition of the hemolymph as the internal environment of the insect body is one of the criteria that characterizes the physiological or pathological state of the honey bee. The cellular basis of the general mechanisms of non – specific resistance of insects is the antimicrobial activity of the hemolymph, which is associated with the functions of its shaped elements-phagocytes, which create prerequisites for activating the synthesis of antibacterial proteins (lysozyme, Agglutinins, apidacins, abecins) in the cells of the fat body of bees and their entry into the plasma of the hemolymph. Proleukocytes are known to be precursors of all other classes of hemocytes. Neutrophilic and eosinophilic phagocytes appear in those that perform a microphagocytic function. Enocytoids have a secretory function. Spherulocytes perform protective and secretory functions.

Each type is an independent group of hemocytes that are not related to each other by origin and do not have morphological transitions. Due to transformations, hemolymph cells located in different morphological positions can perform different functions. Usually, each type of Hemocyte accumulates in the maximum amount during a certain period of life. The number of hemocytes in the hemolymph decreases especially sharply from the 10th day of life of bees.

With age, the number of young forms of hemocytes decreases, and mature ones increase. Any stage of bee development, its age and physiological state is characterized by a specific hemogram that reflects the percentage of different types of hemocytes. According to the hemogram, it is easy to determine physiological changes in the insect's body. It can be used to determine the “fatness” of an insect and diagnose diseases in the early stages of the disease, parasite infestation, and insecticide poisoning.

The insect's hemolymph washes all internal organs and is the environment in which all cells of the Bee's body live and function. It performs a number of functions, including protective. This function involves plasma proteins, hemocytes that have the ability to phagocytosis, and cells that form Hemocyte capsules around multicellular parasites. Hemocytes also have the ability to accumulate in places where damage to the body has occurred, forming a kind of plug that closes the wound. In this case, hemocytes multiply and Dead Cells phagocytosis occurs.

Maintaining the immunity of bees is an important link for the Prevention of infectious diseases. In particular, the cellular mechanisms of the immune defense of Apis mellifera are responsible for a number of protective barriers that promote the destruction of foreign agents and ensure the ability of phagocytic cells to respond by lysis and phagocytosis reactions to the penetration of bacterial pathogens or cause their uptake for

neutralization (Larsen et al., 2019). However, it should be noted that the activity of phagocytic neutrophils of the hemolymph in laboratory conditions is slightly lower than in the conditions of natural existence. Moreover, in bee colonies living in hives, there is a phenomenon of so-called social immunity, in which the bees of a particular colony are able to orally transfer immunological compounds between the members of the hive (Harwood et al., 2021). This type of immune metabolism is probably activated in the event of increased resistance of each individual Bee from the insect colony (synthesis of certain antibodies). Our results indicate that it is advisable to use pro biotic preparations for feeding bees, either with sugar syrup or in the form of Kandy as a stimulant to increase the resistance of bee colonies and maintain protective properties at the proper level.

Conclusion

1. Based on the above, we can conclude that among the scientific and practical directions, the development of modern methods for studying the composition and activity of insect microbiocenoses is relevant; detailing the molecular, biochemical and other mechanisms of action of probiotics for their effective use in the prevention and treatment of various diseases, an in-depth assessment of the safety of drugs containing probiotic strains.

2. Thus, there is no doubt that the scientifically based use of probiotics is an important method for preserving and restoring bee health.

References

Barakat, E.M., AboKersh, M.O., Gomaa, S.A. (2016). Haemocyte Activity and Cellular Defense Reactions in Various Larval Instars of Honey Bee (Apis mellifera L.) following. Natural and Experimental Bacterial Infections. Greener Journal of Biological Sciences, 6 (2): 020-033. http://doi.org/10.15580/GJBS.2016.2.012016017.

Brumfitt W. Salton M., Hamilton-Miller J. (2002).Nisin, alone and combined with peptidoglycan-modulating antibiotics: activity against methicillin-resistant Staphylococcus aureus and vancomycin-resistant enterococci // *Journal of Antimicrobial Chemotherapy*. — Vol. 50, № 5. – P. 731–734.

Daisley, B., Pitek, A., Chmiel, J., Al, K., Chernyshova, A., Faragalla, K., Burton, J., Thompson, G., Reid, G. (2020). Novel probiotic approach to counter Paenibacillus larvae infection in honey bees. The ISME journal, 14(2), 476-491. doi: 10.338/s41396-019-0541-6

Danihlík, J., Aronstein, K., & Petřivalský, M. (2015). Antimicrobial peptides: a key component of honey bee innate immunity: Physiology, biochemistry, and chemical ecology. Journal of Apicultural Research, 54(2), 123-136. doi: 10.1080/00218839.2015.1109919 31.

DeGruttola, A. K., Low, D., Mizoguchi, A., & Mizoguchi, E. (2016). Current understanding of dysbiosis in disease in human and animal models. Inflammatory bowel diseases, 22(5), 1137-1150. https://doi.org/10.1097/MIB.0000000000000750

Frizzera, D., Del Fabbro, S., Ortis, G., Zanni, V., Bortolomeazzi, R., Nazzi, F., & Annoscia, D. (2020). Possible side effects of sugar supplementary nutrition on honey bee health. Apidologie, 51(4), 594-608. doi: 10.1007/s13592-020-00745-6.

Gábor, E., Cinege, G., Csordás, G., Rusvai, M., Honti, V., Kolics, B., Török, T., Williams, M., Kurucz, É. & Andó, I. (2020). Identification of reference markers for characterizing honey bee (Apis mellifera) hemocyte classes. Developmental & Comparative Immunology, 109, 103701. doi: 10.1016/j.dci.2020.103701

Glenny, W., Cavigli, I., Daughenbaugh, K. F., Radford, R., Kegley, S. E., & Flenniken, M. L. (2017). Honey bee (Apis mellifera) colony health and pathogen composition in migratory beekeeping operations involved in California almond pollination. PloS one, 12(8). doi: 10.1371/journal.pone.0182814

Glinski, Z., Jarosz Z. (2001). Infektion and immunity in the honey bee Apis mellifera [Text] // Apiakta. Vol. 36, №1. P. 12–24.

Harwood, G., Salmela, H., Freitak, D., & Amdam, G. (2021). Social immunity in honey bees: royal jelly as a vehicle in transferring bacterial pathogen fragments between nestmates. Journal of Experimental Biology, 224(7). doi: 10.1242/jeb.231076.

Honey Anaalysis. New Advances and Challennges (2020) / Edited by Vagner de Alencar Arnaut de Toledo, Emerson Dechechi Chambo Intechopen (July 15, 2020) ISBN-13: 978-1789851199. 100 P.

Hussain, M. B. (2018). Role of honey in topical and systemic bacterial infections. *The Journal of Alternative and Complementary Medicine*, 24(1), 15-24 doi: 10.1089/acm.2017.0017.

Kalinichenko S. V., Babych Ye. M. , Ryzhkova T. A. , Maslii I. H. , Korotkykh O. O. , Danilina S. S., Solianik O. H. , Shykova O. A. , Skliar N. I. , Tkach L. M. , Balak A. K. , Niemkova S. M. , Desiatnykova O. V., (2013). Suchasnyi stan rozrobky ta zastosuvannia pro biotychnykh, pre biotychnykh ta synbiotychnykh preparativ (ohliad literatury) [Current status of development and use of biotic, prebiotic and synbiotic preparations (literature review)]. Annals of Mechnikov Institute, N 3, www.imiamn.org.ua /journal.htm 5. P. 5–11. (in Ukrainian).

Kotsiumbas I. Ya., Zhyla M. I., Shkil M. I. (2013). Probiotyky – neobkhidna skladova pry suchasnykh tekhnolohiiakh vyroshchuvannia tvaryn [Probiotics are an indispensable component of modern animal husbandry technologies]. *Nauk. visnyk LNUVMBT. im. S. Z. Gzhytskoho.* Vyp. 3 (57). S. 174–181.

Kutsan O. T., Maslii I. H., Niemkova S. M., Stupak L. P., Desiatnykova O. V. (2013). Metodychni rekomendatsii. Vyznachennia kontaminatsii medu mikrofloroiu, patohennoiu dlia bdzhil [Methodological recommendations. Determination of contamination of honey with microflora pathogenic to bees]. (NNTs “IEKVM”). Utv. DVFSS Ukr. № 1 vid 21.12.2012 Vyd. ofits. Kyiv: 40 p. (in Ukrainian).

Labynskaya, A. S. (1978). Mykrobyolohyya s tekhnykoy mykrobyolohycheskykh yssledovanyy [Teskt]. M.: Medytsyna, 394 p.

Larsen, A., Reynaldi, F. J., & Guzmán-Novoa, E. (2019). Fundaments of the honey bee (Apis mellifera) immune system. Review. Revista mexicana de ciencias pecuarias, 10(3), 705-728. https://doi.org/10.22319/rmcp.v10i3.4785

Ogrodowczyk, A. M., Zakrzewska, M., Romaszko, E., & Wróblewska, B. (2020). Gestational dysfunction-driven diets and probiotic supplementation correlate with the profile of allergen-specific antibodies in the serum of allergy sufferers. Nutrients, 12(8), 2381. https://doi.org/10.3390/nu12082381.

Paytuví-Gallart, A., Sanseverino, W., & Winger, A. M. (2020). Daily intake of probiotic strain Bacillus subtilis DE111 supports a healthy microbiome in children attending day-care. Beneficial Microbes, 11(7), 611-620. doi: 10.3920/BM2020.0022.

Piccini C., Antunez K., Zunino P. (2004). An approach to the characterization of the honey bee hive bacterial flora. *J.apic.Res.* Vol.33, N 3, P. 101–104

Ptaszyńska, A. A., Borsuk, G., Zdybicka-Barabas, A., Cytryńska, M., & Małek, W. (2016). Are commercial probiotics and prebiotics effective in the treatment and prevention of honeybee nosemosis C. Parasitology research, 115(1), 397-406. doi: 10.1007/s00436-015-4761-z

Richardson R. T. et al. (2018) This article is an open access publication. Morphological and functional characterization of honey bee, Apis mellifera, hemocyte cell communities Apidologie https://doi.org/10.1007/s13592-018-0566-2

Șapcaliu, A., Rădoi, I., Pavel, C., Tudor, N., Căuia, E., Siceanu, A., & Meiu, F. (2009). Research regarding haemocyte profile from Apis mellifera carpatica bee haemolymph originated in the south of Romania. Lucrari Stiintifice-Universitatea de Stiinte Agricole a Banatului Timisoara, Medicina Veterinara, 42(2), 393-397.

Saranchuk, I. I., Vishchur, V. Y., Gutyj, B. V., & Klim, O. Y. (2021). Effect of various amounts of sunflower oil in feed additives on breast tissues functional condition, reproductivity, and productivity of honey bees. *Ukrainian Journal of Ecology*, 11(1), 344-349. doi: 10.15421/2021_51

Szymaś B.et al. (2003) The influence of different diets on haemocytes of adult worker honey bees, Apis mellifera Apidologie 34 (2003) 97–102 © INRA/DIB-AGIB/ EDP Sciences, DOI: 10.1051/apido:2003012

Tushak, S. (2018). Quantitative changes in hemogram of bees using probiotic «Enteronormin». *Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series: Veterinary Sciences*, 20(83), 61-65. doi: 10.15421/nvlvet8312.

Vlizlo, V.V. et al. (2012). Laboratorni metody doslidzhennya u biologiyi, tvarynnytstvi ta veterynarniy medycyni [Laboratory methods of research in biology, stockbreeding and veterinary medicine: a guide]. Lviv: Spolom 764 c. (in Ukrainian)

William J. (2014) Honey Bee Hemocyte Profiling by Flow Cytometry Marringa, Michael J. Krueger, Nancy L. Burritt, James B. Burritt PLoS One 9: e37235. PLOS ONE www.plosone.org Volume 9 Issue 10 e108486

Yazlovitskaya, L. S., Cherevatov, V. F., Savchuk, G. G., & Khlus, V. K. (2014). Tipologicheskiye osobennosti kletok gemolimfy pchel Apis Mellifera L., rayonirovannykh v Chernovitskoy oblasti [Typological features of hemolymph cells of Apis Mellifera L. bees, zoned in the Chernivtsi region]. *Ekologicheskiy monitoring i bioraznoobraziye*, (1), 134-138

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UDDER TREATMENT AND EVALUATION ARE KEY ELEMENTS IN COW MILKING TECHNOLOGY

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One of the most pressing challenges in the field of agricultural science is the production of high-quality milk on industrial farms. In light of these conditions, there is an urgent need to ensure animal health and prevent mastitis. The generation of new ideas for improving current milk production technologies and technical solutions that ensure their high competitiveness should be a key priority in this process. Drawing on a wide range of information materials from both domestic and international databases in the field of intellectual property, the article contends that manufacturers currently offer a variety of devices for processing and evaluating cow udders. However, it is noted that not all of these devices are universally applicable and that there are some disadvantages associated with their use. The article presents the results of the development of devices for processing and evaluating the udder of cows. These were based on the results of a patent search and theoretical analysis of existing analogs and prototypes. As part of this work, the initial parameters, design features, and main advantages of the developed five devices for processing cow udders both before and after milking are substantiated. These devices are easy to use, provide optimal consumption of mammary gland treatment products, help reduce manual labor costs for machine milking operators, improve milk quality, and reduce the level of animal mastitis. A device for assessing certain properties of teats has been developed. The new technological devices differ from existing analogs not only in their simplified design, but also in being more technologically advanced and cost-effective. The results obtained from implementing the proposed devices in practice will allow us to offer new and effective programs for managing the processes of high-quality milk production in the functional system "man-machine". They will also contribute to the introduction of auxiliary biological tests and rational technological approaches to its evaluation shortly.

Keywords: cattle, udder, processing, device, scheme.

ОБРОБЛЕННЯ ТА ОЦІНЮВАННЯ ВИМЕНІ – КЛЮЧОВІ ЛАНКИ В ТЕХНОЛОГІЇ ДОЇННЯ КОРІВ

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Отримання високоякісного молока на фермах промислового типу є одним із актуальних завдань аграрної науки. За цих умов виникає нагальна необхідність в забезпеченні збереження здоров'я тварин та профілактики виникнення маститів. Чільне місце в цьому процесі має посісти питання генерування якісно нових ідей щодо удосконалення поточних технологій виробництва молока та технічних рішень, які забезпечують його високу конкурентоспроможність. За узагальнення інформаційних матеріалів вітчизняних та зарубіжних баз даних у сфері інтелектуальної власності аргументовано, що на сьогодні виробниками запропоновано цілу низку пристроїв для оброблення та оцінки вимені корів, проте не всі вони є універсальними та мають ряд недоліків за використання. У статті викладено результати щодо розробок пристроїв для оброблення та оцінки вимені корів, які виконували за наслідками патентного пошуку та теоретичного аналізу існуючих аналогів і прототипів. У рамках проведеної роботи обґрунтовано вихідні параметри, конструктивні особливості та основні переваги розроблених п'яти пристроїв для оброблення вимені корів як перед, так і після доїння, які прості у застосуванні, забезпечують оптимальну витрату засобів для оброблення молочної залози, сприяють зниженню витрат ручної праці операторів машинного доїння, дають змогу покращити якість молока та знизити рівень захворювання тварин на мастити. Розроблено пристрій для оцінювання окремих властивостей дійок. Нові технологічні пристрої вирізняються від існуючих аналогів не лише спрощеною конструкцією, але й є більш технологічними та економічно вигідними. Одержані результати щодо реалізації запропонованих пристроїв на практиці дадуть змогу запропонувати нові дієві програми управління процесами виробництва високоякісного молока у функціональній системі ,,людина–машина", а також сприяли запровадженню допоміжних біологічних тестів і раціональних технологічних підходів щодо його оцінювання на найближчу перспективу.

Ключові слова: велика рогата худоба, вим'я, обробка, пристрій, схема.

Today, the problem of providing the population with high-quality and safe products of animal origin is reflected in the works of scientists (Nespolo N. M., 2021; Michalchenko S. A. et al., 2024). The complexity of solving this multifaceted problem implies compliance with high requirements, first of all, for the quality and safety of milk, as it is a unique and most popular human food product (Antunes I. C. et al., 2022). The

industrial realization of this problem is possible only if all sanitary and hygienic requirements are strictly observed during its production and processing (Hadzevych O. V. et al., 2019; Berge A. C. & Baars T., 2020). First of all, this concerns the increased bacterial contamination of milk as a result of non-compliance with the requirements of cow milking hygiene and the use of imperfect technologies and technical solutions.

One of these requirements is to eliminate the possibility of bacterial contamination of the product at the milking stage. In other words, it is necessary to eliminate the possibility of microbial contamination of milk at the primary stage of its production - during the milking process of cows (Singh A. & Ramachandran A., 2020; Vargova M. et al., 2023). At the same time, the number of cases of bovine mastitis can easily be reduced by simple measures, such as treating the udder with antiseptics before and after milking. Pre-milking treatment helps to cleanse the teat skin of dirt and micro-organisms, preventing from entry of microorganisms into the freshly milked milk and transmitting mastitis pathogens from cow to cow via the milking equipment. The most important procedure in the technology for producing high-quality milk is the post-milking treatment of the cow's udder with antiseptics. This treatment reduces the level of pathogenic microflora on the teat skin. It prevents their penetration into the mammary gland by moisturizing the skin and maintaining it in a normal physiological state (Kukhtyn M. et al., 2021).

However, specialists are faced with the problem of choosing an antiseptic, which should be based on the proven effectiveness of its use. In particular, iodine, lactic acid, glycolic acid, and plant extracts are most commonly used to treat teats after milking (Zazharska N. V. & Biben I. A., 2023). The use of chemicals for teat treatment before and after milking can reduce bacterial contamination of teats and enable the production of high-quality milk (Fotina T. I. et al., 2015; Zazharska N. M. & Riaba A. O., 2016), but raises concerns about the risk of chemical residues in milk. Against the background of the use of chemicals for udder treatment, there are many different semiautomatic and automatic devices (Paladiychuk O. R., 2019), but their main disadvantages are complex design and inconvenience in use. Based on these conditions, the development of new technical means for the treatment of cows' udder teats emphasizes the undeniable scientific and practical basis of the problem, determines its relevance, and is the basis for in-depth scientific research.

The research aims to develop innovative devices for processing the teats of cow udders both before and after milking.

Materials and methods. The information base of the study was formed by domestic and foreign patents for inventions and utility models, scientific papers, and Internet resources. The methodological basis of the study was the methods of comparative analysis of existing analogues and prototypes on the studied issues, systematization, and generalization. The work was carried out at the production base of the National Scientific Center "Institute of Experimental and Clinical Veterinary Medicine" from 2018 to 2023.

Research results. As part of the research, several innovative devices have been developed for processing cow udders both before and after milking. The developments differ from existing analogs in their simplified design, are more technologically advanced, and cost-effective.

The innovation of the development lies in the fact that the device contains a container for the disinfectant solution, which is made of elastic transparent material and has a dipping glass with a handle, which is connected to the container by an adapter, while the glass in the upper part has rounded edges and a gutter that prevents overuse of the disinfectant (Fig. 1).

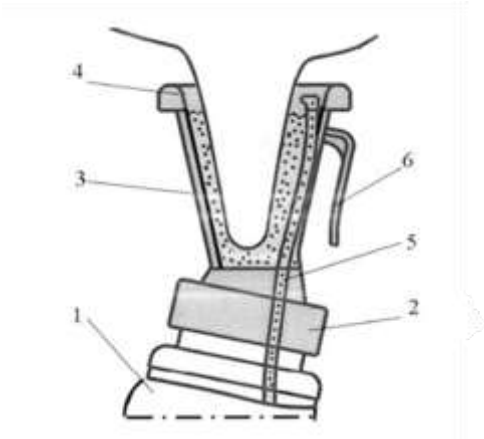

Fig. 1. **Scheme of the device for processing cow udder teats**

The device for treating cow udder teats consists of a container for disinfectant solution 1 made of elastic transparent material, an adapter 2 connecting the container 1 to a dipping glass 3, which has rounded edges 4 at the top to prevent injury to the udder teats during treatment and facilitate their quick entry into the glass, a gutter 5 that prevents overuse of the disinfectant, and a handle 6.

The device for treating cow's udder teats works as follows: the disinfectant solution container 1, made of elastic transparent material, is filled with disinfectant solution. A dip cup 3 is connected to it through an adapter 2. After milking, the cow's udder teats are treated by placing the teat in the dipping glass 3 and then squeezing the container 1 made of elastic transparent material. The rounded edges 4 facilitate quick and painless insertion and positioning of the teat in the inner part of the dipping glass 3. During manipulations, the excess disinfectant solution flows back into the container 1 through the gutter 5. In this way, the udder teats are processed quickly and efficiently, while preventing disinfectant loss and overspray. After use, the device can be placed on any surface of the machine equipment using the handle 6. The advantages of the proposed device are that it reduces the material costs of udder treatment and increases the productivity of the milking operators. The device is reliable and easy to use. The innovation is protected by a utility model patent*.

Another development has a container for a disinfectant solution made of elastic transparent material and a gradation, a body connected to the container at an angle of 5° and made in the shape of a hollow cone. The cone has a fleecy inner surface and a handle, which reduces material costs during processing, improves milk quality, and prevents microorganisms from entering the udder teats.

The design features of the developed device for pre-milking treatment of cow udder teats are shown in Fig. 2.

The device for pre-milking treatment of cow udder teats consists of a container for disinfectant solution 1 made of elastic transparent material and having a gradation 2, an adapter 3 that connects the container 1 to the body 4 at an angle of 5°, which is made in the form of a hollow cone, has a fleecy inner surface 5 and a handle 6.

* Utility model patent No. 133582 Ukraine, IPC A01J 7/00, A01J 7/04. Device for processing of udder teats of cows. Palii A.P., Paliy A.P., Ishchenko K.V. - No. u 2018 11593; applied for on 26.11.2018; published on 10.04.2019; bulletin No. 7.

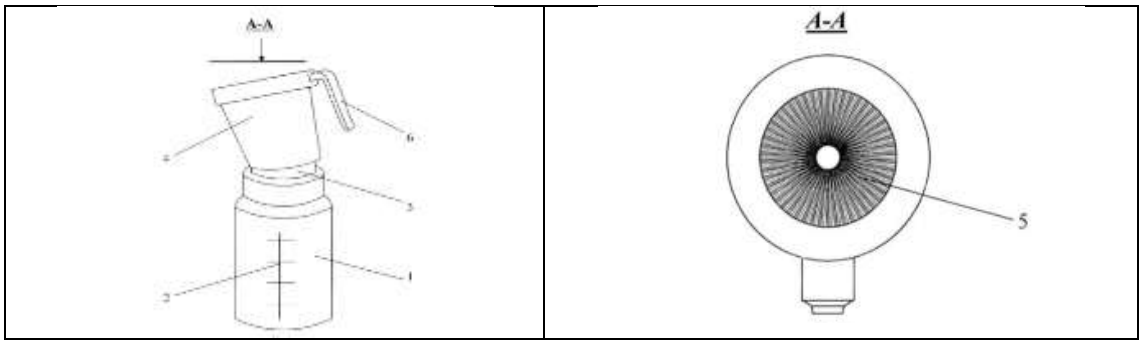

Fig. 2. **Scheme of the device for pre-milking treatment of cow udder teats**

The device for pre-milking treatment of a cow's udder teats works as follows: a container for disinfectant solution made of elastic transparent material 1 is filled with disinfectant solution according to the gradation 2. The body 4, which is presented in the form of a hollow cone, is connected to the filled container 1 through an adapter 3 at an angle of 5°. Before milking, each udder teat is treated separately by placing it in the inner hollow cone of the body 4, then squeezing the container made of elastic transparent material 1 and moving it around the udder teat. Due to the interaction of the udder teat with the fleecy inner surface 5 of the body 4, the udder teat is processed quickly and efficiently. After use, the device can be placed on any surface of the machine tool using the handle 6. The development is protected by a patent of Ukraine for a utility model**.

The next technological task was solved in the direction of creating a device for processing cow udders in milking parlors. The developed device has a vacuum pump (1), filter (2), fan (4), pressure (3) and distribution pipes (5) with nozzles (6) placed on the top along the technological trench and directing the flow of warm air to the animal's udder, which ensures high efficiency of the device. The device helps reduce manual labor costs for the milking operator, as it connects the milking machine to dry teats, which in turn prevents milk contamination. With dry teats, the milking cups at least go up the teats and compress the teat canal, which significantly reduces udder disease (Fig. 3).

The device works as follows: the vacuum pump 1, which draws air from the vacuum system of the milking machine, delivers it to the pressure pipe 3. Passing through filter 2, the warm air is cleaned, picked up by fan 4, creating a slight vacuum in the pressure pipe 3, and fed to the distribution pipe 5 of the warm air flow with nozzles 6, which are located at the top along the technological trench.

The nozzles are designed in such a way that the warm air coming out of them creates a directed movement of streams on the animal's udder from both sides, drying it. The advantages of the proposed device are that it is easy to use, reduces the cost of manual labor of the milking operator during milking, improves the quality of milk and reduces the incidence of udder diseases in cows. The development is protected by utility model patent of Ukraine***.

**Utility model patent No. 134677 Ukraine, IPC A01J 7/00, A01J 7/04. Device for pre-milking treatment of cattle udder teats. Palii A.P., Paliy A.P., Petrov A.M. No. u 2019 00307; applied for on 26.02.2019; published on 27.05.2019; bulletin No. 10

***Utility model patent No. 151014 Ukraine, IPC A01J 7/00. Device for processing cow udders in milking parlors. Rodionova K.O., Palii A.P., Khimych M.S., Paliy A.P., Dubin R.A. No. u 2021 06282; applied for on 08.11.2021; published on 25.05.2022; bulletin No. 21.

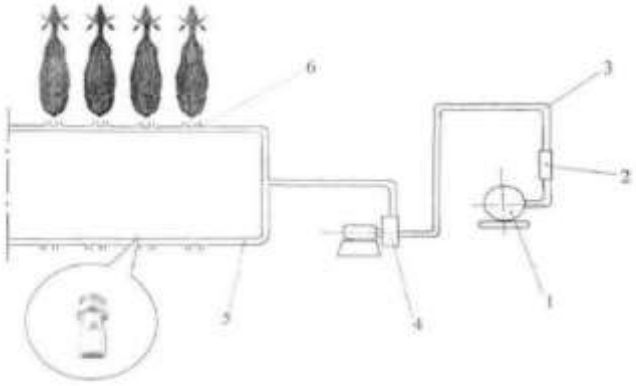

Fig. 3. **Device for treating cow udders in milking parlors**

The following technological solution for the pre-milking treatment of the cow's udder was set as a task, which was solved by designing a device consisting of a body (1), two brushes (2) rotating towards each other, a drive (3), a handle (4) and a wire (5). The proposed device reduces material costs for udder treatment, improves milk quality, prevents microorganisms from entering the udder teats, and reduces the workload of the milking operator (Fig. 4).

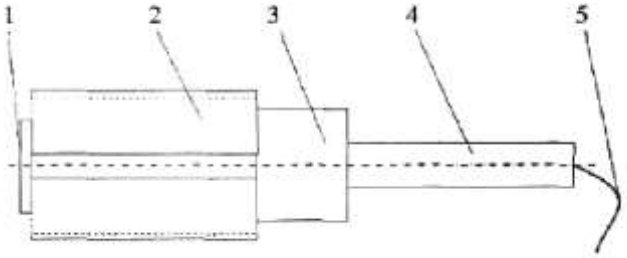

Fig. 4. **Device for pre-milking treatment of the cow udder**

The device operates as follows: after the cow has taken a suitable place in the milking parlor, the body (1), on the axes of which two brushes (2) are fixed, is brought to the base of the animal's udder with the help of the handle (4). The body (1) is placed in such a way that the two teats of the udder are between the brushes (2). At the next stage, the drive (3) of the brushes (2) is turned on and they begin to rotate toward each other, performing the function of removing the first lumps of milk, cleaning, disinfecting, and massaging. The significant advantages of the proposed device are that it is easy to use, reduces material costs for pre-milking udder treatment, improves milk quality and prevents microorganisms from entering the udder teats, reduces the workload of the milking operator, and speeds up preparatory operations. The development is protected by the utility model patent of Ukraine****.

The next technological solution was based on the task of developing a stationary device for processing cows' udders in milking parlors. The task was solved by the fact that this device consists of a platform 1, a base plate 2, a switching device 3, a pump 4, a disinfectant container 5, a piping system 6, and nozzles 7.

****Utility model patent No. 151015 Ukraine, IPC A01J 7/00. Device for pre-milking treatment of cattle udder. Palii A.P., Rodionova K.O., Rodionova K.O., Khymych M.S., Paliy A.P., Anfiorova M.V. - No. u 2021 06294; applied for on 08.11.2021; published 25.05.2022; bulletin No. 21.

A comparative analysis of the developed device and the closest analog allows us to conclude that it differs from the closest analog in that it uses a platform, a base plate, an automatic switching device, a pump, a disinfectant tank, a piping system, and nozzles to treat cows' udders in milking parlors. This helps to reduce the manual labor costs of the milking operator in milking cows by saving time in preparation operations, ensuring optimal consumption of udder treatment products, and improving the quality of the milk produced (Fig. 5).

The device is mounted in the milking parlor under the place where the cow is placed during milking, and it works as follows: the cow enters the milking parlor and is placed on the platform 1. After that, the platform 1 acts on the base plate 2, which starts the pump 4 through the automatic switching device 3.

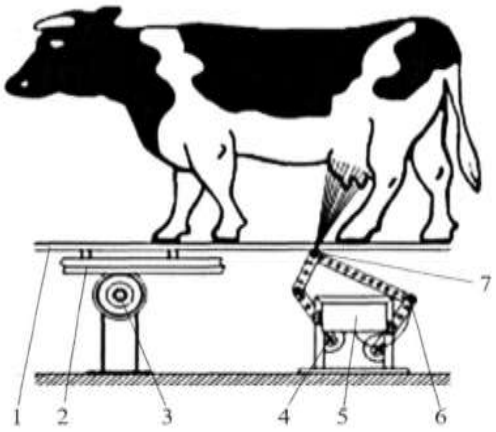

Fig. 5. **Stationary device for processing cow udders in milking parlors**

After the pump 4 is started, the disinfectant is supplied from the disinfectant container 5. The disinfectant is delivered through the pipeline system 6 to the nozzles 7, through which the udder is treated. The advantages of the device are that it is easy to use, provides optimal consumption of products for treating cow udders, reduces manual labor costs for milking operators, improves milk quality, and reduces udder diseases. The development is protected by the utility model patent of Ukraine*****.

The lack of proper equipment for assessing the condition of cows' udder teats causes serious problems for their health and productivity, which requires more thorough research. This technological problem has been solved by developing a device for determining the elastic properties of cows' udder teats. The development is protected by a patent of Ukraine for a utility model******. The main physical and mechanical properties of cow's udder teats, which are necessary for the development of physiologically adapted equipment, include the characteristics of elastic properties: the coefficient of transverse deformation of the teat and, accordingly, the variable modulus of its elasticity (Fig. 6).

*****Patent for utility model No. 153790 Ukraine, IPC A01J 7/00. Stationary device for processing cow udders in milking parlors. Palii A.P., Naumenko A.O., Naumenko O.A., Paliy A.P., Petrov A.M. - No. u 2022 04586; applied for on 05.12.2022; published 30.08.2023; bulletin No. 35.

******Patent for utility model No. 146403 Ukraine, IPC A01J 7/00. Device for determining the elastic properties of cow udder teats. Palii A.P., Naumenko A.O., Paliy A.P. - No. u 2020 06386; applied for on 02.10.2020; published 02.17.2021; bulletin No. 7.

Fig. 6. **Device for determining the elastic properties of cow udder teats**

The device consists of a metal rod-body 1, at the upper end of which is mounted to the udder teat 2, and in the lower part - a disc lock-limiter 3 and metal discs with slots 4. The device works as follows: at the beginning of use, horizontal parallel lines are applied to the middle part of the teat with a marker. Then, a metal rod-body 1 is attached to the teat using a mount 2. Subsequently, a load is gradually and consistently applied to the teat by installing the required number of metal discs with slots 4 on the metal rod-body 1. The metal discs with slots 4 are held on the rod-body 1 through a disc lock-limiter 3. At the end, the determination is fixed, and this is done with each subsequent load. To determine the scale, a ruler is used, which is placed directly near the teat in the required plane. To determine the elastic properties, the load on the udder teat varies from 0.135 kg to 0.675 kg.

During the measurement and determination of the indicators, the distance between the horizontal parallel lines on the teats and, accordingly, the diameter of the mammary gland after each subsequent load are measured. The device provides rapid receipt of reliable data, allows arguing the mechanism of interaction of the teat liner and the teat, and on this basis, to justify the amount of teat liner tension.

Discussion. The increase in milk production is based on the intensive development of the industry, the introduction of innovative technologies and technological solutions, and the improvement of the organization and management of production processes. The corresponding growth rates of milk production make us pay special attention to improving its quality. Ensuring high quality and safety of dairy raw materials for dairy products is a fundamental component of public health protection and, of course, one of the priority challenges of the Ukrainian agro-industrial complex.

As the demand for higher milk yields and improved milking efficiency increases, there is a growing focus on the productive capacity of dairy cows to meet the needs of a growing global population (Neculai-Valeanu A. S. & Ariton A. M., 2022). In this context, the findings from various studies align with previous research (O'Rourke D., 2009; Williamson J. H. & Lacy-Hulbert S. J., 2013; Barkema H. W. et al., 2015), highlighting that proper udder care is crucial not only for preventing mastitis but also for reducing its incidence, particularly after milking. Mastitis can be transmitted during the milking process or through the milking machine. Additionally, other studies have demonstrated a direct link between udder care procedures and the technical and technological parameters of milking cows (Aliiev E. et al., 2022).

To treat udder teats, it is suggested to use special cups with an antiseptic preparation, followed by wiping with reusable napkins (Miseikiene R. et al., 2015; Paladiychuk O. R., 2019), which is emphasized in our work. However, other scientists have reported that pre-milking udder treatment does not ensure sufficient teat cleanliness. Pathogenic organisms remain afterwards and can accumulate in the teat dipping caps

(Borodina O. V. & Nosevych D. K., 2017). Instead, (Paliy A. et al., 2021) demonstrated the high efficiency of udder teat treatment with special brushes, the use of which significantly improved milking hygiene.

The quality of udder treatment is directly related to the antiseptic used on a particular farm. However, it is important to consider the methods used. The use of liquid products, which is the vast majority, requires the use of appropriate containers and equipment. Based on the results of the research, many innovative equipment has been offered to the industry, which is import substitution.

High efficiency of using high-yielding cows, increasing their productivity and milk quality can be ensured only if the animals comply with the rules of machine milking, which requires the introduction of innovative technological solutions based on the latest animal care technologies and rational organization of production processes (Boltianska N. & Zabolotko O., 2020). However, all this is possible with a scientifically based assessment of the suitability of cows for automated milking, taking into account their physiological characteristics (Hovinen M. & Pyörälä S., 2011). Our research complements the existing system for evaluating dairy cows for milking by developing a device for evaluating individual teat characteristics.

Conclusions:

1. The quality and safety of cow milk directly depends on the hygiene of milking. Udder treatment is a key element in milking technology and a preventive measure to reduce the incidence of milk contamination with microorganisms.

2. Five devices have been developed for processing cow udders both before and after milking, as well as a device for determining the elastic properties of teats, which should be used at milk production complexes.

3. The advantage of the developed devices is their ease of use; they provide optimal consumption of means for processing and evaluating the udder of cows, help reduce the cost of manual labor of milking operators, improve milk quality, and reduce cow mastitis. They are more technologically advanced and cost-effective.

4. The results obtained on the implementation of the proposed devices in practice will allow us to offer new effective programs for managing the processes of high-quality milk production in the functional man-machine system, and also contribute to the introduction of auxiliary biological tests and rational technological approaches to its evaluation shortly.

Acknowledgements. The article is dedicated to the blessed memory of the famous scientist, talented organizer of the educational and scientific process, Doctor of Agricultural Sciences, Professor Andrii Palii.

References

Aliiev, E., Paliy, A., Kis, V., Paliy, A., Petrov, R., Plyuta, L., Chekan, O., Musiienko, O., Ukhovskyi, V., & Korniienko, L. (2022). Establishment of the influence of technical and technological parameters of dairy and milking equipment on the efficiency of machining. *Eastern-European Journal of Enterprise Technologies*. 1 (1 (115)), 44–55. https://doi.org/ 10.15587/1729-4061.2022.251172

Antunes, I. C., Bexiga, R., Pinto, C., Roseiro, L. C., & Quaresma, M. A. G. (2022). Cow's milk in human nutrition and the emergence of plant-based milk alternatives. *Foods.* 12(1), 99–120. https://doi.org/10.3390/foods12010099

Barkema, H. W., von Keyserlingk, M. A., Kastelic, J. P., Lam, T. J., Luby, C., Roy, J. P., LeBlanc, S. J., Keefe, G. P., & Kelton, D. F. (2015). Invited review: Changes in the dairy industry affecting dairy cattle health and welfare. *Journal of Dairy Science.* 98(11), 7426–7445. https://doi.org/10.3168/jds.2015-9377

Berge, A. C., & Baars, T. (2020). Raw milk producers with high levels of hygiene and safety. *Epidemiology and Infection.* 148, 14. https://doi.org/10.1017/S0950268820000060

Boltianska, N. I. & Boltianskyi, O. V. (2020). Vyznachennia napriamiv enerhozberezhennia v silskomu hospodarstvi [Defining directions of energy saving in agriculture]. *Naukovyi visnyk TDATU.* 10(1), 3–9. (in Ukrainian). https://doi.org/10.31388/2220-8674-2020-1-5

Borodina, O. V., & Nosevych, D. K. (2017). Bakterial′na zabrudnenist′ diyok pid chas doyinnya koriv na doyil′nomu maydanchyku [Bacterial contamination of teats during caw milking in the milking area]. *Scientific Bulletin of the National University of Life Resources and Environmental Management of Ukraine. Series: Technology of Production and Processing of Livestock Products*. 271, 210–216. (in Ukrainian). http://nbuv.gov.ua/UJRN/nvnau_tevppt_2017_271_27

Fotina, T. I., Zazharska, N. M., & Kostiuchenko, V. Yu. (2015). Vplyv zasobiv dlia doinnia na sanitarnu yakist kozynoho moloka [Influence of facilities for milking on sanitary quality of goat's milk]. *Visnik Sumskogo Nacionalnogo Agrarnogo Universitetu*. 7 (37), 59–65. (in Ukrainian).

Hadzevych, O. V., Paliy, A. P., Kinash, O. V, Petrov, R. V., & Paliy, A. P. (2019). Antibiotic resistance of microorganisms isolated from milk. *World of Medicine and Biology.* 3(69), 245–250. https://doi.org/10.26724/2079-8334-2019-3-69-245-250

Hovinen, M., & Pyörälä, S. (2011). Invited review: udder health of dairy cows in automatic milking. *Journal of Dairy Science*. 94(2), 547–562. https://doi.org/10.3168/jds.2010-3556

Kukhtyn, M., Boltyk, N., Perkii, Yu., & Protsenko, T. (2021). Rol sanatsii shkiry diiok vymeni koriv u profilaktytsi mastytu [The role of skin sanitation of cows' udders in the prevention of mastitis]. Conference "Modern Methods of Diagnostic, Treatment and Prevention in Veterinary Medicine&quot" 95. Retrieved from. (in Ukrainian). https://nvlvet.com.ua/index.php/conference/article/view/4513

Michalchenko, S. A., Korkh, I. V., Paliy, A. P., Boiko, N. V., Kovalenko, L. V., Pavlichenko, O. V., Vyrvykyshka, S. M., & Morozov, M. G. (2024). Amino acid composition of beef depending on the breed and age of dairy bulls. *International Journal of Agricultural Technology.* 20(6), 2405–2422.

Mišeikienė, R., Rudejevienė, J., & Gerulis, G. (2015). Effect of pre-milking antiseptic treatment on the bacterial contamination of cow teats' skin. *Bulgarian Journal of Veterinary Medicine.* 18(2), 159–166. https://doi.org/0.15547/bjvm.833

Neculai-Valeanu, A. S., & Ariton, A. M. (2022). Udder health monitoring for prevention of bovine mastitis and improvement of milk quality. *Bioengineering (Basel).* 9(11), 608–632. https://doi.org/10.3390/bioengineering9110608

Nespolo, N. M. (2021). The behavior of consumers and producers of food of animal origin and their impacts in One Health. *Frontiers in Veterinary Science.* 8, 641634. https://doi.org/10.3389/fvets.2021.641634.

O'Rourke, D. (2009). Nutrition and udder health in dairy cows: a review. *Irish Veterinary Journal.* 62(4), 15–20. https://doi.org/10.1186/2046-0481-62-S4-S15

Paladiichuk, O. P. (2019). Predypinh ta postdypinh – efektyvni metody profilaktyky mastytiv u molochnykh koriv [Pre-dipping and post-dipping – effective methods for the preventition of dairy cows mastitis]. Ahrarna nauka ta kharchovi tekhnolohii. 4(107)1, 100–112. (in Ukrainian).

Paliy, A., Aliiev, E., Paliy, A., Ishchenko, K., Shkromada, O., Musiienko, Y., Plyuta, L., Chekan, O., Dubin, R., & Mohutova, V. (2021). Development of a device for cleansing cow udder teats and testing it under industrial conditions. *Eastern-*

European Journal of Enterprise Technologies. 1/1 (109), 43–53. https://doi.org/10.15587/1729-4061.2021.224927

Singh, A., & Ramachandran, A. (2020). Assessment of hygienic milking practices and prevalence of bovine mastitis in small dairy farms of peri-urban area of Jaipur. *Indian journal of community medicine.* 45(1). 21–25. https://doi.org/10.4103/ijcm.IJCM_363_19

Vargova, M., Vyrostkova, J., Lakticova, K. V., & Zigo, F. (2023). Effectiveness of sanitation regime in a milking parlour to control microbial contamination of teats and surfaces teat cups'. *Annals of Agricultural and Environmental Medicine.* 30(1). 55–60. https://doi.org/10.26444/aaem/161037

Williamson, J. H., & Lacy-Hulbert, S. J. (2013). Effect of disinfecting teats post-milking or pre- and post-milking on intramammary infection and somatic cell count. *New Zealand Veterinary Journal.* 61(5), 262–268. https://doi.org/10.1080/00480169.2012.751576

Zazharska, N. V., & Biben, I. A. (2023). Means for pre-milking and post-milking processing of cow udders. Bulletin of Sumy National Agrarian University. *The Series: Veterinary Medicine.* 4(63), 43–50. https://doi.org/10.32782/bsnau.vet.2023.4.7

Zazharska, N. M., & Riaba, A. O. (2016). Sanitarna yakist kozynoho moloka za vykorystannia homeopatychnykh zasobiv dlia doinnia [Sanitary quality of goat milk in the application of the homeopathic preparations for milking]. Naukovo-tekhnichnyi biuleten Derzhavnoho naukovo-doslidnoho kontrolnoho instytutu veterynarnykh preparativ ta kormovykh dobavok i Instytutu biolohii tvaryn, 17 (1), 72–77. (in Ukrainian).

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THE META-ANALYSIS OF CERVICAL AND POST-CERVICAL ARTIFICIAL INSEMINATION OF SOWS

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The efficiency of artificial insemination (AI) in pig breeding plays a critical role in improving reproductive performance, reducing breeding costs, and increasing the sustainability of the pig farming industry. Unlike cervical insemination (CAI), PCAI allows for more precise sperm deposition in the uterus, improving the chances of conception and this ensures more effective intensive boar management.

To evaluate the effectiveness of PCAI in improving pig breeding efficiency, a meta-analysis was conducted using Jamovi software and algorithms from the metaphor package (R). Publication bias estimation was integrated into the analysis to ensure the reliability of the findings. The meta-analysis compared PCAI and CAI across three critical reproductive parameters: farrowing rate, fecundity index, and litter size. Criteria for study inclusion were defined to ensure the integrity of the analysis: the exclusion of studies involving exogenous hormonal treatments that could interfere with natural reproductive processes, the inclusion of studies with at least 20 animals per group, availability of group sizes and insemination doses, and provision of relevant variation statistics.

A total of 34 studies were included in the analysis for the farrowing rate, 33 studies for the fecundity index, and 33 studies for the litter size. The inclusion of such a large number of studies enhanced the robustness of the meta-analysis and allowed for a comprehensive evaluation of PCAI's impact on pig breeding efficiency.

The meta-analysis results showed that PCAI does not significantly worsen the farrowing rate. Specifically, the average log odds ratio based on the random-effects model was 0.0061 (95% CI: -0.2042 to 0.2163), indicating no detrimental effect on the farrowing rate when PCAI was used. Similarly, no significant differences were found between PCAI and CAI for the fecundity index (average standardized mean difference was 0.1156; 95% CI: -0.0790 to 0.3103), nor for litter size (average standardized mean difference was 0.0226; 95% CI: -0.0670 to 0.1123). These findings suggest that PCAI is comparable to traditional AI methods in terms of key reproductive parameters, which is a crucial consideration for breeders seeking to improve their production efficiency.

The publication showed that use of PCAI can offer several economic benefits. By increasing the efficiency of boars and making desirable genetics more accessible, PCAI reduces the need for maintaining a large number of boars on farms. This not only reduces the costs associated with keeping boars, but also promotes the wider use of high-quality genetic material, leading to the genetic improvement of industrial pigs.

Keywords: Sows artificial insemination, traditional cervical insemination (CAI), post-cervical artificial insemination *(PCAI)*, meta-analysis, the farrowing rate, fecundity index, the litter size.

МЕТА-АНАЛІЗ МЕТОДІВ ЦЕРВІКАЛЬНОГО ТА ПОСТЦЕРВІКАЛЬНОГО ОСІМЕНІННЯ СВИНОМАТОК

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Ефективність штучного осіменіння (ШІ) у свинарстві відіграє вирішальну роль у покращенні репродуктивної продуктивності, зниженні витрат на племінну діяльність та підвищенні стійкості свинарської галузі. На відміну від цервікального осіменіння (CAI), PCAI дозволяє точніше осідати сперму в матці, підвищуючи шанси на зачаття, а це забезпечує більш ефективне інтенсивне утримання кнурів.

Щоб оцінити ефективність PCAI у покращенні ефективності розведення свиней, було проведено мета-аналіз з використанням програмного забезпечення Jamovi та алгоритмів із пакету метафор (R). Оцінка похибки публікації була інтегрована в аналіз, щоб забезпечити надійність результатів. Метааналіз порівнював PCAI та CAI за трьома критичними репродуктивними параметрами: частотою опоросів, індексом плодючості та розміром посліду. Критерії включення дослідження були визначені для забезпечення цілісності аналізу: виключення досліджень, що включають екзогенне гормональне лікування, яке може заважати природним репродуктивним процесам, включення досліджень із принаймні 20 тваринами на групу, наявність розмірів груп і доз осіменіння, і надання відповідної статистики варіацій.

Загалом 34 дослідження були включені в аналіз частоти опоросів, 33 дослідження індексу плодючості та 33 дослідження розміру приплоду. Включення такої великої кількості досліджень підвищило надійність мета-аналізу та дозволило провести комплексну оцінку впливу PCAI на ефективність розведення свиней.

Результати мета-аналізу показали, що PCAI істотно не погіршує частоту опоросів. Зокрема, середнє логарифмічне співвідношення шансів на основі моделі випадкових ефектів становило 0,0061 (95% ДІ: від -0,2042 до 0,2163), що вказує на відсутність шкідливого впливу на частоту опоросів при застосуванні PCAI. Подібним чином не було виявлено істотних відмінностей між PCAI та CAI для індексу плодючості (середня стандартизована середня різниця становила 0,1156; 95% ДІ: від -0,0790 до 0,3103), а також для розміру посліду (середня стандартизована середня різниця становила 0,0226; 95% ДІ: від -0,0670 до 0,1123). Ці результати свідчать про те, що PCAI можна порівняти з традиційними методами штучного інтелекту з точки зору ключових репродуктивних параметрів, що є вирішальним фактором для селекціонерів, які прагнуть підвищити ефективність виробництва.

Дослідження показали, що використання PCAI може принести кілька економічних переваг. Підвищуючи ефективність кнурів і роблячи бажану генетику більш доступною, PCAI зменшує потребу утримувати велику кількість кнурів на фермах. Це не тільки знижує витрати, пов'язані з утриманням кнурів, але й сприяє більш широкому використанню високоякісного генетичного матеріалу, що призводить до генетичного вдосконалення промислових свиней.

Ключові слова: штучне осіменіння свиноматок, традиційне цервікальне осіменіння (CAI), постцервікальне штучне осіменіння (PCAI), мета-аналіз, частота опоросів, індекс плодючості, розмір гнізда.

Artificial insemination (AI) has been a widely-used practice in pig breeding since the 1930s, but it wasn't until the 1980s that it saw significant commercial use in the industry. Artificial insemination has been an important part of pig breeding for decades, with traditional cervical insemination (CAI) being the most common method used in commercial settings. However, post-cervical AI (PCAI) has been proposed as an alternative method that can reduce insemination time, decrease the number of sperm required per dose, and improve time management for pig farms. The insemination procedure typically involves depositing a dose of semen into the ca of the cervical canal using cervical insemination (CAI), with 2.5-4.0 billion spermatozoa being used per insemination in an extender (70-100 ml). A non-surgical technique for depositing semen into the uterus was proposed by Hancock in 1959 (Hancock, J., & Hovell, G., 1961). This technique, known as post-cervical AI (PCAI), reduces insemination time, allows for a reduction in total sperm number per dose (and it allows for reducing the loss of sperm by backflow (Bortolozzo, F., et al., 2015).), and improves time management for pig farms, resulting in significant productivity gains. By reducing the number of spermatozoa required per dose, PCAI saves on some consumables for artificial insemination and reduces the cost of keeping boars while making desirable genetics more affordable, making it an efficient and effective tool for improving pig breeding efficiency.

The technology of post-cervical insemination, in comparison with traditional cervical insemination, has some specific features, which are related to inner catheter insertion. The cervix must be relaxed for the successful insertion of the internal catheter. Therefore, having the boar present during the process is not recommended. After inserting the basic catheter, it is necessary to wait for 1-3 minutes to allow the cervix to relax before inserting the internal catheter. Typically, insemination is performed for a group of sows at the same time, so while waiting for the cervix to relax, the basic catheters can be inserted for the next sows. It is recommended to allow sows to have contact with the boar after insemination, as this can stimulate uterine motility and improve the transport of sperm to the upper third of the oviduct, where fertilization takes place.

However, the different studies presented controversial results of this method, and the different methodological aspects of research hindered independent analysis. A meta-analysis is a tool capable of creating a comprehensive assessment of results through the data integration of different publications.

In view of this, the aim of the study was to analyze the effects of the post-cervical insemination method in terms of more effective use of boar semen using reproduction parameters (farrowing rate, litter size, and fecundity index of sows).

Materials and methods. The purpose of the meta-analysis was to compare the effectiveness of sows' cervical and post-cervical artificial insemination (by using three reproduction parameters: farrowing rate, fecundity index, and the litter size). The meta-analysis was conducted using Jamovi software and algorithms of the metaphor package (R) algorithms, with publication bias estimation (The Jamovi project, 2022; R Core Team, 2021; Viechtbauer, W., 2010; Francis, G., 2013). The articles selected to create the database were. The following criteria were used for the inclusion of publications in the meta-analysis:

- The study should not involve exogenous hormonal effects on the reproductive system
- The study must have at least 20 animals in each group;
- Information regarding the size of the control and experimental groups, the volume of insemination doses, the total sperm count in one dose, and variation statistics should be available, which are essential for meta-analysis.

We conducted a search of relevant keywords: ("sows" OR "swine" OR "pig") AND ("cervical" OR "post-cervical" AND "artificial insemination» on online databases,

including PubMed and Google Scholar. We then examined the sources cited in the received articles and used them in our study if they were available in full-text versions and contained non-redundant data. In cases where a study included data from multiple experiments, each experiment was considered separately (Table 1). The random effect model was used.

Table 1

Characteristics of included studies

Author, year(n CAI, n PSAI)	Group CAI		Group PSAI	
	Sperm cells in insemination dose, bn	Volume of insemination dose, ml	Sperm cells in insemination dose, bn	Volume of insemination dose, ml
Will K. J., 2021, (158, 90)	1.5	50	1.5	50
Will K. J., 2021, (159, 97)	2.5	80	2.5	80
Singh M., 2020, (40, 30)	1.5	80	1.5	80
Singh M., 2020, (40, 30)	3	80	3	80
Apić J., 2015, (30, 30)	2	50	2	50
Apić J., 2015, (30, 30)	4	50	4	50
Apić J., 2015, (30, 30)	4	100	4	100
Apić J., 2015, (30, 30)	2	100	2	100
Pylypenko S.V., 2006, (25, 25)	2	100	2	20
Kovalenko V.F., 2005, (25, 25)	2	100	2	20
Ternus E. M., 2017, (273, 279)	2.5	80	1.5	45
Llamas-López P. J., 2019, (130, 1036)	2.5	85	1.5	45
Suárez-Usbeck A., 2019, (324, 248)	3	90	1.5	45
Cane F., 2019, (280, 280)	3	100	1.5	50
Hernández-Caravaca I., 2012, (1716, 1683)	3	80	1	25
Hernández-Caravaca I., 2012, (1716, 1664)	3	80	1.5	40
Hernández-Caravaca I., 2017, (38, 104)	3	80	1.5	40
Hernández-Caravaca I., 2017, (38, 38)	3	80	1.5	40
Hernández-Caravaca I., 2017, (38, 42)	3	80	1.5	40
Hernández-Caravaca I., 2017, (47, 56)	3	80	1.5	40
Hernández-Caravaca I., 2017, (47, 54)	3	80	1.5	40
Hernández-Caravaca I., 2017, (47, 63)	3	80	1.5	40
Mellado M., 2018, (1773, 7078)	3	90	3	90
Sbardella P. E., 2014, (165, 165)	3	90	1.5	45
Pearodwong P., 2020, (88, 124)	3	100	1.5	50
Pearodwong P., 2020, (83, 129)	3	100	1.5	50
Fitzgerald R. F., 2008, (196, 193)	3	100	3	100
Dimitrov S., 2009, (49, 67)	3	100	1.5	50
Dimitrov S., 2009, (51, 33)	3	100	1.5	50
Roberts P.K., 2005, (859, 924)	3	80	1	80
Serret C.G., 2005, (95, 83)	3.5	100	2	50
Serret C.G., 2005, (95, 77)	3.5	100	1	50
Serret C.G., 2005, (95, 79)	3.5	100	0.5	50
Llanes Chalé J. E., 2007, (1074, 1510)	4	100	0.5	50

Farrowing rate. We performed a meta-analysis of the farrowing rate using the log odds ratio as the outcome measure. A random-effects model was utilized to analyze the data. The amount of heterogeneity (i.e., tau^2) was estimated using Hedges' estimator (Hedges, 1985). Along with the tau^2 estimate, we reported the Q-test for heterogeneity (Cochran, 1954) and the I^2 statistic. If any level of heterogeneity was detected (i.e., $tau^2 > 0$, regardless of the Q-test results), a prediction interval for the true outcomes was calculated. Studies with a Cook's distance larger than the median plus six times the interquartile range of the Cook's distances were considered influential. To check for funnel plot asymmetry, we used the rank correlation test and the regression test with the standard error of the observed outcomes as predictors. In total, we included k=34 studies in the analysis, with a total of n (CAI) = 9962 and n (PSAI) = 16582 for the farrowing rate.

Fecundity index. We conducted an analysis of the sow's fecundity index using the standardized mean difference (d Hedges') as the outcome measure. We fitted a random-effects model to the data. In total, we included k=33 studies in the analysis, with a total of n (CAI) = 8111 and n (PSAI) = 9348 for the fecundity index.

The litter size. We analyzed the variation in the litter size between the study groups of CAI and PSAI insemination using the standardized mean difference as the outcome measure. We fitted a random-effects model to the data. The number of studies included in the analysis was the same as that described in the previous section.

Results

Farrowing rate. The observed log odds ratios for farrowing after PSAI insemination compared to CAI inseminations ranged from -2.1647 to 1.0561, with the majority of estimates being positive for PSAI (56%). The estimated average log odds ratio based on the random-effects model was 0.0061 (95% CI: -0.2042 to 0.2163) (Fig. 1). Therefore, the average outcome did not significantly differ from zero ($z = 0.0564$, $p = 0.9550$). According to the Q-test, the true outcomes of farrowing rates appeared to be heterogeneous ($Q(33) = 68.7814$, $p = 0.0003$, $tau^2 = 0.1852$, $I^2 = 76.4080\%$). A 95% prediction interval for the true outcomes was -0.8631 to 0.8752. Hence, although the average outcome was estimated to be positive, it did not significantly differ from zero, and in some studies, the true outcome may be negative. An examination of the studentized residuals revealed that none of the studies had a value larger than ±3.1804, indicating no outliers in the context of this model.

According to Cook's distances, two studies (Serret C.G., 2005, (95, 83); Serret C.G., 2007, (95, 79)) could be considered overly influential. A funnel plot for estimating the systematic error associated with publication bias (Fig. 2) revealed a low probability of such error. Neither the rank correlation nor the Egger regression test indicated any funnel plot asymmetry ($p = 0.4437$ and $p = 0.0567$, respectively).

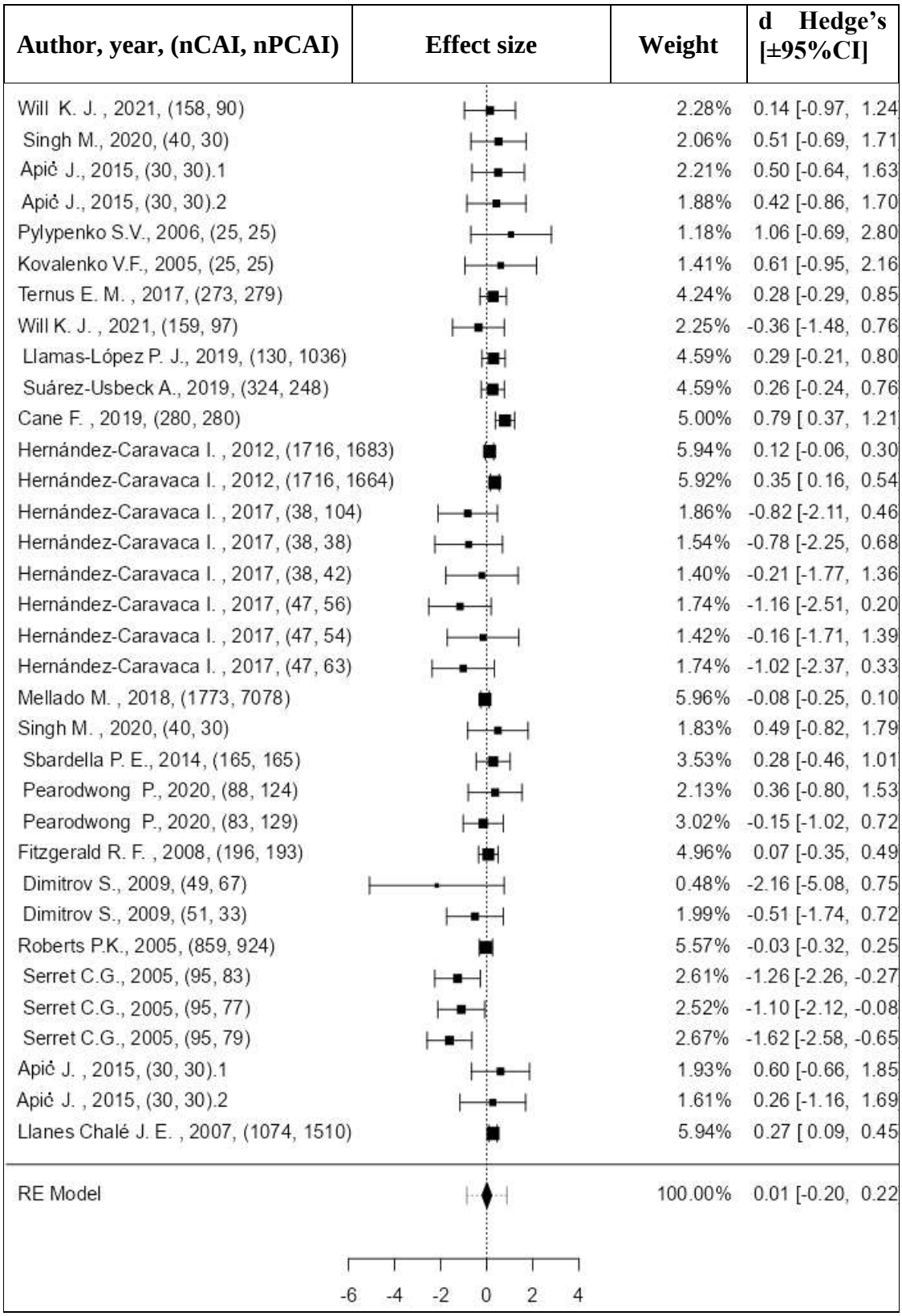

Fig 1. **Forest plot showing effects of artificial insemination method of sows on farrowing rate using the log odds ratio as the outcome measure. Figure lists number sows in groups (nCAI and nPCAI), d Hedge's and 95% confidence interval and the weight accounting for the total statistics (weight).**

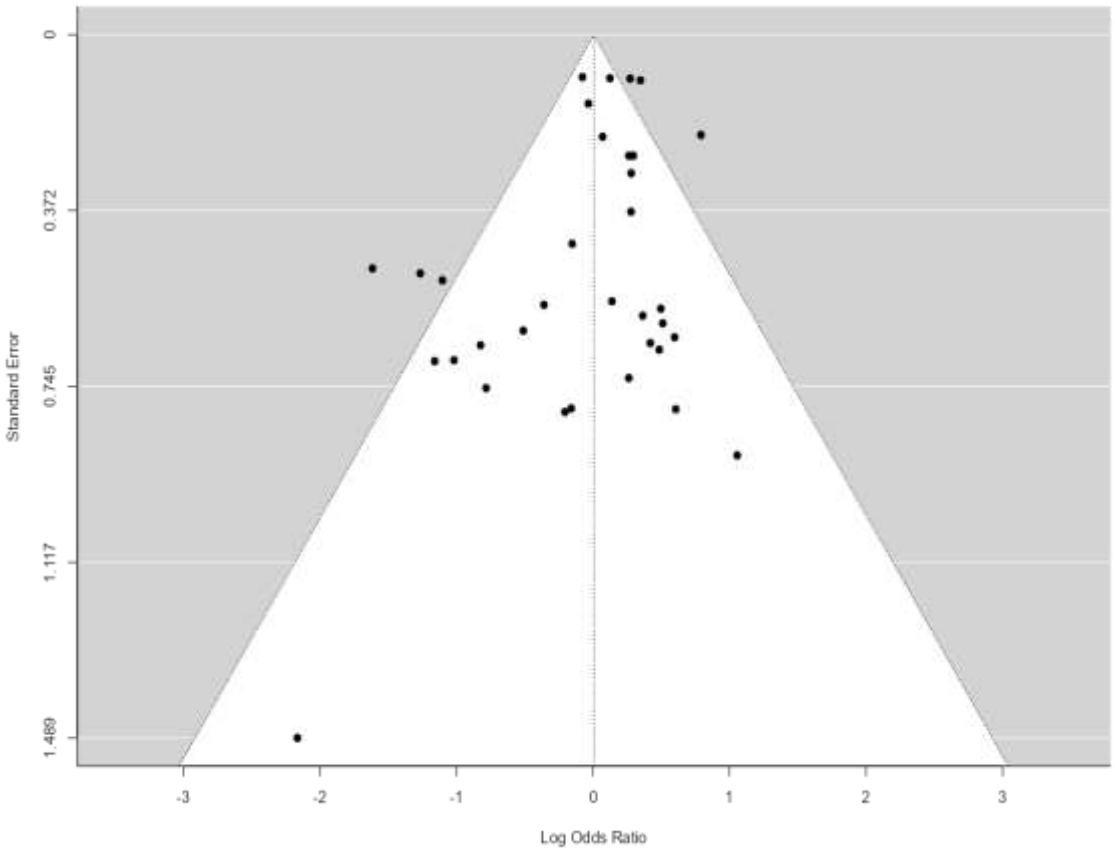

Fig. 2. **A funnel plot for estimating the farrowing rate systematic error associated with publication bias. The middle line is the effect size and the side two lines are the corresponding confidence ranges.**

Fecundity index. The observed standardized mean differences ranged from -0.7213 to 1.8843, with 52% of the estimates being positive. The estimated average standardized mean difference based on the random-effects model was 0.1156 (95% CI: -0.0790 to 0.3103) (Fig. 3). Therefore, the average outcome did not differ significantly from zero ($z = 1.1642$, $p = 0.2443$). According to the Q-test, the true outcomes appear to be heterogeneous ($Q(32) = 295.6778$, $p < 0.0001$, $tau^2 = 0.2926$, $I^2 = 97.0358\%$). A 95% prediction interval for the true outcomes is given by -0.9623 to 1.1935. Neither the rank correlation nor Egger's regression test indicated any funnel plot asymmetry ($p = 0.3165$ and $p = 0.0877$, respectively) (Fig. 4).

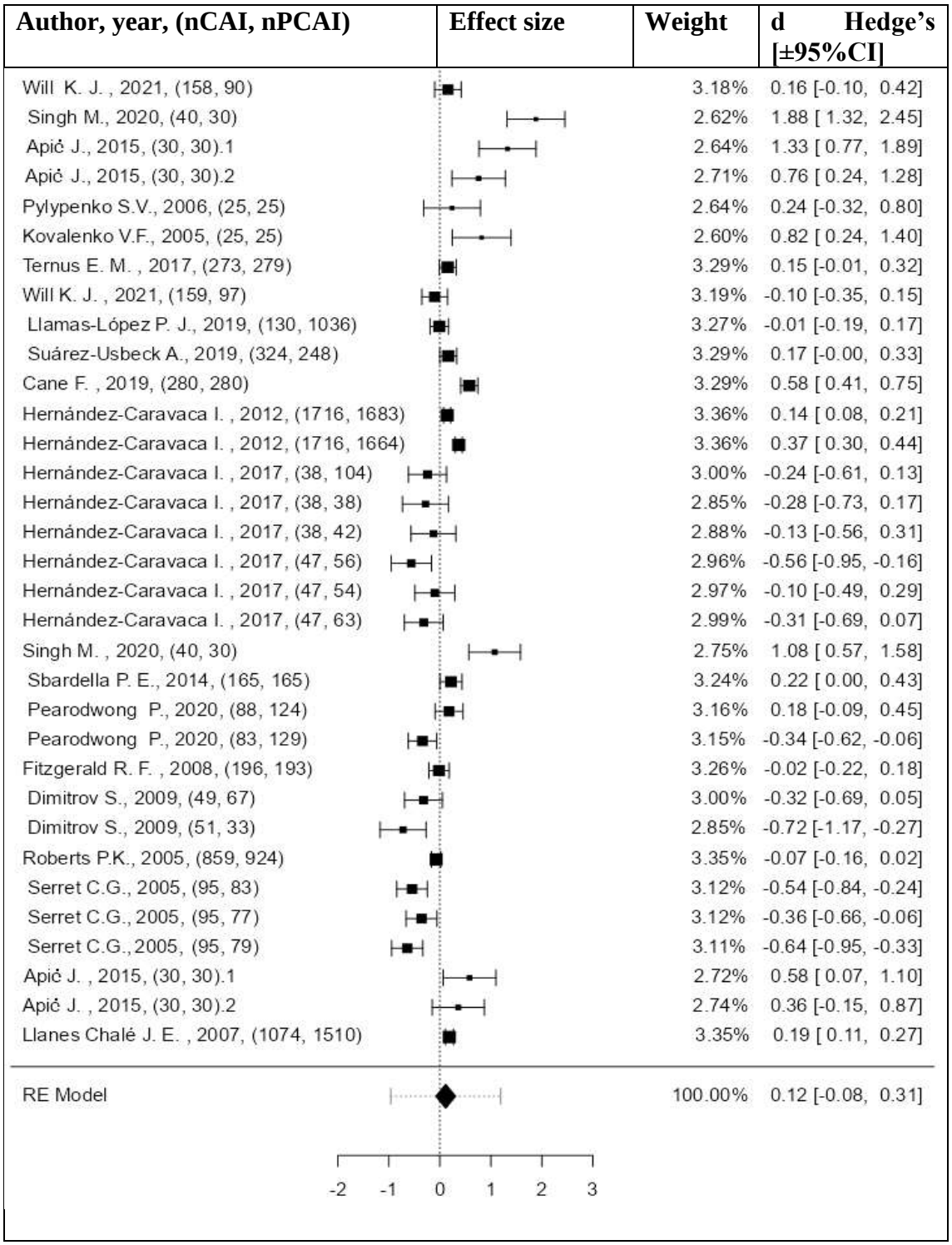

Fig. 3. **Forest plot showing effects of artificial insemination method on sow's fecundity index the standardized mean difference as the outcome measure. Figure lists number sows in groups (nCAI and nPCAI), d Hedge's and 95% confidence interval and the weight accounting for the total statistics (weight).**

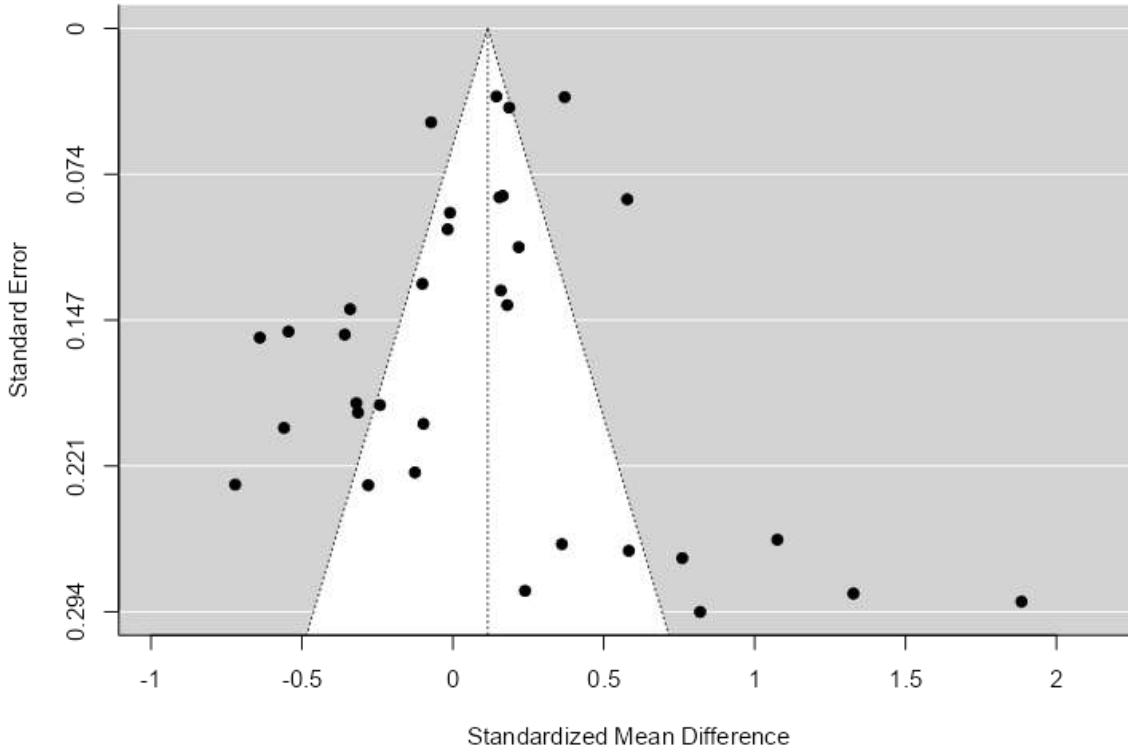

Fig. 4. **A funnel plot for estimating the sows' fecundity index systematic error associated with publication bias. The middle line is the effect size and the side two lines are the corresponding confidence ranges.**

The litter size. The observed standardized mean differences ranged from -0.4466 to 1.1583, with positive estimates in 52% of cases. The estimated average standardized mean difference, based on the random-effects model, was 0.0226 (95% CI: -0.0670 to 0.1123) (Fig. 5). Therefore, the average outcome did not differ significantly from zero ($z = 0.4950$, $p = 0.6206$). According to the Q-test, the true outcomes appear to be heterogeneous ($Q(32) = 71.9705$, $p < 0.0001$, $tau^2 = 0.0435$, $I^2 = 83.0904\%$). A 95% prediction interval for the true outcomes is given by -0.3958 to 0.4410. Neither the rank correlation nor Egger's regression test indicated any funnel plot asymmetry ($p = 0.2736$ and $p = 0.3391$, respectively) (Fig. 6).

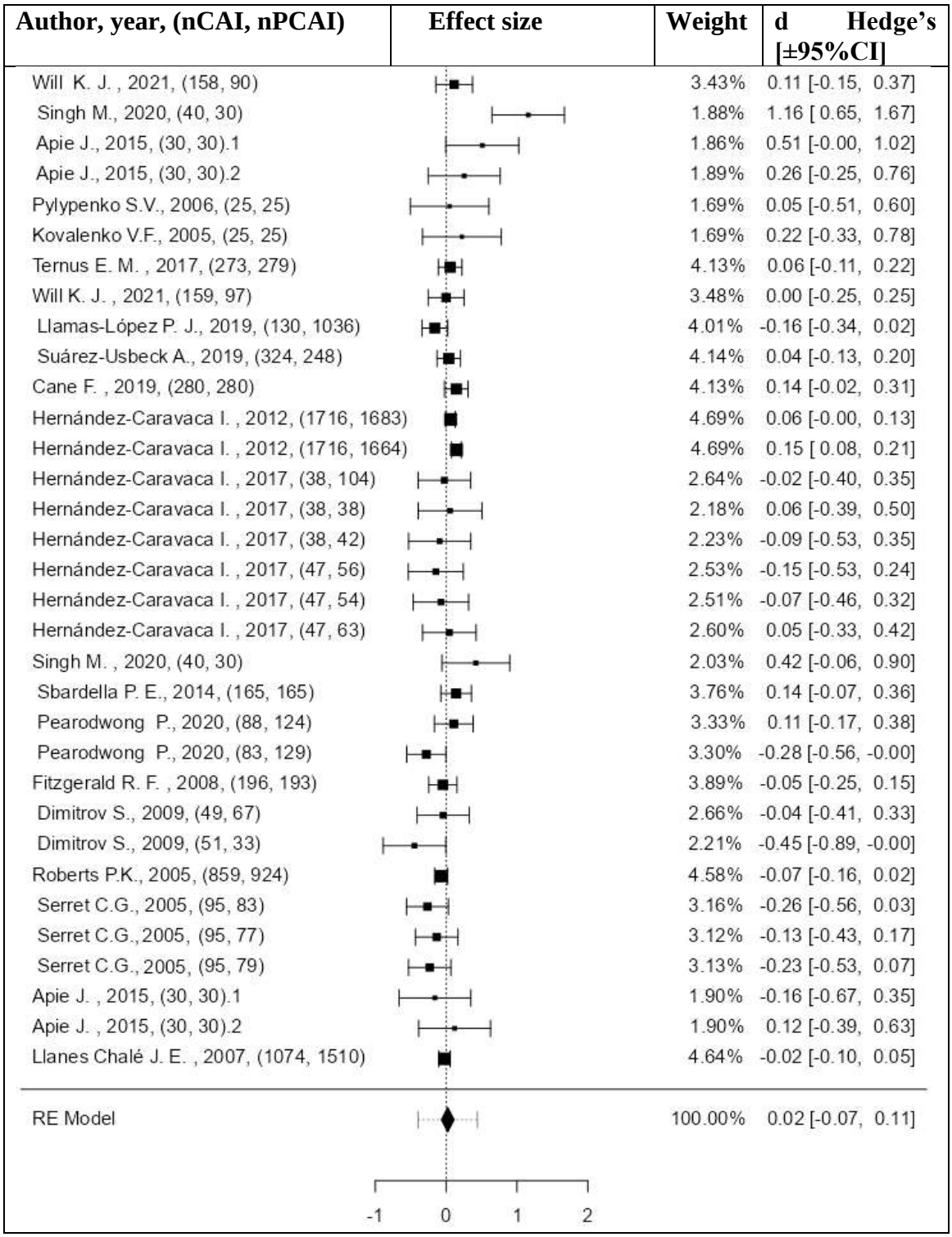

Fig. 5. **Forest plot showing effects of sows artificial insemination method on litter size the standardized mean difference as the outcome measure. Figure lists number sows in groups (nCAI and nPCAI), d Hedge's and 95% confidence interval, and the weight accounting for the total statistics (weight).**

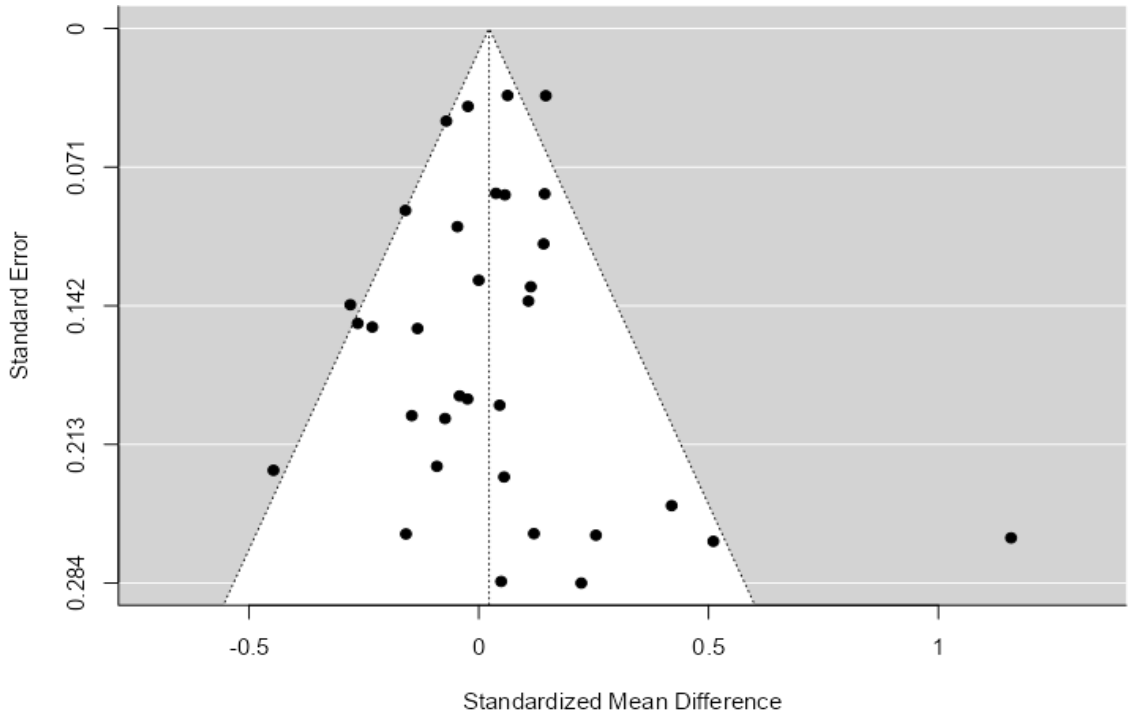

Fig. 6. **A funnel plot for estimating the litter size's systematic error associated with publication bias. The middle line is the effect size and the side two lines are the corresponding confidence ranges.**

Discussion

The present study demonstrates that none of the investigated parameters (including the farrowing rate, the fecundity index, and the litter size) decreased with the use of the post-cervical insemination method. Therefore, based on the data from various authors analyzed in this meta-analysis, we can conclude that this method offers all the advantages without any loss of reproductive efficiency.

From a methodological standpoint, our dataset was not stratified based on the number of spermatozoa in the semen dose or the volume of the dose when comparing the two methods.

In the analyzed dataset, the number of sperm per dose for traditional insemination ranged from 1.5 billion to 4 billion and the volume of the dose was from 50 to 100 ml. Realistically, industrial doses of less than 70 ml and a total sperm count of less than 2.5 billion are not used. Usually, breeding companies use about 3 billion sperm per dose and a volume of 80-100 ml, this is an effective way to get a good level of fertility in group insemination. So, (Knox R.V., 2016;. Roca J., et al., 2006) wrote about 3 billion and 80 ml. The number of sperm per dose for post-cervical insemination ranged from 0.5 billion to 4 billion and the volume of the dose was from 20 to 100 ml. The actual industrial use is about 1.5 billion sperm (Feitsma H., 2009) and the volume of the dose is 35-50 ml.

We separated each group into subgroups where the grouping factor was the number of spermatozoa in the insemination dose. For CAI group, these were concentrations of 1.5-2 million (n=6), 2.5-3 million (n=22), and >3 million (n=6) (Fig. 7, a, b, c). For the PCAI group, these were concentrations of 0.5-1 million (n=5), 1.5-2 million (n=23), and >2 million (n=6) (Fig. 7 (d, e, f). The significance of differences between groups was evaluated by ANOVA (Tables 2, 3).

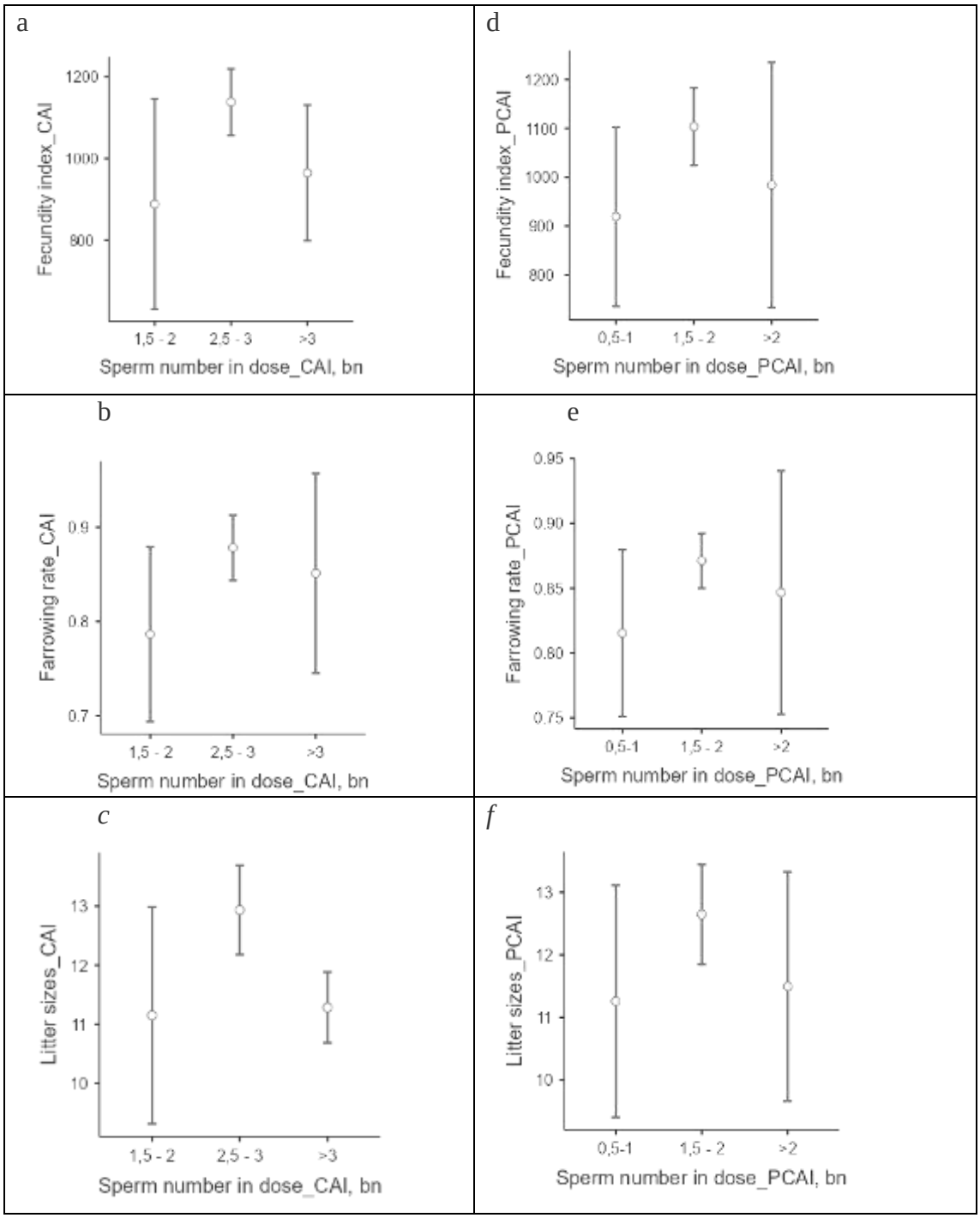

Fig. 7. **Fecundity index (a, d), farrowing rate (b, e), and litter sizes (c, f) for CAI and PSAI sow artificial insemination in groups with different total sperm numbers in one dose. Means and 95% CI are shown.**

Table 2

One-way ANOVA (Fisher's) result of the effect of sperm numbers per dose on reproductive performance in sows at PCAI

Variables	**F**	**df1**	**df2**	**p**
Farrowing rate	2.08	2	31	0.142
Fecundity index	2.45	2	31	0.103
Litter sizes	1.87	2	31	0.171

Table 3

One-way ANOVA (Fisher's) result of the effect of sperm numbers per dose on reproductive performance in sows at CAI

Variables	F	df1	df2	p
Farrowing rate	2.84	2	31	0.074
Fecundity index	5.01	2	31	0.013
Litter sizes	4.56	2	31	0.018

In all cases, the absolute values of average groups were optimal. The lowest values were in the case of the smallest sperm counts per dose. However, the differences in fecundity index and litter sizes were significant in the CAE group Post-cervical insemination offers several benefits in pig breeding, including increased control over semen placement, reduced use of semen material, the potential for increased genetic diversity, and improved herd performance. These advantages make it a popular choice for many farms looking to enhance their breeding programs.

In recent years, there has been a growing interest in the use of post-cervical insemination in the pig industry. The need for more efficient breeding programs has driven the adoption of this technology, as it allows farmers to achieve the same or better results with less effort and resources.

As technology and equipment continue to improve, the use of post-cervical insemination is likely to increase further. It is also expected to play a crucial role in the speed of selective breeding and genetic improvement, making it a valuable tool for the pig industry.

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As technology and equipment continue to improve, the use of post-cervical insemination is likely to increase further. It is also expected to play a crucial role in the speed of selective breeding and genetic improvement, making it a valuable tool for the pig industry.

Conclusion. Thus, according to the results of the meta-analysis of the available published data, the method of post-cervical insemination of pigs does not lead to a deterioration of reproductive performance and allows the use of its technological, economic, breeding, and genetic advantages in commercial pig breeding. Also, according to the results obtained, there is no reliable systematic error associated with publication bias.

References

Apić, J., Vakanjac, S., Stančić, I., Radović, I., Jotanović, S., Kanački, Z., Stanković, B. (2015). Sow fertility after insemination with varying doses of volume and spermatozoa count. Turkish Journal of Veterinary and Animal Sciences, 39(6), 709–713. https://doi.org/10.3906/vet-1503-50.

Bortolozzo, F., Menegat, M., Mellagi, A., Bernardi, M., & Wentz, I. (2015). New Artificial Insemination Technologies for Swine. *Reproduction in Domestic Animals*, 50(S2), 80–84. https://doi.org/10.1111/rda.12544

Cane, F., Pereyra, N., Cane, V., Patricia, M., Teijeiro, J. M. (2019). Improved farrowing rate using intrauterine insemination in sows. *Revista Mexicana De Ciencias Pecuarias*, 10(3), 583–594. https://doi.org/10.22319/rmcp.v10i3.4772.

Dimitrov, S., Zmudzki, J. (2009). Post-cervical insemination of sows with low sperm concentration dose in the commercial pig farm. *Bulletin of the Veterinary Institute in Pulawy*, 53(2), 225–228.

Feitsma H. (2009). Artificial insemination in pigs, research and developments in The Netherlands, a review. *Acta Scientiae Veterinariae*, 37 (1), 61-71. https://www.ufrgs.br/actavet/37-suple-1/suinos-07.pdf

Fitzgerald, R. F., Jones, G. F., Stalder, K. J. (2008). Effects of intrauterine and cervical artificial-insemination catheters on farrowing rate and litter size. *Journal of Swine Health and Production*, 16(1), 10–15.

Francis, G. (2013). Replication, statistical consistency, and publication bias. *Journal of Mathematical Psychology.* l, 57, 153-169. https://www.sciencedirect.com/science/article/pii/S002224961300014X?via%3 Dihub, https://doi.org/10.1016/j.jmp.2013.02.003

Hancock, J., & Hovell, G. (1961). The effect of semen volume and number of spermatozoa on the fertility of intra-uterine inseminations of pigs. *Animal Production*, 3(2), 153-161. Doi:10.1017/S0003356100033912.

Hernández-Caravaca, I., Izquierdo-Rico, M. J., Matás, C., Carvajal, J. A., Vieira, L., Abril, D., Soriano-úbeda, C., García-Vázquez, F. A. (2012). Reproductive performance and backflow study in cervical and post-cervical artificial insemination in sows. *Animal Reproduction Science,* 136(1), 14–22. https://doi.org/10.1016/j.anireprosci.2012.10.007.

Hernández-Caravaca, I., Llamas-López, P. J., Izquierdo-Rico, M. J., Soriano-Úbeda, C., Matás, C., Gardón, J. C., García-Vázquez, F. A. (2017). Optimization of post-cervical artificial insemination in gilts: Effect of cervical relaxation procedures and catheter type. *Theriogenology*, 90, 147–152. https://doi.org/10.1016/j.theriogenology.2016.11.027.

Knox R.V. (2016). Artificial insemination in pigs today. *Theriogenology,* 85(1), 83-93. https://doi.org/10.1016/j.theriogenology.2015.07.009.

Kovalenko, V., Pilipenko S. (2005). Intrauterine method of inseminating sows. Animal breeding of Russia, 11, 31.

Llamas-López, P. J., López-Úbeda, R., López, G., Antinoja, E., García-Vázquez, F. A. (2019). A new device for deep cervical artificial insemination in gilts reduces the number of sperm per dose without impairing final reproductive performance. *Journal of Animal Science and Biotechnology,* 10(1). https://doi.org/10.1186/s40104-019-0313-1.

Llanes Chalé, J. E., López, A.-A., Segura Correa, J. C., Álvarez Fleites, M. J., Góngora Castro, G. (2007). Gestation percentage and prolificacy of sows under tropical conditions using traditional and intra-uterine artificial insemination techniques. Livestock *Research for Rural Development,* 19(10), https://www.lrrd.org/lrrd19/10/llan19145.htm.

Mellado, M., Gaytán, L., Macías-Cruz, U., Avendaño, L., Meza-Herrera, C., Lozano, E. A., Rodríguez, Á. Mellado, J. (2018). Effect of climate and insemination technique on reproductive performance of gilts and sows in a subtropical zone of

Mexico. *Austral Journal of Veterinary Sciences,* 50(1), 27–34. https://doi.org/10.4067/s0719-81322018000100106.

Pearodwong, P., Tretipskul, C., Panyathong, R., Sang-Gassanee, K., Collell, M., Muns, R., Tummaruk, P. (2020). Reproductive performance of weaned sows after single fixed-time artificial insemination under a tropical climate: Influences of season and insemination technique. *Theriogenology,* 142, 54–61. https://doi.org/10.1016/j.theriogenology.2019.09.032.

Pylypenko S.V. Physiological grounds and improvement of the pigs' intrauterine insemination. Manuscript. Poltava, 2006, 32.

R Core Team (2021). R: A Language and environment for statistical computing. (Version 4.1) [Computer software]. Retrieved from https://cran.r-project.org. (R packages retrieved from MRAN snapshot 2022-01-01).

Roberts, P., Bilkei, G. (2005). Field Experiences on Post-cervical Artificial Insemination in the Sow. *Reproduction in Domestic Animals,* 40(5), 489–491. doi:10.1111/j.1439-0531.2005.00616.x.

Roca J., Vázquez J.M., Gil M.A., Cuello C., Parrilla I., Martínez E.A. (2006). Challenges in pig artificial insemination. *Reproduction in Domestic Animals*, 41 (2), 43 – 53. DOI: 10.1111/j.1439-0531.2006.00664.x.

Sbardella, P. E., Ulguim, R. R., Fontana, D. L., Ferrari, C. V., Bernardi, M. L., Wentz, I., Bortolozzo, F. P. (2014). The post-cervical insemination does not impair the reproductive performance of primiparous sows. *Reproduction in Domestic Animals,* 49(1), 59–64. https://doi.org/10.1111/rda.12224.

Serret, C. G., Alvarenga, M. V. F., Cória, A. L. P., Dias, C. P., Corcini, C. D., Corrêa, M. N., Deschamps, J. C., Bianchi, I., Jr, T. L. (2005). Intrauterine artificial insemination of swine with different sperm concentrations, parities, and methods for prediction of ovulation. *Anim Reprod,* 2(4), 250-256.

Singh, M., Mollier, R. T., Sharma, P. H. R., Chaudhary, J. K. (2020). Reproductive performance in cervical and postcervical artificial insemination (PCAI) with liquid boar semen in Gunghroo × Hampshire crossbreed pig in Nagaland. Indian Journal of Animal Sciences, 90(5), 708–711. https://doi.org/10.56093/ijans.v90i5.104610.

Suárez-Usbeck, A., Mitjana, O., Tejedor, M. T., Bonastre, C., Moll, D., Coll, J., Ballester, C., Falceto, M. (2019). Post-cervical compared with cervical insemination in gilts: Reproductive variable assessments. *Animal Reproduction Science,* 211. https://doi.org/10.1016/j.anireprosci.2019.106207.

Ternus, E. M., Vanz, A. R., Lesskiu, P. E., Preis, G. M., Serafini, L., Consoni, W., Traverso, S. D., Cristani, J. (2017). Reproductive performance of gilts submitted to post-cervical artificial insemination. Arquivo Brasileiro de Medicina Veterinaria e Zootecnia, 69(4), 777–784. https://doi.org/10.1590/1678-4162-9285.

The Jamovi project (2022). Jamovi. (Version 2.3) [Computer Software]. Retrieved from https://www.jamovi.org.

Viechtbauer, W. (2010). Conducting meta-analyses in R with the metafor package. *Journal of Statistical Software,* 36, 1-48. https://www.jstatsoft. org/article/view/v036i03, https://doi.org/10.18637/jss.v036.i03

Will, K. J., Mellagi, A. P. G., Bernardi, M. L., Bortolozzo, F. P., Ulguim, R. D. R. (2021). Perspectives of intrauterine artificial insemination applicability in gilts. Ciencia Rural, 51(5). https://doi.org/10.1590/0103-8478cr20200612.

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INNOVATIVE TECHNOLOGIES IN EQUINERY

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The research is devoted to a review of innovative technologies used in the world horse breeding. Innovations include ensuring a long, healthy life for horses, precision medicine, laser and PRP therapy, rehabilitation and training programs, monitoring media platforms, genetic and biotechnological methods, development of ecological and safe coatings, use of recycled materials for buildings, automated microclimate systems and waste collection, automatic feed dispensers with customized schedules, real-time feed and water consumption monitoring tools.

The most revolutionary ideas concern the development of smart technologies for horse training with monitoring of physical condition. Such developments include wearable GPS tracking devices and heart rate monitors, trackers for tracking horse movement over distances, satellite tracking technologies, saddle pads with sensors of active points on the horse's body, a horse movement analysis system, 3D imaging and motion capture tools, and mounting headsets for athletes. Much attention is paid to the safety of equestrian sports, especially children's and parasports, in connection with which protective equipment has been developed (helmets made of modern materials, pneumatic jackets and vests). An important sector of innovation in horse breeding is technologies that have a positive impact on the environment - the use of environmentally friendly materials and construction technologies in the construction of stables and sports arenas, the use of ecological bedding, manure composting systems with subsequent processing into organic fertilizers, energy-efficient design, the use of renewable energy, landscaping of horse breeding areas. It is noted that the horse breeding of the future, along with all areas of human activity, is closely related to the further development and integration of technologies.

According to the results of the research, the prospects for the development of technologies in horse breeding are outlined.

Keywords: horses, equestrian industry, equestrian sport, innovative technologies, monitoring

ІННОВАЦІЙНІ ТЕХНОЛОГІЇ У КОНЯРСТВІ

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Дослідження присвячені огляду інноваційних технологій, що застосовуються у світовому конярстві. Інновації стосуються забезпечення тривалого здорового життя коней, презицизійної медицини, лазерної та PRP-терапії, реабілітаційних і тренувальних програм, моніторингових медіа-платформ, генетичних та біотехнологічних методів, розробки екологічних та безпечних покриттів, застосування перероблених матеріалів для будівель, автоматизовані системи мікроклімату і прибирання відходів, автоматичні дозатори кормів із налаштованими графіками, моніторингові інструменти споживання корму та води в реальному часі.

Найбільш революційні ідеї стосуються розвитку розумних технологій тренінгу коней з моніторингом фізичного стану. До таких розробок відносяться натільні пристрої GPS-відстеження і монітори серцевого ритму, трекери відстеження пересування коней на дистанції, супутникові технології стеження, килимки під сідло з датчиками активних точок на тілі коня, систему аналізу рухів коня, інструменти 3D-зображень і зйомки рухів, гарнітури кріплення для спортсменів. Багато уваги приділяють безпеці кінного спорту, особливо дитячого та параспорту, у зв'язку з чим розроблено засоби захисту (шлеми з сучасних матеріалів, пневматичні куртки та жилети). Важливим сектором інновацій у конярстві є технології, що позитивно впливають на екологію - використання екологічно чистих матеріалів і будівельних технологій при будівництві стаєнь і спортивних манежів, застосування екологічної підстилки, системи компостування гною з подальшою переробкою в органічні добрива, енергоефективний дизайн, застосування відновлюваної енергії, озеленіння територій конярських підприємств. Зазначено, що конярство майбутнього поряд з усіма сферами діяльності людини, тісно пов'язане із подальшим розвитком та інтеграцією технологій.

За результатами досліджень окреслено перспективи розвитку технологій у конярстві.

Ключові слова: коні, кінна індустрія, кінний спорт, інноваційні технології, моніторинг.

Introduction. Ukraine is a country with ancient traditions and a rich history of horse breeding and horse breeding. Despite the current challenges caused by the war, the industry is promising. With its development, the requirements for the technology of running the industry, innovative solutions for ensuring all elements of keeping, training, protection, and monitoring the productivity of horses will also grow. The horse industry

in developed countries of the world is currently undergoing a significant transformation, new high-tech means of monitoring the health and fitness of horses are being developed, robotic technologies for keeping, training, and restoring the body of horses after physical and nervous stress are being introduced. Innovative technologies not only increase the quality and accuracy of methods, but also open up new prospects for the development of scientific thought regarding improving the sports results of horses. Therefore, innovative technologies should radically and positively change the horse industry, including in Ukraine.

Given the relevance of this issue, the purpose of the work was to study innovative technologies used in the horse industry of the world.

Materials and methods. A systematic review of literature sources was carried out by searching for publications in the databases Scopus, Web of Sciences, Google Scholar, etc., published in Ukrainian and English in recent years in accordance with the inclusion criteria. The systematization of published data was carried out with the aim of accumulating modern scientific knowledge on innovative technologies in horse breeding for further building our own methodological basis for research in this area in domestic conditions, taking into account international experience.

Research results. The countries with the largest number of horses in the world are the USA (over 10 million), China (9 million), Mexico (6.4 million), Brazil (5.5 million), Argentina (3.6 million). In total, according to FAO data (DAD-IS, FAO 2023), there are about 60 million horses of more than 200 breeds in the world. More than half of them are kept on the American continent (55.2%).

The horse breeding of Ukraine is currently represented by a horse population of about 200 thousand individuals, with about 3 thousand of breeding animals identified as pedigree (Tkachova I. V., 2024). Most of the horses are used (agricultural, rental, tourist, hobby-class horses), horses used in sports - about 20%. The leading horse breeds are the Ukrainian riding horse, thoroughbred riding horse, Ukrainian and Oryol trotter, Novooleksandrivskaya heavy-duty. Recently, riding horses of European selection have been gaining considerable popularity, which are actively used in sports.

The priority in the horse industry is to ensure a long healthy life for horses, and in this direction, innovative tools and methods of diagnostics, treatment and health prevention have developed significantly (Zhang, Y., & Liu, H., 2020). These tools allow for faster and more accurate detection of health problems in horses that were previously difficult to detect (for example, tendon damage that is not visible on X-rays). Video surveillance equipment allows the veterinarian to remotely monitor the health of the horse, which is effective in sparsely populated regions. Recently, precision medicine has been increasingly used in veterinary medicine - laser and PRP therapy (platelet-rich plasma) of tendons and joints of horses that are exposed to significant physical exertion, accelerating recovery time after strenuous training and competitions. The advantages of laser therapy are that it is non-invasive and stimulates blood circulation and metabolic activity using intense, focused light. PRP is an injection treatment that promotes healing by delivering a concentrated dose of platelet-rich plasma to the damaged area of the body. The use of platelet-rich plasma and allogeneic mesenchymal stem cells in veterinary therapy promotes the healing of tendons and ligaments, however, this sometimes triggers an immune response in the horse, destroying the added stem cells. According to studies of the last decade (Trelford C.B. et all., 2022; Deng Z. et all., 2024), the addition of the growth factor TGF-ß2 successfully blocks this immune response, with the survival of treated stem cells being 50% higher than that of untreated cell cultures. Preliminary observations suggest that in the future this therapy may be used to treat similar injuries in humans.

Innovative approaches are also being used in the treatment of musculoskeletal injuries in horses, ensuring full restoration of functionality. Therefore, hydrogels and bioactive dressings are being improved, which provide a favorable environment for wound healing, while preventing the formation of granulation tissue (Fatima A. et all., 2024). Rehabilitation also plays a crucial role for sports horses, as it helps to prevent re-injury and prolong active life (Annamiyradov K. et all., 2025). Specially designed training programs using innovative tools that take into account the individual characteristics of each horse can significantly increase its physical fitness and performance (Kurhaluk N. et all., 2023). It is also important to pay attention to the psycho-emotional state of horses, as stress and anxiety can negatively affect the recovery process. Here, innovative means of creating comfortable conditions for rapid recovery come to the rescue. In addition, an important aspect of rehabilitation is teaching owners the correct methods of caring for animals during the recovery period. This includes recommendations on diet, physical activity and regime, which allows to provide optimal conditions for recovery. Developed applications for gadgets and media platforms have brought communication between horse owners, veterinarians and trainers to another level, exchanging information and learning from each other's experiences and knowledge (Broms, L. et all., 2021; Hii C. et all., 2020). Monitoring the processes occurring with the horse (training regimen, rations and feeding schedules, medical history, pregnancy progress, etc.) in real time can be carried out more effectively and organized thanks to a mobile application on a regular smartphone or tablet. For example, the “Sleip” smartphone application uses a video of a horse in motion to track the movement of certain points of the body for further evaluation of the data by the veterinarian and trainer (Fig. 1).

Fig. 1. **The Sleip smartphone application for step-by-step analysis of a horse’s condition in real time (photo from the Sleip website)**

Due to the high value of individual horses (outstanding sports horses, especially valuable stallions and mares, rare breeds), reproductive technologies are being developed

to increase fertility and further improve genetic selection (Palmer E. & Chavatte-Palmer P., 2020). The most important effect of new technologies is their contribution to genetic selection and genetic progress (increasing genetic value in a population from one generation to the next). Innovations in biotechnology – cell division, embryo transplantation, cloning, preservation of biological material in vitro – allow to accelerate the improvement of horses (Cabeza J. P. & Gambini A., 2023). Embryo transfer technology in equine reproduction is not very new and has been used quite successfully to reproduce the outstanding Landim horses (Alvarenga F.C. et all., 2008), but it is constantly being improved to increase survival and maintain viability during transportation. Thus, recently it has become possible to store embryos at 5°C for 12-24 hours and transport them to the transplantation site. Embryo freezing has become more common using the technique of vitrification of embryos >300 μm or dehydration of embryos >300 μm before freezing. The ability to wash oocytes from mares and fertilize them by sperm injection has revolutionized the approach of veterinarians to infertility in mares and stallions (Squires E. et all., 2020). Despite the complexity of implementation and the high cost of innovative reproductive technologies, they are a promising tool that can have a positive impact on the preservation of horse populations that are under threat of extinction and reproduce unique individuals (Turghan M. A. et all., 2022).

Regarding horse keeping technologies, the development of innovative floor coverings can be highlighted, because traditional materials - concrete, wood - have a negative effect on the joints and hooves of horses. Innovative materials provide shock absorption, hygiene, better grip (prevention of slipping), easy cleaning, and therefore - reduced veterinary costs. Indoor arenas ensure the comfort of sports horses and riders, environmental friendliness, and the ability to train in any season and weather in optimal conditions. In an indoor training room, it is possible to provide comfortable temperature, humidity, lighting, install smart microclimate systems, and cleaning (McGill S. et all., 2023). Modern climate control systems play an important role in the horse keeping system, which is especially relevant in conditions of climate change and in difficult natural and climatic regions. Automated climate control systems provide comfortable living conditions in horse facilities and equestrian facilities regardless of the external environment. They create optimal temperature and humidity indicators, improve air quality through filtration, reduce respiratory problems and heat stress, improve the comfort of horses during rest and recovery from stress in any climatic conditions. Animal husbandry systems are closely related to environmental safety issues. Environmental safety is a priority issue of the modern world, and the horse industry is no exception, although it is one of the safest for the environment (Kic P. et all., 2024). In this direction, innovative solutions include the use of environmentally friendly local, as well as recycled and recovered materials and construction technologies in the construction of horse-breeding facilities, which has a positive impact on environmental safety, as well as energy-efficient design - maximizing natural lighting and ventilation, the use of renewable energy devices (solar panels and wind generators), landscaping the territory of the horse-breeding enterprise, which has a positive impact on both the environment and the psycho-emotional state of horses and people. An interesting environmental innovation is a comfortable, well-absorbing bedding for horses made of recycled paper or hemp, which is more profitable than traditional straw. Horse manure composting systems that convert waste into highly nutritious fertilizers for the soil also require attention, which solves the problems of pasture fertilization and prevents pollution of the stable area and the environment.

Innovative horse feeding systems play a vital role in rations for horses of different purposes and workloads, maximizing bioconversion and reducing feed waste. Advanced

feeding systems are revolutionizing our approach to horse feeding, offering precision and efficiency that were previously unattainable. Innovative horse feeding systems include automatic feed dispensers with customizable schedules, real-time feed and water consumption monitoring tools, and integration with health monitoring systems for personalized feeding plans. These systems not only ensure that horses are getting the right amount of food at the right time, but also provide valuable data to veterinarians and nutritionists to optimize horse diets.

Individual balancing of horse diets is not a new process, however, in recent years, various manufacturers have offered a huge number of feed additives aimed at leveling oxidative stress, which contribute to the rapid recovery of the body, improving sports performance and health (Andriychuk A.V. et all., 2021). Such additives contain amino acids, vitamins, macro- and microelements, Omega-3 fatty acids and other elements that protect cell membranes, structural proteins, DNA and contribute to excellent health and high sports performance of horses. Recently, more and more animal nutrition products have been developed using nano-technologies. Such products, as prebiotics and probiotics, help solve problems in the digestive system of horses caused by pH instability, improve nutrient absorption and strengthen immunity. Today, the market offers a huge range of horse feed products and it is developing with the progress of scientific research into the physiology of horse nutrition and the chemical industry. In this situation, the horse owner needs to decide what exactly and in what quantities his horses need, and what can harm their health and turn out to be just an unnecessary expense. Timely hydration is of great importance in the daily routine of the stable and the comfort of the horses, and therefore the introduction of controlled drinkers is an important element of technology. An innovation in this process are non-contact ultrasonic sensors, similar to those of the Cascada Automatic Waterer, which monitor hydration using algorithms based on data collected over a long period of time, with information available via smartphone from anywhere in the world (Fig. 2). The device is attached to a stable wall, holds a 20-liter bucket and can be configured to heat the water flow to the desired temperature to prevent freezing.

Fig. 2. **Cascada Automatic Waterer**

A real revolution in horse training technology has been the introduction of wearable GPS tracking devices and heart rate monitors (Horsepal), which allow real-time monitoring of the horse's speed, distance and heart rate during training, and help the trainer adjust the training. The development of wearable devices (smart halters, limb sensors) has changed the level of horse health management, such devices provide real-time data on the horse's vital signs, activity level and sleep pattern (Fig. 3).

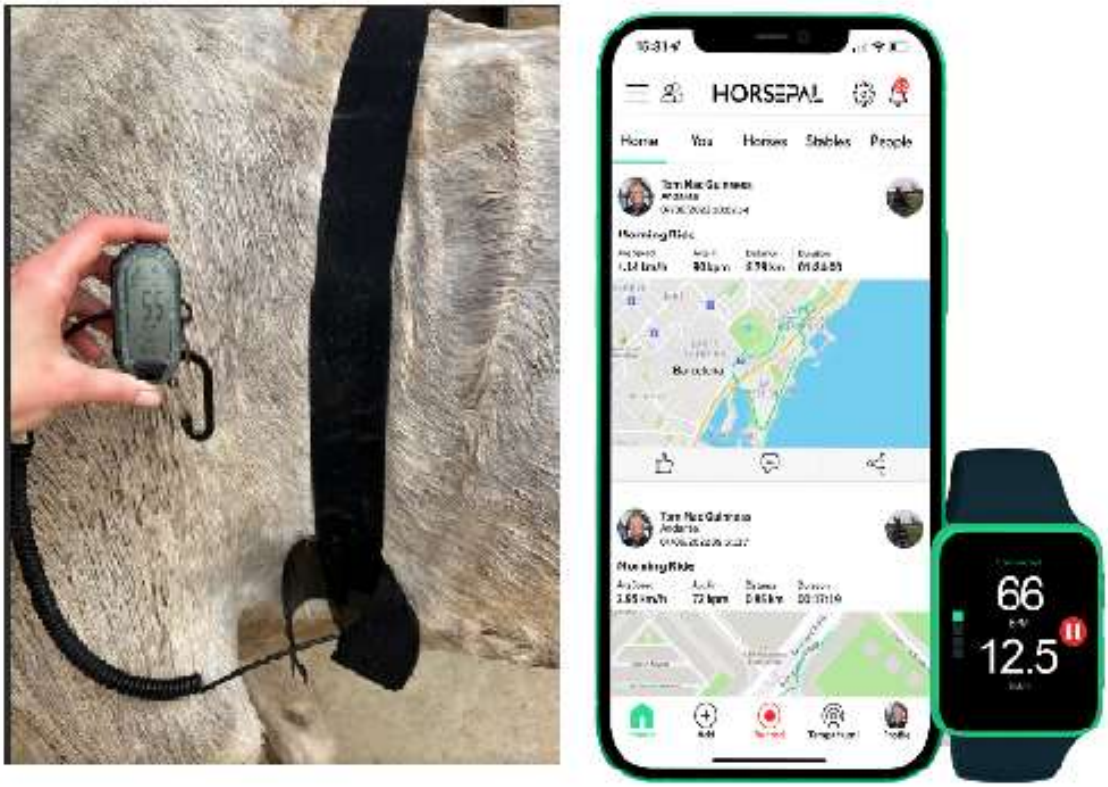

Fig. 3. **GPS tracking device and heart rate monitor (Horsepal)**

GPS tracking devices for distance analysis such as (EQUIMO type trackers) help to track how a horse moves over a distance on a racetrack or competition field and monitor liveliness, distance, height, angle and specific turns, etc. (Fig. 4). The tracker determines the time spent on each gait to adjust the optimal pace of movement over the distance. The recording function allows you to compare sessions to determine trends and progress. By measuring and analyzing precise changes in limb extension and joint flexion, trainers know what the horse is feeling. The value of these measurements is that they are completely unbiased and eliminate any subjectivity.

In recent years, the horse identification system has become mandatory to implant electronic chips into the horse's muscles with unique numbers, which allows for quick identification of horses. Connecting such chips to a GPS satellite navigation system makes it possible to track the movements of horses, especially when kept in herds or tracking feral populations (for example, the Przewalski's horse population in the Chernobyl Exclusion Zone).

Fig. 4. **Sensor for a horse's limb**

Interesting devices are saddle pads with sensors (such as Estride with 900 sensors) for monitoring saddle pressure, which measure pressure points on the horse's back during riding and jumping. Such pads control whether the saddle is properly fitted, which helps to avoid discomfort for the horse and back injuries. Thermography with a heat sensor tracks pain points due to ill-fitting equipment or excessive pressure (weight) which leads to increased blood flow and body temperature in the affected area of the horse's body. The readings from these sensors can also be useful for manufacturers of equine equipment for further adjustment and improvement (Figure 5-6).

Fig. 5. **Sensors on the saddle and key points of the horse's movement (photo from the Fairfax website)**

Fig. 6. **Horse gait analysis mapping (photo from Fairfax Saddles Ltd)**

Horse gait analysis systems (such as StrideSAFE carriers) allow for accurate assessment of horse gait, pre-purchase lameness detection, and general gait monitoring (Calle-González, N., 2024). Objectively identifying potential problems such as uneven strides, lameness, asymmetry, or gait inefficiency assists in the subjective examination of horses by veterinarians. This technology can also be used to detect hidden injuries. A horse will move in a way that minimizes discomfort, so naturally, if it is in pain, it will choose a different mode of movement. The StrideSAFE system analyzes about 3,000 readings per second and captures even the smallest changes in the horse's gait, helping to detect potential injury before the rider feels the change.

The use of 3D imaging tools makes it possible to analyze the conformation of the horse, identifying parts of the body where there has been an improvement in musculature or movement, as well as to monitor the ontogenesis of foals. In this direction, very interesting research was carried out by Giraudet C. et all. (2023), who developed a system based on underwater camera markers and cameras on the horse's body during hydrotherapy of sport horses (Fig. 7).

In addition, 3D motion capture (Mocap) systems are used to assess the biomechanics of a horse during a jump, identifying inefficiencies, stress zones and risk factors for injury. Cameras are placed around the sports field and track the movement of reflectors attached to the horse's body, accumulating data on limb movement, joint angles, and overall symmetry (Fercher, C., 2024). The Mocap device is the most accurate for measuring movement analysis and is used by athletes, but it is expensive and difficult to set up, requiring special training for the user. A more economical analogue of this device are inertial measurement unit (IMU) sensors, which are small wireless devices that measure acceleration, orientation, movements of different parts of the body, and provide information about joint angles (Parmentier J. I. M. et all., 2023). Thus, Hatrisse C. and colleagues (2022) used an inertial measurement unit located on the pasterns, hooves, and withers of trotting horses to determine gait phases on different sections of the racetrack with different track surfaces and set the load on the limbs (McGill S., 2023). In the training system of equestrian athletes, a very interesting niche is increasingly occupied

by virtual reality technologies in the form of video monitors and headsets attached to the athlete, reacting to body movements and rein pressure. The devices simulate the movements of a horse and allow the athlete to improve riding and jumping skills, orientation on the competition site or on the racetrack track where he plans to perform. Using this innovation allows the athlete to train without unnecessary strain on the horse and additional risk of injury. In addition, virtual reality systems allow people to feel like riders when it is impossible to engage in real horseback riding.

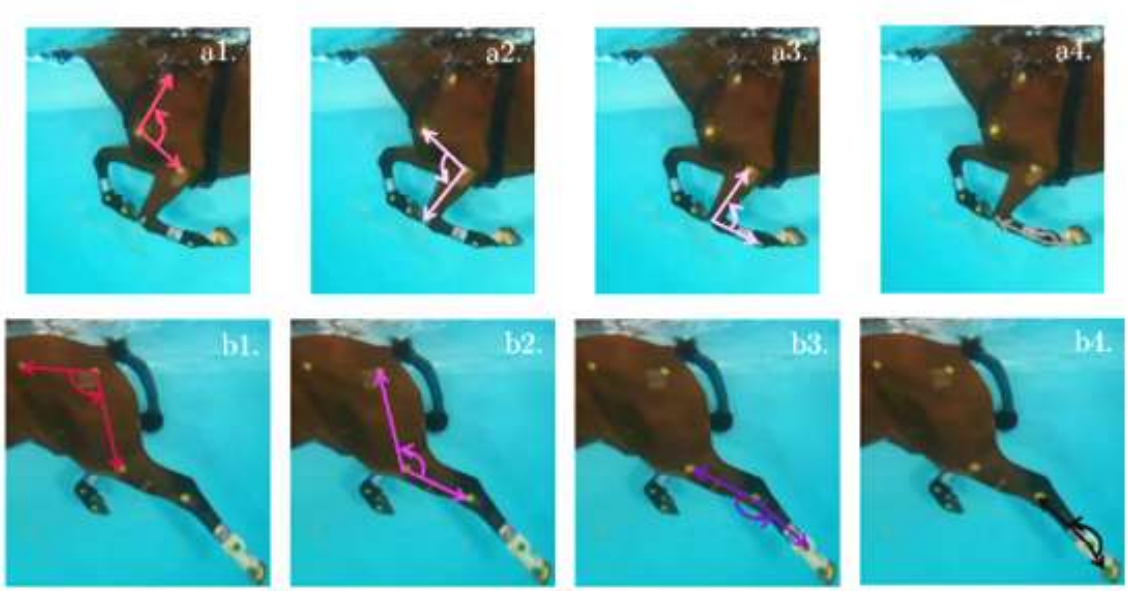

Fig. 7. **Calculating the anatomical angles of a horse's limbs during hydrotherapy using 3D reconstruction (Giraudet C. et all., 2023)**

An urgent problem is the safety of athletes, especially children and people with disabilities, because equestrian sports are considered one of the most dangerous sports. Therefore, various companies producing sports equipment and gear are working on creating and improving protective equipment for equestrian athletes. A good example is the Point Two USA campaign, which created innovative pneumatic jackets and vests for riders like airbags in cars (Dyer R. F. & Irizarry R. P., 2014). Riding helmets are made of modern materials (advanced polycarbonate), they combine lightness, maximum impact resistance, durability, advanced design. Such helmets have a carefully designed ventilation system, sun visors. Some models are equipped with a built-in microchip, onto which you can download data about the rider's health using the KER program, which can speed up the provision of first aid and in addition - equipped with video cameras for further analysis of the performance

In recent years, the use of unmanned aerial vehicles in economic activities and the agricultural sector in particular has developed significantly. Unmanned aerial vehicles can be very useful for filming and further studying the horse's test route, recording the course of the race (Fig. 8), as well as when overcoming obstacles, which allows you to monitor jumps from different angles and perform a detailed analysis of the jump model and the overall sports performance of the rider-horse pair.

Fig. 8. **Recording of hippodrome races using a drone (image created using AI)**

Management of a horse breeding enterprise is becoming increasingly efficient thanks to the introduction of satellite platforms that provide information on land use optimization, the state of the forage crop harvest, pasture health, control of horse movement during grazing, and allow you to track weather conditions for planning outdoor activities (Lupoae O.-D., 2024). Maintaining pedigree, accounting and other documentation, exchanging information between horse owners and stud inspectors, etc. on paper is a thing of the past thanks to the spread of software and mobile technologies (Johnson, M. & Carter, R., 2021). Management takes place in an automated mode in real time, which allows for rapid receipt and analysis of information.

As a result of the conducted research, it can be stated that innovative technologies introduced into the horse industry are directed in the directions presented in Table 1.

Discussion. According to the results of research, innovative technologies in horse breeding can be divided into separate blocks:

- innovative tools and diagnostic methods, means of treatment and prevention of horse health (precision medicine, modern drugs for treatment and prevention, means of protection and rehabilitation of horses);
- innovations in genetics and reproduction (genetic selection, DNA technologies, embryo transplantation, cloning);
- innovations in housing technology and environmental safety (use of recycled materials for the construction of stables and arenas, use of modern floor covering materials, introduction of renewable energy sources and waste disposal technologies);
- innovations in feeding (innovative feeds enriched with probiotics, vitamin-mineral premixes, preparations for strengthening immunity);
- digital technologies, automation and robotization of technological processes (software platforms for monitoring the condition of horses in real time, activity sensors, feeding and watering monitoring systems, robotic cleaning systems, microclimate regulation systems);
- automated management in real time.

Innovative achievements not only improve the lives of horses and the sporting achievements of riders, but also revolutionize the approach of equestrian professionals to their work. From small family farms to large competitive horse breeding enterprises, the impact of modern technologies is increasingly developing throughout the world's equestrian industry.

Table 1

Innovative technologies in horse breeding

Direction of development	Innovative technologies, means
Instruments and diagnostic methods, means of treatment and prevention of horse health	Precision medicine, modern drugs for treatment and prevention, means of protection and rehabilitation of horses
Genetics and reproduction	Genetic improvement programs, DNA technologies, embryo transfer, cloning
Comfortable life of horses, environmental safety,	Environmentally safe materials for buildings, floors, bedding
Healthy feeding, economical hydration systems	Innovative safe and nutritious feeds enriched with probiotics, vitamin and mineral premixes, preparations for strengthening immunity, controlled drinkers with moisture sensors, heart rate monitors
Digital technologies, artificial intelligence in training horses	GPS tracking devices, smart equipment for horses, trackers, satellite systems, 3D imaging and video recording tools, virtual reality technologies
Equestrian safety	Innovative equipment for riders (impact-resistant helmets with health analyzers, pneumatic jackets and vests)
Surveillance systems	Use of unmanned aerial vehicles to monitor training, movement, and horse protection
Automated management	Management of a horse breeding enterprise on satellite platforms using programs adapted to business conditions

Technologies are progressing and revolutionizing our daily lives, providing us with more information about health, finances and the world around us, while making it easier to perform tasks that may have seemed impossible a few years ago. The future of horse breeding, like any other field of human activity, is closely linked to the further integration of technologies. The development of new methods of genetic analysis, reproductive technologies, the creation of new materials and smart management systems, and the improvement of environmental sustainability allow the horse breeding industry to be in demand and competitive.

Conclusion. The modern horse industry in the world is constantly improving due to the development and implementation of innovative technologies that concern all sectors of the industry - genetics and reproduction, maintenance and feeding, training, monitoring and management. The priority in the horse industry is to ensure a long healthy life for horses, which is ensured by developments in veterinary medicine and pharmacology, genetics and biotechnology, nutrition physiology and feed production, nanotechnology and nanomaterials, and the use of artificial intelligence in monitoring systems and management of horse breeding enterprises.

References

Andriychuk A.V., Tkachenko G.M., Tkachova I. V., Kurgalyuk N. M., Vartovnyk M. S., Kulibaba R.O. (2021). Antioxidant mechanisms of adaptation of horses in the process of physical training: monograph [ed. I. V. Tkachova, G. M. Tkachenko. K.: Agrarna nauka, 2021. 232 p.

Annamyradov K., Rovshenkulyev B., Bailiev B., & Djumaniyazov E. (2025). Therapy and rehabilitation: how veterinary medicine helps restore animal health. *Symbol of science*, (1-1-2), 111-113.

Broms, L., Bentzen, M., Radmann, A., and Hedenborg, S. (2021). Stable Cultures in Cyberspace. Scandinavian Sport Studies Forum issn 2000-088x volume eleven. Available online at: www.sportstudies.org

Cabeza Juan P., Gambini A. (2023). Advancements and challenges in in vitro reproductive technologies for the conservation of equine species, Theriogenology Wild, Volume 2, 2023, 100036, ISSN 2773-093X, doi:10.1016/j.therwi.2023.100036.

Calle-González, N., Lo Feudo, C. M., Ferrucci, F., Requena, F., Stucchi, L., & Muñoz, A. (2024). Objective Assessment of Equine Locomotor Symmetry Using an Inertial Sensor System and Artificial Intelligence: A Comparative Study. *Animals*, *14*(6), 921. https://doi.org/10.3390/ani14060921

Chapman, M., Fenner, K., Thomas, M. J. W., & Thompson, K. (2024). Stakeholder Views on the Potential Benefits and Feasibility of an Equestrian Industry-Specific Health, Safety and Welfare Management System. ***Animals***, ***14***(23), 3450. doi:10.3390/ani14233450

DAD-IS, FAO 2023. 〈https://www.fao.org/dad-is/breed-diversity/en/〉. (Accessed 10 March 2025).

Dyer R. F. & Irizarry R. P. (2014). Point Two USA: Marketing an Innovation in Equestrian Safety in Equestrian Safety. *Case Studies in Sport Management*, Vol. 3: Issue 1: 54–67. doi:10.1123/cssm.2014-0021.

Deng Z., Fan T., Xiao C., Tian H., Zheng Y., Li C., He J. (2024). TGF-β signaling in health, disease, and therapeutics. *Signal Transduct. Target Ther*. Mar 22;9(1):61. doi: 10.1038/s41392-024-01764-w

Fatima A., Muhammad S.A., Mateen A. (2024). Innovative approaches in equine wound management: Addressing challenges and their remedies. Int. J. Vet. Sci. Res. 10(2): 016-020. doi:10.17352/ijvsr.000145

Fercher, C., Bartsch, J., Kluge, S., Schneider, F., Liedtke, A. M., Schleichardt, A., & Ueberschär, O. (2024). Applying Multi-Purpose Commercial Inertial Sensors for Monitoring Equine Locomotion in Equestrian Training. *Sensors*, *24*(24), 8170. https://doi.org/10.3390/s24248170

Giraudet, C., Moiroud, C., Beaumont, A., Gaulmin, P., Hatrisse, C., Azevedo, E., Denoix, J.-M., Ben Mansour, K., Martin, P., Audigié, F., Chateau, H., & Marin, F. (2023). Development of a Methodology for Low-Cost 3D Underwater Motion Capture: Application to the Biomechanics of Horse Swimming. ***Sensors***, ***23***(21), 8832. https://doi.org/10.3390/s23218832

Hatrisse, C., Macaire, C., Sapone, M., Hebert, C., Hanne-Poujade, S., De Azevedo, E., Marin, F., Martin, P., & Chateau, H. (2022). Stance Phase Detection by Inertial Measurement Unit Placed on the Metacarpus of Horses Trotting on Hard and Soft Straight Lines and Circles. *Sensors*, *22*(3), 703. https://doi.org/10.3390/s22030703

Hii, C., Dhand, N. K., Toribio, J.-A. L., Taylor, M. R., Wiethoelter, A., Schembri, N., et al. (2020). Information delivery and the veterinarian-horse owner relationship in the context of Hendra virus in Australia. Prevent. Vet. Med. 179, 104988. doi: 10.1016/j.prevetmed.2020.104988

Johnson, M. & Carter, R. (2021). Digital Innovations in Equine Management. *Journal of Equine Science*, Vol. 12(4): 89–102.

Kic P., Wohlmuthová M., & Starostová L. (2024). Effects on the Indoor Environment in a Stable for Horses in Winter: A Case Study. ***Agriculture***, ***14***(8), 1287. https://doi.org/10.3390/agriculture14081287

Kurhaluk N., Tkaczenko H., Tkachova I., Lukash O. (2023). Lipid peroxidation and the total antioxidant capacity in the plasma of Shetland pony mares and stallions involved in recreational horseback riding: role of photoperiod and exercise. *The Scientific and Technical Bulletin of Livestock farming institute of NAAS*, Is. 130: 112-127. doi:10.32900/2312-8402-2023-130-112-127

Landim-Alvarenga F.C., Fernandes C.B., Devito L.G., Derussi A.A.P., Blanco I.D.P., Alvarenga M.A. (2008). New assisted reproductive technologies applied to the horse industry: successes and limitations. *Anim. Reprod*., vol. 5, № 3/4: 67-82.

Lupoae O.-D., Wilk V., Radu R. I. (2024). Sustainable entrepreneurship in equine services. *Journal of Business Research*, Vol. 170, 114361, ISSN 0148-2963, doi:10.1016/j.jbusres.2023.114361.

McGill S., Coleman R., Jackson J., Tumlin K., Stanton V. and Hayes M. (2023) Environmental spatial mapping within equine indoor arenas. Front. Anim. Sci. 4:1083332. doi: 10.3389/fanim.2023.1083332

Palmer E., Chavatte-Palmer P. (2020). Contribution of Reproduction Management and Technologies to Genetic Progress in Horse Breeding, *Journal of Equine Veterinary Science*, Vol. 89, 2020, 103016, ISSN 0737-0806, doi:10.1016/j.jevs.2020.103016.

Parmentier, J. I. M., Bosch, S., van der Zwaag, B. J. *et al.* (2023). Prediction of continuous and discrete kinetic parameters in horses from inertial measurement units data using recurrent artificial neural networks. *Sci Rep.* 13, 740 https://doi.org/10.1038/s41598-023-27899-4

Squires E. (2020). Current Reproductive Technologies Impacting Equine Embryo Production. *Journal of Equine Veterinary Science*, Vol. 89, 102981, ISSN 0737-0806, https://doi.org/10.1016/j.jevs.2020.102981.

Tkachova I. V. (2024). Genetic resources of horses in Ukraine and directions of their effective use. *«One World – One Health». Proceedings of the I International Scientific and Practical Conference*, 4-5 June 2024, Słupsk, Poland. Słupsk: Institute of Biology, Pomeranian University in Słupsk, 2024, 287-293.

Trelford C. B, Dagnino L., Di Guglielmo G. M. (2022). Transforming growth factor-β in tumour development. *Front. Mol. Biosci*. 2022 Oct 4;9:991612. doi: 10.3389/fmolb.2022.991612

Turghan, M. A., Jiang, Z., & Niu, Z. (2022). An Update on Status and Conservation of the Przewalski's Horse (*Equus ferus przewalskii*): Captive Breeding and Reintroduction Projects. *Animals***,** *12*(22), 3158. https://doi.org/10.3390/ani12223158

Zhang, Y., & Liu, H. (2020). Sustainable Practices in Modern Horse Breeding. *Equine Veterinary Journal*, Vol. 9(2): 35–47.

DOI 10.32900/2312-8402-2025-133-122-133
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SORGHUM IS A PROMISING FEED RESOURCE FOR ANIMAL HUSBANDRY IN THE CONTEXT OF CLIMATE CHANGE
(Review article)

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The article is devoted to an overview of trends, volumes and state of sorghum cultivation in Ukraine and on a global scale. Sorghum is one of the most important forage, industrial and food crops, which is characterized by high drought resistance, ecological plasticity and versatility of use. It ranks fifth among the most widespread grain crops in the world, grown in more than 85 countries on an area of about 50 million hectares. The main producers are the United States, Australia, Argentina, China, Mexico and Japan.

Ukraine is one of the key European producers of sorghum, actively introducing modern agricultural technologies and increasing its export potential. Despite the temporary reduction in acreage, they are projected to expand significantly in the coming years.

Historical aspects of sorghum domestication and the variety of its use are traced. Sorghum has a long history, it was grown in Ancient Egypt more than 3000 years ago. It has spread all over the world, adapting to different climatic conditions. Thanks to breeding achievements, various types of sorghum were bred: grain, sugar, broom and fodder, each of which has its own specific application.

The article considers the features and advantages of this promising crop for our climatic conditions. Attention is focused not only on the botanical features of sorghum as a fodder crop in the agro-industrial complex of Ukraine, but also on the diversity of its species forms and hybrids.

In particular, the main directions of breeding sugar and grain sorghum are analyzed. A comparative analysis of the chemical components of grain sorghum and the content of individual mineral elements in seeds is carried out.

The main advantages of sorghum in terms of its resistance to diseases and pests in comparison with the range of forage crops similar in botanical characteristics are evaluated.

The article highlights the experience of specialists in using sorghum in feeding farm animals, and its impact on product quality. In animal husbandry, sorghum is used as a highly nutritious feed containing protein, starch and trace elements, which helps to improve the productivity of livestock. Its grain is actively used in the production of mixed feed, gluten-free products and bioethanol, and green mass for the production of silage.

Keywords: sorghum, crop distribution, breeding directions, hybrids, soriz, sorghum grain, sorghum silage, animal feeding.

СОРГО – ПЕРСПЕКТИВНИЙ КОРМОВИЙ РЕСУРС ДЛЯ ТВАРИННИЦТВА В УМОВАХ КЛІМАТИЧНИХ ЗМІН (ОГЛЯДОВА)

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Стаття присвячена огляду тенденцій, об'ємів і стану вирощування сорго в Україні та у світовому масштабі. Сорго – одна з найважливіших кормових, технічних і продовольчих культур, яка вирізняється високою посухостійкістю, екологічною пластичністю та універсальністю використання. Воно займає п'яте місце серед найбільш поширених зернових культур світу, вирощується у понад 85 країнах на площі близько 50 млн га. Головними виробниками є США, Австралія, Аргентина, Китай, Мексика та Японія.

Україна є одним із ключових європейських виробників сорго, активно впроваджуючи сучасні агротехнології та нарощуючи експортний потенціал. Незважаючи на тимчасове скорочення посівних площ, прогнозується їхнє суттєве розширення у найближчі роки.

Простежено історичні аспекти одомашнення сорго та різноманіття його використання. Сорго має давню історію, його вирощували ще в Давньому Єгипті понад 3000 років тому. Воно поширилось по всьому світу, адаптуючись до різних кліматичних умов. Завдяки селекційним досягненням було виведено різні види сорго: зернове, цукрове, віничне та кормове, кожен із яких має свою специфіку застосування.

У статті розглянуто особливості та переваги цієї перспективної для наших кліматичних умов культури. Сконцентровано увагу не лише на ботанічних особливостям сорго як кормової культури в агропромисловому комплексі України, але й на різноманітті його видових форм та гібридів.

Зокрема проаналізовано основні напрямки селекції сорго цукрового та зернового. Проведено порівняльний аналіз хімічних складових зернового сорго та вмісту окремих мінеральних елементів в насінні.

Оцінено основні плюси сорго щодо його стійкості до хвороб та шкідників порівняно зі спектром подібних за ботанічними характеристиками кормових культур.

Висвітлено досвід фахівців в використанні сорго в годівлі сільськогосподарських тварин, та його вплив на якість продукції. У тваринництві сорго використовується як високопоживний корм із вмістом білка, крохмалю та мікроелементів, що сприяє покращенню продуктивності худоби. Його зерно активно застосовують у виробництві комбікормів, безглютенових продуктів і біоетанолу, а зелена маса для виготовлення силосу.

Ключові слова: сорго, поширення культури, напрями селекції, гібриди, сориз, зерно сорго, силос з сорго, годівля тварин.

We live in times of change that occur regardless of our attitude to them. And in such conditions, it is important not only to be able to adapt to new challenges, but also to

try to stay ahead of them. This also applies to the agricultural sector, as one of the most traditional in the global economy.

The use in agricultural production of crops characterized by high yields, resistance to adverse environmental conditions, pests and diseases is one of the sources of increasing the volume of feed production and strengthening the feed base.

Sorghum is the most important fodder, technical and food crop. It is cultivated in many countries of the world on all continents. Different peoples of the world call sorghum culture differently: in Central Asia - jugara, in India - jovar, in China - gaolyan, hundzu, shoshu, in Korea - Su-Su, in Georgia - Gomi, in Asia Minor - Dora, in the African country - Durra, zorrat, in Spain, France - sorgo, etc. This culture got its name for its height from the Latin "sorgos" - "to rise, to rise" (Cardoso, L. M. et al., 2015). In World Agriculture, sorghum ranks fifth among the most common grain crops and is grown in 85 countries around the world on an area of about 50 million hectares. Ga (Rakshit, S. et al., 2014). Such large acreage of sorghum is associated with its unpretentiousness, high environmental plasticity and heat resistance. High productivity, feed advantages and versatility put sorghum among the most promising crops (Hossain, S. et al., 2022; Pravdyva, L. A. et al., 2023; Widodo, S. et al., 2024). Taking into account the relevance of this issue, the aim of the work was to analyze the experience of specialists in using sorghum in feeding farm animals, and its impact on product quality.

Research materials and methods. A systematic review of literature sources is carried out by searching for publications in the databases Scopus, Web of Sciences, Google Sscholar, etc., published in Ukrainian and English in recent years in accordance with the inclusion criteria. The systematization of the published data was carried out in order to accumulate modern scientific knowledge on the use of sorghum as a feed crop in cow feeding and its impact on the quality of dairy products to further build their own methodological basis for research in this direction in domestic conditions, taking into account international experience.

Research results. The world's main producers of sorghum are North America and Africa, the main exporters are the United States, Australia and Argentina, and importers are China, Mexico and Japan (Kondratiuk, S. 2018).

It should be noted that Ukraine is assigned one of the key roles in this regard, since our state is already one of the largest producers of this crop in Europe with an area of 70 thousand hectares and a production volume of 256 thousand tons. Another 18 thousand hectares are occupied by fodder sorghum crops. However, in recent seasons, its acreage has significantly decreased, which was due to the higher profitability of corn, soybeans and sunflower. In a few years,the area under sorghum will occupy at least 1-1.3 million hectares, and in 5-10 years it can partially replace corn and even sugar beet. Ukrainian farmers are actively implementing precision farming technologies, which allows them to optimize costs and increase production efficiency.

Sorghum has significant potential for development both domestically and for export. Due to its high nutritional value and versatility of use, sorghum can become a key crop for many farms. Ukrainian farmers have the opportunity to expand their sales markets, including Europe and Asia, where the demand for sorghum is constantly growing (Ozturk, I. 2018).

Sorghum is native to Equatorial Africa. Archaeological finds indicate that the practice of domestication of the culture was transferred from Egypt to Ethiopia around 3000 Years BC.E. on ancient monuments in Egypt, built long before 2200 BC, drawings of harvesting and found sorghum grain, which indicates the cultivation of this culture from time immemorial. Its antiquity is confirmed by ancient monuments in the countries of East and South Asia (Cardoso, L. M. et al., 2015; Smith, C. W. 2000). Sorghum came

to India by land and sea routes. Its cultivation in India is mentioned in Legends dating back to the First Century AD, under the name "cane barley". In this country, grain sorghum is considered the third most important food crop after wheat and rice. It is believed that India and China are secondary foci of origin and formation. It is known that when sorghum genotypes are transferred to new geographical zones, modification variability occurs. In the new conditions, the crop may be more productive and precocious than at home, as a result of which it occupies larger areas. Thus, the sorghum nevrosum race is not widespread in Africa, and in China and India it is an important national culture (Burgarella, C. et al. , 2021). Natural and artificial selection has led to the emergence of new forms that have led to the varieties of sorghum that we now grow (Ge, F. et al., 2022). From India and China, sorghum was imported to Central Asia 2-3 millennia ago.

Weed sorghum was first cultivated and grown in America and Europe earlier than all other cultivated sorghum imported later directly or indirectly from Africa. In China, the first documented mention of sorghum appears as early as the 3rd century AD.the widespread use of sorghum in Italy dates back to the late 17th century. Dried sorghum shoots were collected in the form of brooms and used to clean clothes in Italy, France and Germany. By the 1700s, the widespread use of sorghum in Europe had attracted the attention of Benjamin Franklin. During the 1960s and 1970s, production was moved to Mexico, which is still one of the most significant suppliers of sorghum to the US market. The main sorghum producing countries in southeastern Europe remain Serbia, Hungary and Turkey, with smaller crops in Romania, Bulgaria and Ukraine (Berenji, J. et al., 2011).

In North and South America, the genus Sorghum was introduced in the XVII century, simultaneously with the development of the slave trade, which took sorghum grain with them as a food product. In the United States, sugar sorghum was first imported from China via France in 1851 under the name Chinese Amber. White and brown forms of sorghum, called Egyptian corn, were imported from Egypt in 1874. Currently, the main areas under sorghum are concentrated in the" sorghum belt": the states of Nebraska, Missouri, Kansas, Oklahoma, Texas (Assefa, Y. et al., 2024; McGinnis, M. J. et al., 2020). In the United States, the greatest success has been achieved in the selection, seed production and agricultural technology of grain sorghum. Only 14% of the world's acreage is concentrated here, and the gross harvest is about 40%. Currently, the main companies in the United States engaged in the production of sorghum seeds are Monsanto, Pioneer, Pfister, Gareth and Thomas, Syngenta, Frontier. In Europe (Italy, Romania, Hungary, Albania, Bulgaria, Hungary), sorghum crops occupy about 20% of the area (5 million hectares). ha).

In the XVIII century, there was a sharp increase in the area under sugar sorghum, which is associated with its use for the production of sorghum honey (Eggleston, G. et al., 2022). The spread of sugar sorghum contributed to the construction of small sugar factories. However, by the end of the XIX century, the production of sorghum sugar was abandoned.

Interest in sorghum increased after prolonged droughts, when other crops sharply reduced yields. In the context of climate change, there is a need to reorient to more sustainable forage crops that are highly nutritious and adaptable to drought and abnormal weather events. On the one hand, sorghum has always been characterized by increased grain productivity and drought resistance, and on the other hand, it remained a secondary, poorly studied crop, which determined the opinion about its unreliability. In addition, with the advent of new white-grain varieties, the possibilities of using sorghum for starch, alcohol (Abah, C. R. et al., 2020).

There are about 50 species of sorghum in the world. Classification of sorghum was started by American breeders, who at the beginning of the 20th century divided sorghum species into four groups: grain, sugar, broom and herbaceous, identified among them ecological and geographical subgroups and varietal types (Wondimu, Z. et al., 2021). A number of requirements are put forward for modern varieties: compliance with soil and climatic conditions, high potential yield, resistance to adverse conditions, high product quality, adaptability to mechanical processing. All these tasks can be achieved by various breeding methods (selection, hybridization, polyploidy, mutagenesis, etc.). In recent years, breeders ' attention has been drawn to the prospect of creating hybrid populations (Wu, Y. P. et al., 2023).

Since 1977, new hybrid sorghum populations have been created based on phenotypically aligned but genotypically sharply different hybrid material. In sorghum breeding, a significant event is the creation of a rice-like form of sorghum (soriza), which has valuable nutritional properties. Soriz grain is characterized by good vitreous, high endosperm hardness and therefore high extrusion capacity, good taste, and resistance to diseases (Dremlyuk, H. K. et al., 2020; Voitovska, V. I. et al., 2022).

The main areas of breeding work for grain sorghum are: early ripening, suitability for mechanized harvesting, grain yield and quality (starch, protein, tannin, lysine content) (Gichile, H. 2022; Treviño-Salinas, M. et al., 2021). With the advent of such varieties, prospects open up for the use of grain for starch production, in the production of gluten-free bakery products (Szabłowska, E. et al 2021; Gómez, M. 2022; Khoddami, A, et al., 2023).

The main direction of sugar sorghum breeding is low growth, resistance to lodging, early ripening, disease damage, high yield of green mass, high content of protein and sugars in green mass (Herniwati, H. et al., 2024). The world has developed technologies for obtaining feed concentrate with a sugar content of 50-55% for a balanced sugar-protein ratio in animal feeding, production of food syrup for the confectionery industry, production of bio-alcohol, etc. (Leite, P. et al., 2020).

On average, over the years of research, in terms of nutritional value, namely the highest content of protein, fat and carbohydrates among the studied hybrids, American – Prime stood out, whose caloric content was 320 kcal., from the French – hybrid-Burggo with a caloric content of 318 kcal. The highest values of thiamine were found in the seeds of hybrids of American selection – Yuki and Prime, which amounted to – respectively – 0.43–0.45 mg/100 g. in the seeds of the American hybrid of grain sorghum – Prime, the highest biotin content was established – 0.018 mg/100 g. The studied hybrid of grain sorghum of the American selection Prime, over the years of research, was characterized by the highest na content in its composition, which was – 26 mg/100g. as a result of the research, it was revealed that the co content in all the studied hybrids of grain sorghum was in the same amount and amounted to – 2.0 mg/100 g. the highest Al content was found in the seeds of French hybrids, these indicators were at the level of 1,550 to 1,555 mg/100 g. In terms of B content, the American Yuki hybrid turned out to be the best, since the content of this trace element was 0.347 mg, while the French Burggo hybrid had the lowest boron content – 0.325 mg. The seeds of the American hybrid sorghum grain Prime 160 have the highest content of such a trace element as zinc at the level of 2.15 mg/100 g.

There are 5 types of sorghum in Ukraine, 4 of them are cultivated, mainly in the southern parts of the country. Dzhugara and sugar sorghum are grown in small areas in the steppe. Important varieties and hybrids in the steppe zone of Ukraine: Ukrainian 107, Kuban red 1677, Genichesk 1, Steppe hybrid 5, feed hybrid 5, Orange 160.

The agricultural sector of Ukraine in its functioning requires an urgent solution to the problem of further development and improvement of livestock productivity by organizing a scientifically based feed base and mainly focuses on the use of modern scientific achievements and the widespread introduction of the latest innovative and knowledge-intensive technologies aimed at actively increasing production volumes. In this regard, there is a need for special attention to technological solutions for storing sorghum grain, its effective use in feeding cattle with the study of the influence of a new feed ingredient in the diets of dairy cows on the physical and chemical parameters of milk, the physiological state of cows, their reproductive ability. Therefore, the study of the productive effect of sorghum grain when used in feeding cattle (dairy cows) as part of the feed diet is relevant.

What is the advantage of sorghum over the range of similar forage crops in biology? Grain sorghum, unlike corn, is not demanding on the presence of moisture in the soil, so it is perfectly suitable for those regions where the cultivation of moisture-loving fodder crops is difficult. Sorghum is less susceptible to pests and diseases, which not only reduces the cost of pesticides by an average of 30%, but also minimizes the risk of mycotoxins in feed. The structure of the plant does not allow fungi to actively develop (Suszkiw, J. 2023; Lee, S. et al., 2022).

Problems with water supply are extremely acute. According to the transpiration indicator, sorghum bypasses the main fodder crop - silage corn, a high yield of which can be obtained only with a high level of moisture. For example, sorghum consumes only 300 parts of water per unit of dry matter. For comparison, Sudan grass - 340, corn - 388, wheat-515.

There is no universal culture that meets absolutely all the requirements of livestock breeders. Moreover, it is also impossible to ensure a timely conveyor belt with one crop. Therefore, the selection of the most appropriate and hardy plants that can ensure the production of products despite stressful conditions is the best option. In this case, sugar sorghum is considered as the only alternative to using corn as a silage crop. After all, when it is grown outside irrigated areas, corn loses all its advantages as a highly productive plant. (Getachew, G. et al., 2016; Pujiharti, Y. et al., 2022).

In terms of the content of basic nutrients, sorghum grain is practically not inferior to corn. It is characterized by a high protein content (from 11%) and an increased lysine content (protein quality depends on this amino acid). According to scientists, increasing the amount of lysine by 0.1 g increases the level of protein digestibility per 1 g per 100 g of feed. Sorghum grains also have a high starch content (Beta, T. et al., 2000).

Sorghum contributes to the achievement of average weight gains similar to those achieved when feeding cattle on fattening grounds with a diet with corn. Sorghum not only has a positive effect on the growth and development of farm animals, but also improves the quality of meat due to its low content of polyunsaturated acids (Ochieng, B. et al., 2020; Jiao, J. et al., 2022).

There are several types of sorghum, and each has its own preferences. Grain sorghum is a fodder, food and technical crop. Sorghum grain is often used for the preparation of mixed feed, as well as as a concentrated feed not only for pigs, but also for cows, horses and poultry. The nutritional value of sorghum grains is quite high, in its composition this crop is very similar to corn and barley, which belong to grain crops. Sorghum has significantly more protein than corn, but instead it is inferior in such a factor as digestibility of substances (Ronda, V. et al., 2019).

Experts say that the use of sorghum grains in feed additives is equivalent to barley grains, pigs give the same increase and quality of meat. But there is one advantage of this crop, which is manifested in the fact that the yield of sorghum significantly exceeds spring

barley, so you can get twice as much pork from 1 hectare of sorghum as from 1 hectare of barley.

Sorghum grains contain 12-15 percent protein, about 70 percent starch, and 3.5 - 4.5 percent fat. In one hundredweight of grain, there are from 118 to 130 feed units. This indicator of nutritional value is considered quite good.

Some experts insist that the total percentage of sorghum grains in pig feed should be approximately 30-50 %, while they note accelerated growth of piglets, especially late farrowing. Pork turns out to be a dense, intense pink color and is well sold out on the market (Sotak, K. M. et al., 2015; Thomas, L. L. et al., 2020).

Sorghum grain is covered with a dense shell, during feeding it is not fully digested and transits through the gastrointestinal tract. Sorghum proteins and starches are digested in the rumen for up to two days (Yahaghi, M. et al., 2014; Risyahadi, S. T. et al., 2023). Therefore, it is better to pre-process sorghum grain before use:

- Grinding is the cheapest way. Sorghum is processed almost in the same way as corn. Good grain grinding up to 1.8 mm for better assimilation.
- Flattening, pre-treated with steam.
- Flattening of dry grain.
- Extrusion.

All these methods of pretreatment of grain give different degrees of digestibility. The "glassy" state of the endosperm slows down the breakdown of starch and proteins in the rumen, which reduces the risk of acidosis and optimizes the delivery of dietary proteins digested in the intestines.

When feeding sorghum grain, the consumption of dry matter decreases (Aguerre, M. et al., 2009). Unlike traditionally used grain types, sorghum is digested very slowly in ruminant Rumen due to the fact that sorghum starches and proteins are more resistant to enzymes. On meat animals, this is not essential, but during feeding dairy cattle and raising calves, problems may arise. When flattening with steam treatment, eating can be increased by 15%, but this treatment greatly increases the cost of food.

Sugar sorghum and sorghum-Sudan hybrid are successfully used for silage and haylage. Such top dressing is simply irreplaceable in winter in the diet of feeding pigs and other farm animals. Sorghum silage has a pleasant aroma, reminiscent of fruit, its taste qualities are quite high, so it is much better eaten by animals. The energy content in the sorghum crop is 18.3 MJ per kilogram (Jabbari, H. et al., 2011; Cattani, M. et al., 2017; Guo, F. et al., 2024).

Sorghum produces a green mass from the beginning of July to the end of August, surpassing other crops in yield. After mowing, sorghum grows quickly and vegetates until late autumn. With timely mowing for green fodder, it can produce 2-3 mowing per year.

Some sorghum varieties retain the ability to effectively silage their green mass for up to one and a half months from the moment of the optimal phase (milk-wax) of grain ripeness. This means that for Farms Limited in harvesting facilities and transport equipment, this crop is a real godsend.

Sorghum is suitable for silage. At the same time, its nutritional value will be lower than that of corn silage. The actual composition may vary depending on hybrids, harvesting dates, and storage losses.

The efficiency of sorghum silage depends on the level of livestock productivity. (Abdelhadi, L. O. et al., 2006). Experiments conducted on lactating cows proved that the complete replacement of corn silage with sorghum silage did not affect milk yield (Zhang, S. et al., 2024). However, such results were obtained by adding starch sources (such as corn flour) to the sorghum diet to compensate for the lower starch content in sorghum silage compared to corn silage. At the same time, there are opinions that changing the

type of silage and including starch sources in the diet will affect the nature of fermentation in the rumen (Pimentel, J. et al., 2013; Li, S. S. et al., 2020; Lv, X. et al., 2023).

What to be afraid of when using sorghum in feeding farm animals? It is necessary to pay attention to the sorghum variety itself. Since some varieties have a high percentage of tannins (it does not affect the feed value, but affects its digestibility and digestion, tannin itself is an astringent); monoenzymes can be added to improve digestion. In modern hybrids, the tannin index is low, on average 0.3-0.5%, which has a positive effect on feed digestion, but it is necessary to pay attention to the indicators. It is also worth considering the indicators of hydrocyanic acid - it is not allowed in sorghum. In silage, its neutralization occurs in about 1-2 weeks naturally.

Discussion. The analysis of information sources covers a wide range of issues related to the production and use of sorghum, including its history, global distribution, breeding, nutritional and feed value for animals.

Concluding the review of known sources on the use of sorghum as a priority fodder crop in the context of climate change, it should be noted that climate change will continue to affect many areas in the entire chain of feed and food production, and therefore the selection of fodder crops adapted to the impact of negative climatic factors becomes particularly relevant.

Encouraging producers to resume sorghum cultivation is based on the high potential of Ukraine in the form of acreage for this crop, the projected growth of which to 1-1.3 million hectares indicates its prospects. If we take into account adaptability, compared to corn, this crop is less demanding of moisture, which makes it suitable for regions with irrigation problems. In the face of climate change, this is an important advantage. No less significant is the search for areas of sorghum breeding and obtaining hybrids that will produce high yields in different climatic zones of Ukraine. Sorghum has a high nutritional value, which can contribute to its wider use in the food industry (gluten-free products, starch, alcohol). This promising crop is a valuable feed for livestock, pigs, and poultry. At the same time, it is important to take into account the nuances of digestibility and methods of pre-processing grain. The introduction of this crop into the feed wedge of farms for the production of livestock products will allow timely receiving a sufficient amount of sorghum grain and harvesting silage necessary for feeding cattle, especially lactating cows, which will contribute to obtaining high-quality dairy and meat products and preserving the health and productive longevity of livestock.

Conclusions. Thus, the spread of sorghum in the world indicates the great potential of this crop. Due to the origin and species diversity of sorghum, even in the driest and hottest regions of the world, unlike other agricultural crops, it allows you to get stable, high yields of grain and green mass. This makes it one of the leading grain and food crops. The use of sorghum silage is an effective solution in animal husbandry, especially in conditions of shortage of corn silage. However, for optimal effect, it is recommended to adjust the diet and carefully control the quality of feed. At the same time, the potential of this culture in Ukraine is still far from being revealed.

References

Abah, C., Ishiwu, C., Obiegbuna, J., & Oladejo, A. (2020). Sorghum Grains: Nutritional Composition, Functional Properties and Its Food Applications. *European Journal of Nutrition & Food Safety, 12* (5), 101–111. https://doi.org/10.9734/ejnfs/2020/v12i530232

Abdelhadi, L. O., & Santini, F. J. (2006). Corn silage versus grain sorghum silage as a supplement to growing steers grazing high quality pastures: Effects on

performance and ruminal fermentation. *Animal Feed Science and Technology*, *127* (2), 33–43. https://doi.org/10.1016/j.anifeedsci.2005.08.010

Aguerre, M., Cajarville, C. , Machado, V., Persak, G., Brambillasca, S., & Repetto, J. (2009). Dry matter intake and digestibility of temperate pastures supplemented with sorghum grain in wethers and heifers. *South African Journal Of Animal Science*, *39* (1), 252. https://doi.org/10.4314/sajas.v39i1.61158

Assefa, Y., Holman, J., Obour, A., O'Brien, D., & Prasad, P. (2024). Historic Grain Sorghum Production, Value, Yield Gap, and Weather Relation Trends. *Agronomy*, *14* (11), 1–15. https://doi.org/10.3390/agronomy14112582

Berenji, J., Dahlberg, J., Sikora, V., & Dragana, L. (2011). Origin, History, Morphology, Production, Improvement, and Utilization of Broomcorn [Sorghum bicolor (L.) Moench] in Serbia. *Economic Botany*, *65* (2), 190-208. https://doi.org/10.1007/s12231-011-9155-2

Beta, T., Corke, H., Rooney, L., & Taylor, J. (2000). Starch properties as affected by sorghum grain chemistry. *Journal of The Science of Food and Agriculture*, *81* (2), 245–251. https://doi.org/10.1002/1097-0010(20010115)81:2<245::AID-JSFA805>3.0.CO;2-S

Burgarella, C., Berger, A., Glémin, S., David, J., Terrier, N., Deu, M., & Pot, D. (2021). The Road to Sorghum Domestication: Evidence From Nucleotide Diversity and Gene. *Expression Patterns Front Plant Sci.*, *12*, 1–15. https://doi.org/10.3389/fpls.2021.666075

Cardoso, L. M., Silva Pinheiro, S., Duarte Martino, S.H., & Pinheiro-Sant'Ana, H. M. (2015). Sorghum (*Sorghum bicolor* L.): nutrients, bioactive compounds, and potential impact on human health. *Critical Reviews in Food Science and Nutritio*, *57* (2), 372–390. https://doi.org/10.1080/10408398.2014.887057

Cattani, M., Guzzo, N., Mantovani, R., & Bailoni, L. (2017). Effects of total replacement of corn silage with sorghum silage on milk yield, composition, and quality. *Journal of Animal Science and Biotechnology*, *8* (1). https://doi.org/10.1186/s40104-017-0146-8

Dremlyuk, H. K., Topal, I. A., Vlashchenkov, V. M. (2020). Soriz conditions of success (spring worries about harvest). Available at: https://olis.com.ua/en/press-centre-en/articles/article13/

Eggleston, G., Triplett, A., Bettarber, K., Boue, S., & Bechtel, P. (2022). Macronutrient and mineral contents in sweet sorghum syrups compared to other commercial syrup sweeteners. *Journal of Agriculture and Food Research*, *7*, 1–11. https://doi.org/10.1016/j.jafr.2022.100276

Ge, F., Xie, P., Wu, Y., & Xie, Q. (2022). Genetic architecture and molecular regulation of sorghum domestication. *aBIOTECH*, *4* (1), 57–71. https://doi.org/10.1007/s42994-022-00089-y

Getachew, G., Putnam, D., Ben, C., & Peters, E. (2016). Potential of Sorghum as an Alternative to Corn Forage. *American Journal of Plant Sciences*, *7* (7), 1106–1121. https://doi.org/10.4236/ajps.2016.77106

Gichile, H. (2022). Review On Breeding Sorghum (Sorghum Bicolor (L.) Moench) for Nutritional Quality Improvement *International Journal of Research Studies in Agricultural Sciences (IJRSAS)*, *8* (2), 13–19, https://doi.org/10.20431/2454-6224.0802002

Gómez, M. (2022). Gluten-free bakery products: Ingredients and processes. *Advances in Food and Nutrition Research*, *99*, 189–238. https://doi.org/10.1016/BS.AFNR.2021.11.005

Guo, F., Wang, S., Dong, M., Sun, X., Xu, F., Chen, J., Song, T., & He, B. (2024). Effects of replacing whole-plant corn silage with sweet sorghum silage in diets on the slaughter performance and meat quality of beef cattle. *Journal of Applied Animal Research, 52* (1), 1–9. https://doi.org/10.1080/09712119.2024.2425174

Herniwati, H., Pabendon, M., Wicaksono, K., Waluyo, B., & Widaryanto, E. (2024). Estimation of genetic diversity of sweet sorghum (Sorghum bicolor (L.) Moench) genotypes as a bioethanol source using SSRs markers. *Czech J. Genet. Plant Breed, 60* (2*),* 86–96. https://doi.org/10.17221/79/2023-CJGPB

Hossain, S., Nahidul Islam, Md., Mamunur Rahman, Md., Golam Mostofa, Md. & Arifur Rahman Khan, Md. (2022). Sorghum: A prospective crop for climatic vulnerability, food and nutritional security. *Journal of Agriculture and Food Research, 8*, 1–9. https://doi.org/10.1016/j.jafr.2022.100300

Jabbari, H., Tabatabaei, S., Kordnejad, E., Modarresi, M., & Tabeidian, S. A. (2011). Effect of dietary corn silage replacement with sorghum silage on performance and feed cost of growing steers. *Online Journal of Animal and Feed Research, 1* (1), 14–21. Available at: https://www.ojafr.ir/main/attachments/article/54/OJAFR,%20A03.pdf

Jiao, J., Wang, T., Li, S., Gou, N., Degen, A., Long, R., Wang, H., & Shang, Z. (2022). Effects of supplementing sweet sorghum with grapeseeds on carcass parameters, and meat quality, amino acid, and fatty acid composition of lambs. *Animal Bioscience, 36* (3), 461–470. https://doi.org/10.5713/ab.22.0189

Khoddami, A., Messina, V., Venkata, K., Farahnaky, A., Blanchard, C., & Roberts, T. (2023). Sorghum in foods: Functionality and potential in innovative products. *Critical Reviews in Food Science and Nutrition, 63* (9), 1170–1186. https://doi.org/10.1080/10408398.2021.1960793

Kondratiuk, S. (2018). *Sorho – kultura maibutnoho* [Sorghum - the crop of the future]. Ahronom. Available at: https://www.agronom.com.ua/sorgo-kultura-budushhego/ (in Ukrainian)

Lee, S., Fu, F., Liao, C., Bayable, D., Adeyanju, A., Ejeta, G., Lisch, D., & Broad, T. (2022). Spectrum fungal resistance in sorghum is conferred through the complex regulation of an immune receptor gene embedded in a natural antisense transcript. *The Plant Cell*, 34 (5), 1641–1665. https://doi.org/10.1093/plcell/koab305

Leite ,P., Botelho, T., Ribeiro, P., Schaffert, R., Parrella, R., & Rodrigues, J. (2020). Intrapopulation recurrent selection in sweet sorghum for improving sugar yield. *Industrial Crops and Products, 8*, 1–16. https://doi.org/10.1016/j.indcrop.2019.111910

Li, S., Zhang, J., Bai ,Y., Degen, A., W, T., Shang, Z., Ding, L., & Long, R. (2020). Sorghum silage substituted for corn silage in diets for dairy cows: Effects on feed intake, milk yield and quality, and serum metabolites. *Applied Animal Science, 36* (2), 228–236. https://doi.org/10.15232/aas.2019-01923

Lv, X., Chen, L., Zhou ,C., Zhang, G., Xie, J., Kang, J., Tan, Z., Tang, S., Kong, Z., Liu, Z., & Du, Z. (2023). Application of different proportions of sweet sorghum silage as a substitute for corn silage in dairy cows. *Food Science & Nutrition, 11*, 3575–3587. https://doi.org/10.1002/fsn3.3347

McGinnis, M. J., & Painter, J. E. (2020). Sorghum: History, Use, and Health Benefits. *Nutrition Today*, 55 (1), 8–44. https://doi.org/10.1097/NT.0000000000000391

Ochieng, B., Owino, W., Kinyuru, J., Mburu, J., &. Gicheha, M. (2020). Effect of low tannin sorghum based feeds on broiler meat nutritional quality. *Journal of Agriculture and Food Research, 2*, 1–7. https://doi.org/10.1016/j.jafr.2020.100078

Ozturk, I. (2018). *Solo dlia sorho, abo chomu ahrovyrobnyky obyraiut tsiu kulturu* [Solo for sorghum, or why farmers choose this crop]. Agravery. Available at: https://agravery.com/uk/posts/author/show?slug=solo-dla-sorgo-abo-comu-agrovirobniki-obiraut-cu-kulturu (in Ukrainian)

Pimentel, J., Lana, R., Oliveira, A., Teixeira, R., & Abreu, D. (2013). Dairy cows feeding with sorghum silage supplemented with concentrate. *Pesquisa Agropecuária Tropical, 43* (3), 255–261. https://doi.org/10.1590/S1983-40632013000300013

Pravdyva, L. A., Hanzhenko, O. M., & Honcharuk, H. S. (2023). Productivity of sorghum [Sorghum bicolor (L.) Moench] and soryz (S. orysoidum) depending on methods of weed control. *Plant Varieties Studying and Protection, 19* (3), 176–184. https://doi.org/10.21498/2518-1017.19.3.2023.287641

Pujiharti, Y., Paturohman, E., & Ikhwani. (2022). Prospect of sorghum development as corn substitution in Indonesia. *IOP Conf. Series: Earth and Environmental Science, 978*, 1–6, 012019. https://doi.org/10.1088/1755-1315/978/1/012019

Rakshit, S., Hariprasanna, K., Gomashe, S. S., Ganapathy, K .N., Das, I. K., Ramana, O. V., Dhandapani, A., & Patil, J. V. (2014). Changes in area, yield gains, and yield stability of sorghum in major sorghum-producing countries, 1970 to 2009. *Crop Science, 54* (4), 1570–1584. https://doi.org/10.2135/cropsci2012.12.0697

Risyahadi, S., Martin, R., Qomariyah, N., Suryahadi, S., Sukria, H., & Jayanegara, A. (2023). Effects of dietary extrusion on rumen fermentation, nutrient digestibility, performance and milk composition of dairy cattle: a meta-analysis. *Animal Bioscience, 36* (10), 1546–1557. https://doi.org/10.5713/ab.23.0012

Ronda, V., Aruna, C., Visarada, K., B., & Bhat, V. (2019). Chapter 14 – Sorghum for Animal Feed. *Breeding Sorghum for Diverse End Uses*, 229–238. https://doi.org/10.1016/B978-0-08-101879-8.00014-0

Smith, C. W. & Frederiksen, R. A. (2000). *Sorghum Origin, History, Technology and Production.* New York, NY: John Wiley & Sons. Available at: https://www.wiley.com/en-us/Sorghum%3A+Origin%2C+History%2C+Technology%2C+and+Production-p-9780471242376

Sotak, K. M., Houser, T. A., Goodband, R. D., Tokach, M. D., Dritz, S. S., DeRouchey, J. M., Goehring, B. L., Skaar, G. R., & Nelssen, J. L. (2015). The effects of feeding sorghum dried distillers grains with solubles on finishing pig growth performance, carcass characteristics, and fat quality. *Journal of Animal Science, 93* (6), 2904–2915. https://doi.org/10.2527/jas.2014-8022

Suszkiw, J. (2023). Strengthening Sorghum Against a Worldwide Fungal Threat. *Agricultural Research Service U.S. DEPARTMENT OF AGRICULTURE*. Available at: https://www.ars.usda.gov/news-events/news/research-news/2023/strengthening-sorghum-against-a-worldwide-fungal-threat/

Szabłowska, E., & Tańska, M. (2021). Acorn flour properties depending on the production method and laboratory baking test results: A review. *Comprehensive Reviews in Food Science and Food Safety, 20* (1), 980–1008. https://doi.org/10.1111/1541-4337.12683

Thomas, L. L., Espinosa, C. D., Goodband, R. D. , Stein, H. H., Tokach, M. D., Dritz, S. S., Woodworth, J. C., & DeRouchey, J. M. (2020). Nutritional evaluation of different varieties of sorghum and the effects on nursery pig growth performance. *Journal of Animal Science, 98* (5), skaa120. https://doi.org/10.1093/jas/skaa120

Treviño-Salinas, M., Perales-Torres, A., Castillo-Ruíz, O., Montes-García, N., Lizarazo-Ortega, C., Navarro-Cortez, R., & RodríguezCastillejos, G. (2021). Proximal analysis and profile of fatty acids on six varieties of white grain sorghum with

potential use in human consumption. *CyTA - Journal of Food*, *19* (1), 547–551. https://doi.org/10.1080/19476337.2021.1928757

Voitovska, V. I., Storozhyk, L. I., Liubych, V. V., & Yalanskyi, O. V. (2022). Evaluation of productivity of different varieties of soryz (Sorghum orysoidum). *Plant Varieties Studying and Protection*, *18* (1), 50–56. https://doi.org/10.21498/2518-1017.18.1.2022.257587

Widodo, S., Purwaningsih, H., Budi Pustika, A., Widyayanti, S., Muazam, A., Putri Hanifa, A., Triastono, J., Sahara, D., Sulistyawati Purwaning Rahayu, H., Laksono, P., Fahmi, D. A., Pramono, J. & Rachmiwati, Y. (2024). Sorghum Productivity and Its Farming Feasibility in Dryland Agriculture: Genotypic and Planting Distance Insights. *Phyton-International Journal of Experimental Botany*, *93* (5), 1–15. https://doi.org/10.32604/phyton.2024.048770

Wondimu, Z., Dong, H., Paterson, A., Worku, W., & Bantte, K. (2021). Genetic diversity, population structure, and selection signature in Ethiopian sorghum [Sorghum bicolor L. (Moench)] germplasm. *G3 Genes|Genomes|Genetics*, *11* (6), 1–10. https://doi.org/10.1093/g3journal/jkab087

Wu, Y., Chang, Y., Kuo, S., Liao, D., Shen, T., Kuo, H., Wang, S., & Tseng, Y. (2023). The Breeding of Waxy Sorghum Using Traditional Three-Line Method and Marker-Assisted Selection. *Agriculture*, *13* (11), 1–14. https://doi.org/10.3390/agriculture13112054

Yahaghi, M., Liang, J., Balcells, J., Valizadeh, R., Jahromi, M., Alimon, R., & Ho, Y. (2014). Extrusion of sorghum starch enhances ruminal and intestinal digestibility, rumen microbial yield and growth in lambs fed on high-concentrate diets. *Animal Feed Science and Technology*, *189*, 30–40. https://doi.org/10.1016/j.anifeedsci.2013.12.009

Zhang, S., Wang, J., Lu, S., Shakoor Chaudhry, A., Tarla, D., Khanaki, H., Raja, I., & Shan, A. (2024). Effects of Sweet and Forge Sorghum Silages Compared to Maize Silage without Additional Grain Supplement on Lactation Performance and Digestibility of Lactating Dairy Cows. *Animals*, *14* (11), 1702. https://doi.org/10.3390/ani14111702

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INFLUENCE OF MORPHOFUNCTIONAL PARAMETERS OF MARES OF NOVOOLEXANDRIVSKII DRAFT ON THEIR MILK PRODUCTIVITY

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The research is devoted to determining the relationship of milk productivity of mares of the Novoolexandrivskii Draft with their morph functional indicators: torso and udder measurements. in two independent experiments (in two different farms), the indicator of milk productivity of mares of the Novoolexandrivskii Draft was studied depending on morph functional indicators – torso and udder measurements. the highest level of milk productivity was established in large-type mares by height at the Withers (150 cm) and chest circumference (190 cm). At the same time, minor correlations were established between the indicator of milk productivity and height at the withers (r=0.112) and oblique trunk length (r=0.109). In the second experiment, milk productivity was most correlated with chest circumference (r=0.280), metacarpal circumference (r=0.245), and trunk circumference (r=0.232). Body measurements of the studied mares are quite closely related: height at the withers × circumference of the body (r=0.811), circumference of the body × circumference of the metacarpus (r=0.573), chest circumference × circumference of the metacarpus (r=0.559), height at the withers × circumference of the metacarpus (r=0.520). By determining the development indicators of foals from Mares of various types, it was established that both foals and mares obtained from large-type mares prevailed over peers obtained from small-type mares by live weight in the development periods from birth to 18 months of age.

It was found that large-type mares are also characterized by higher indicators of udder girth and length, while small-type mares predominated in udder depth. Positive correlation coefficients of the average bond strength were found between the milk productivity of mares and udder circumference (r=0.370) and udder length (r=0.301), with udder depth the bond is weak and negative (r=-0.113). A fairly strong relationship was found between udder measurements: girth × length (r=0.665), length × depth (r=0.570), girth × depth (r=0.361). The udder girth index significantly and positively correlated with the indicators of body structure indices: format (r=0.654), massiveness (r=0.514), Bony (r=0.391). The udder length index is positively and significantly correlated with the bony index (r=0.486) and format index (r=0.323).

Keywords: horses (Equus caballus), Novoolexandrivskii Draft, milk productivity, body measurements, udder parameters.

ВПЛИВ МОРФОФУНКЦІОНАЛЬНИХ ПАРАМЕТРІВ КОБИЛ НОВООЛЕКСАНДРІВСЬКОЇ ВАГОВОЗНОЇ ПОРОДИ НА ЇХ МОЛОЧНУ ПРОДУКТИВНІСТЬ

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Дослідження присвячені визначенню взаємозв'язків молочної продуктивності кобил новоолександрівської ваговозної породи з їх морфофункціональними показниками: промірами тулуба і вим'я. У двох незалежних дослідах (у двох різних господарствах) вивчали показник молочної продуктивності кобил новоолександрівської ваговозної породи залежно від морфофункціональних показників – промірів тулуба і вим'я. Встановлено вищий рівень молочної продуктивності у кобил крупного типу за висотою в холці (<150 см) та обхватом грудей (<190 см). При цьому встановлено незначні кореляційні зв'язки між показником молочної продуктивності та висотою в холці (r=0,112) та косою довжиною тулуба (r=0,109). У другому досліді показник молочної продуктивності найбільше корелював з обхватом грудей (r=0,280), обхватом п'ястка (r=0,245) та навкісною довжиною тулуба (r=0,232). Проміри тіла досліджених кобил досить тісно пов'язані між собою: висота в холці × навкісна довжина тулуба (r=0,811), навкісна довжина тулуба × обхват п'ястка (r=0,573), обхват грудей × обхват п'ястка (r=0,559), висота в холці × обхват п'ястка (r=0,520). Визначенням показників розвитку лошат від кобил різних типів встановлено, що і жеребчики і кобилки, одержані від кобил крупного типу, переважали ровесників, одержаних від кобил дрібного типу за живою масою по періодах розвитку від народження до 18-місячного віку.

Встановлено, що кобили крупного типу характеризуються також вищими показниками обхвату і довжини вим'я, за глибиною вим'я переважали кобили дрібного типу. Виявлені позитивні коефіцієнти кореляції середньої сили зв'язку між молочною продуктивністю кобил та обхватом вим'я (r=0,370) і довжиною вим'я (r=0,301), з глибиною вим'я зв'язок слабкий і негативний (r=-0,113). Між промірами вим'я виявлено досить міцний зв'язок: обхват × довжина (r=0,665), довжина × глибина (r=0,570), обхват × глибина (r=0,361). Показник обхвату вим'я значно і позитивно корелював із показниками індексів будови тіла: формату (r=0,654), масивності (r=0,514), костистості (r=0,391). Показник довжини вим'я позитивно і значно корелює з індексами костистості (r=0,486) і формату (r=0,323).

Ключові слова: коні (Equus caballus), новоолександрівська ваговозна порода, молочна продуктивність, проміри тіла, параметри вим'я.

Introduction. Horses have long not played the role of the main labor force in agricultural production, although cattle (DAP) is recognized as one of the 14 renewable energy sources selected by the UN Conference on new and renewable energy, held in Nairobi in 1981 (Spugnoli P. & Dainelli R., 2012). According to the food and Agriculture Organization of the United Nations FAO (Elcio P. et all., 2007; Miraglia N., 2015), about 300 million people are used in the world. working animals (horses, donkeys, mules, cows, camels) that provide life support for 300-600 million people., especially in poor regions where working animal energy is an important energy resource (Asmare B. & Yayeh Z., 2017; Burn C.C. et all., 2010; Miraglia N. et all, 2020; Romaniuk K. et all, 2019).

Horses of heavy breeds originate from large fighting horses of the Middle Ages, whose task was to carry soldiers in metal armor, while they themselves had metal protection from Spears and arrows of opponents (https://www.horsejournals). In the future, heavy horses were used to move cannons and other combat weapons and cargo, as well as used in agricultural work (Stephens T.D. & Splan R.K., 2013). As the agricultural implements became heavier and the amount of work increased at the same time, the horses needed to get bigger and stronger. Heavy horse breeding reached a special heyday in the late XIX – early XX centuries with the development of industry and transport. Horses were used for logging, mining, railway construction, roads, etc., as well as for transporting metro cars and horse-drawn trams. In the first World War, heavy horses were indispensable tools for transporting artillery and ammunition. Historically, the use of horses as productive animals has been proven since their domestication by primitive humans (Langlois B., 2011).

With the development of mechanical engineering, the need for heavy horses almost disappeared and because of this, many breeds of horses with a unique genotype (Hudson R.S. & Cole C.L., 1939). However, in many countries, local breeds of heavy trucks are part of the traditions and are preserved at the level of national heritage. Thus, the following breeds are preserved horses: Arden, Belgian, Clydesdale, Shire, Percheron, Breton, Suffolk, etc. A good example is the Clydesdale Outpost program, which aims to preserve the Clydesdale horse breed, its unique characteristics and genetic originality (https://www.clydesdaleoutpost). Draft horses are used in the production of organic agricultural products, for recreational purposes, equestrian tourism, logging and other works where the use of equipment is impossible or impractical. Some ethnoreligious communities (Mennonites, Amish, etc.) abandon modern intensive technologies and use horse-drawn harnesses in everyday life. Original breeds of heavy horses are used in cultural and traditional events in many countries (https://www.horsejournals).

Noteworthy are the productive breeds of Asian horses, which are often direct descendants of relict horses tamed by nomadic tribes, and are characterized by high adaptability to the harsh conditions of year-round herd maintenance. These are Kazakh horses of the Jabe type, the Aday branch of Kazakh horses, the Kabinsky meat type of the Kazakh horse, the Kushum and mugalzhar breeds (Iskhan K. Zh. et all., 2019; Pabat V.O. & Goncharenko I.V., 2019; Sansyzbaev et all., 2024; Turabaev, 2015).

Original breeds of heavy horses are used in cultural and traditional events in many countries: various shows, folk festivals, competitions, cargo transportation championships, tourism, historical reconstructions (Sawers L., 2003; Rzekęć A. et all., 2020). In addition, heavy breeds have become the genetic basis of many modern sports horse breeds (Asmare B. & Yayeh Z., 2017; Edmonds J. L., 1940; Garre A., 2022).

Ukraine is the originator country of a unique heavy-duty breed – Novoolexandrivskii Draft, created by the efforts of domestic scientists and breeders. The best heavy breeds of Europe appeared in Ukrainian farms in the 1860s and 1880s with the development of Agriculture and industry. On the genetic basis of Belgian Ardennes

and Brabansons, French Percherons, Scottish Clydesdales and local horses well adapted to the climatic conditions of Ukraine, for more than a century, the new Alexander heavy breed (recognized in 1998) (Liutykh S.V., 2002; Program, 2014).

The growing popularity of «green» or organic food production gives a new impetus to the restoration of the popularity of Draft horses (Aguilera E. et all., 2019; Rzekęć A. et all., 2020). The use of Novoolexandrivskii Draft is also not limited to transport and agricultural use, the breed is successfully used for the production of milk and koumiss. Mare's milk has a high nutritional value and practically does not contain allergenic proteins, and therefore can be a raw material for the production of children's and dietary food products, medicines and cosmetology products (Businesso L. et all., 2000; Centoducati P., 2012; Jastrzębska E. et all., 2017; Pieszka M. et all., 2016; Ranadheera C. S. et all., 2018; Romaniuk K. et all., 2019; Yakunin A. V. et all., 2017). It is worth noting that in developed economies, the production of mare's milk on organic farms is one of the most promising areas of animal husbandry. This production is highly profitable, does not require significant expenditures of funds, energy and human resources, and is safe for the environment, because it involves organic, natural animal husbandry. The high identity of the chemical composition of mare's milk to female milk gives grounds for its widespread use in baby food, both complementary feeding of infants on artificial nutrition and the production of dairy products with prebiotic qualities for children of all age groups (Pieszka M. et all., 2016; Romaniuk K. et all., 2019; Yakunin A. V. et all., 2017). In Finland, Germany, and Kazakhstan, mare's milk has been widely studied in pediatrics, and technologies for its processing and long-term storage have been developed. So, in Germany (TM «Saumalmilk», TM «Zollmann», GmbH & Co Kazakhstan (Kazakh Academy of nutrition) uses sublimation technology, which makes it possible to obtain powdered milk of mares, which corresponds to 99% of its fresh counterpart. The best example of profitable productive horse breeding can be considered the farm «Kurgestüt Hoher Odenwald» in Germany, where 400 mares (such as a small heavy truck) are kept on 450 hectares of land, there is a deep freezing shop and a milk sublimation shop for the production of koumiss, other bio-products and cosmetics (https://www.demeter-bw). The technology of obtaining powdered milk allows you to sell bio-koumiss from this farm under its own trademark «Zollmann» throughout Europe. The high profit of this production is evidenced by the fact that the cost of 200 ml of bio-kumiss is 4.90 euros (https://www.stutenmilch).

In Ukraine, an interesting example of popularizing productive horse breeding is the company «Dendoff Agro Family», which in the Tetiivskii District of the Kiev region founded the trademark MLK PWR ("Milk Power") for the production of kumiss and other products from the milk of mares and cows (Balagura B.; 2019).

So, the study of milk productivity of mares is an urgent task in the future development of the industry and productive horse breeding in particular. Like any quantitative trait, the milk productivity of mares is formed under the influence of various factors, determining the strength of which is the task of researchers to predict and improve productive traits. It is known that the milk productivity of livestock is closely related to the size of the body and udder (Polupan Yu., 2024). There are few similar data on horses in our country (Yusyuk T. A., 2017; Yusyuk T. A., Gopka B. M., 2018).

The aim of the work was to determine the milk productivity of mares of the Novoolexandrivskii Draft Horse in Ukraine, and the impact of body and udder measurements on it.

Materials and methods. On Mares of the Novoolexandrivskii Draft of Stud «Lann» of Donetsk region (n=32) and the Dibrivskii Stud of Poltava region (n=16), experiments were conducted to study the indicators of milk productivity of mares

depending on their morph functional parameters (body and udder measurements). For the experiment, healthy mares were selected that safely gave birth to healthy foals. Control milking operations were carried out using DDU-2 portable milking machines during the milking season of mares for kumiss production (May-September). Mares were milked three times during the day with an interval of 2 hours, the duration of milking is up to 2 minutes. During milking, the foals were in the milking parlor in a separate close visibility of the mares, which contributed to the milk return reflex and the nervous balance of the mares and foals. Foals on the day of control milking received the required amount of milk through artificial drinking, which did not affect their condition. After each milking, the amount of milk in the bucket was measured, and the total daily milk yield was determined as the sum of all milks per day. Milk productivity was determined based on the results of control milks for the entire milking season. The relationship of milk productivity of mares with body measurements and udder parameters was established. Body measurements were determined – height at the withers and circumferential length of the body – with a measuring stick, chest circumference and metacarpal circumference – with a measuring tape. Based on the obtained indicators, the indices of body structure were calculated: format and massiveness. Udder parameters (girth, length, depth) were measured with a measuring tape. The relationships between the studied indicators were determined by calculating the correlation coefficient (r).

Indicators of the development of foals obtained from large and small mares by live birth weight and at 1, 3, 6, 9 and 18 months of age were also determined.

All experimental studies were conducted in accordance with modern methodological approaches, requirements and standards (DSTU ISO/IEC 17025:2019, 2021), directive 2010/63/EC (2010), the procedure for conducting animal testing in research institutions (Law of Ukraine No. 249, 2012) and in accordance with the provisions of the European Convention for the protection of vertebrates used for experimental and other scientific purposes (Strasbourg, 1985).

Research results. The first experiment to study the indicator of milk productivity of mares of the Novoolexandrivskii Draft depending on morph functional indicators was conducted in Stud «Lann» of the Donetsk region (n=32). Control milking was performed during the main lactation season in May-September. According to measurements, mares were divided into gradations according to height at the withers: Group I – small (150 cm), Group II – large (150 cm) and chest circumference – group I – small (190 cm), Group II – large (190 cm).

Indicators of milk productivity of mares of various types: small (group I) and large (group II) are shown in Table 1.

A higher level of milk productivity was established in mares of large type and with a higher live weight. At the same time, the analysis of correlation coefficients did not establish a significant relationship between the studied indicators. The highest relationship was found between milk productivity and height (r=0.112) and oblique trunk length (r=0.109). Higher ligaments were established between body measurements: height × circumference of the trunk (r=0.811), circumference of the trunk × circumference of the metacarpus (r=0.573), chest circumference × circumference of the metacarpus (r=0.559), height × circumference of the metacarpus (r=0.520).

Table 1

Indicators of measurements, live weight and milk productivity of experimental mares of the Novoolexandrivskii Druft Horse

Groups of mares by body measurement type		Body measurements, cm				Live weight, kg	Milk productivity, l
		height	body length	chest circumference	metacarpal girth		
I (n=20)	M±m	146,9 ±0,51	156,5 ±0,93	188,8 ±1,77	21,17 ±0,19	660,9 ±6,20	684,0 ±66,8
	Cv, %	1,21	2,05	3,25	3,08	3,25	33,8
II (n=12)	M±m	154,4 ±0,51	162,8 ±0,70	192,1 ±1,84	21,70 ±0,13	673,9 ±6,29	761,0 ±72,7
	Cv, %	1,46	1,93	4,28	2,63	4,17	42,7
Together (n=32)	M±m	151,9 ±0,75	160,4 ±0,78	190,9 ±1,34	21,5 ±0,11	669,1 ±4,64	732,1 ±51,6
	Cv, %	2,78	2,74	3,97	3,01	3,93	39,9

Indicators of the development of foals obtained from mares of various types are established (table 2). Both stallions and mares obtained from large-type mares outnumbered peers obtained from small-type mares in live weight at all developmental periods.

Table 2

Dynamics of development of foals obtained from mares of various types

Age, month	Type of mares							
	Group I (Small)				Group II (large)			
	Stallions		Fillies		Stallions		Fillies	
	M±m	Cv, %	M±m	Cv, %	M±m	Cv, %	M±m	Cv, %
3 days	64,2 ±0,38	1,46	63,5 ±0,32	2,63	66,5*** ±0,58	2,48	64,7** ±0,61	2,14
1	79,4 ±0,46	2,54	77,8 ±0,63	2,56	88,6** ±1,51	2,98	79,4*** ±1,28	2,22
3	139,6 ±0,98	2,57	128,9 ±1,01	2,68	143,1** ±2,25	3,24	138,6*** ±2,08	3,11
6	246,7 ±3,18	2,89	229,2 ±3,10	2,74	256,4** ±3,18	5,36	338,4** ±3,78	6,28
9	331,2 ±3,58	3,11	318,5 ±3,17	3,16	356,4** ±4,12	7,11	324,9** ±4,01	6,57
18	578,4 ±5,48	6,37	556,9 ±4,81	5,21	595,9* ±6,42	9,24	529,3* ±5,67	8,21

*Note: * - p≤ 0.05; * * - p≤0.01; * * * - p≤0.001*

In order to verify the obtained data, a study was conducted on Mares of the Dibrivskii Stud (n=16). In addition to indicators of milk productivity and body measurements, udder measurements (centimeters) are determined: girth, length, depth.

Indicators of milk productivity of mares of various types: small (group I) and large (group II) are shown in Table 3.

Table 3

Indicators of body measurements and body structure indices of mares of the Novoolexandrivskii Draft Horse

Groups of mares by body measurement type		Body measurements, cm				Body structure indices, %			Milk productivity, l
		height	body length	chest circumference	metacarpal girth	format	massiveness	bone dirth	
I (n=7)	M±m	148,1 ±0,34	156,7 ±0,78	191,9 ±2,41	21,6 ±0,34	105,8 ±0,68	129,5 ±1,59	14,6 ±0,21	2761,0 ±280,0
	Cv, %	0,61	1,31	3,32	4,16	1,69	3,27	3,76	26,83
II (n=9)	M±m	152,2 ±0,94	159,7 ±1,55	193,4 ±2,53	22,1 ±0,29	104,9 ±0,72	130,4 ±3,73	14,5 ±0,17	2872,7 ±191,26
	Cv, %	1,85	2,90	3,92	3,88	2,05	8,59	3,44	19,97
Together (n=16)	M±m	150,4 ±0,75	158,4 ±0,98	192,8 ±1,72	21,91 ±0,22	105,3 ±0,49	130,0 ±2,15	14,6 ±0,13	2761,3 ±161,77
	Cv, %	1,99	2,48	3,58	4,02	1,89	6,63	3,47	23,43

The milk productivity of the studied mares for 150 days of lactation averaged 2448.81±163.6 kg of milk with limits lim=1603-3792 kg. The height of the studied mares averaged 150.4±0.75 cm, which means that horses of the Novoolexandrivskii Draft Horse belong to small heavy horses close to the Ardennes. The circumferential length of the body – 158.4±0.98 cm – significantly exceeds the height at the Withers, which indicates a distinct harness type. Average chest circumference (192.8±1.72 cm) and metacarpal circumference (21.9±0.22 cm) indicate the desired massiveness and bony nature of the studied mares. Udder parameters averaged: girth-65.6±1.25 cm, length – 19.7±0.44 cm, depth – 18.4±0.33 cm.

It was found that mares whose height exceeded 150 cm (large type) were characterized by significantly higher milk yield during lactation than smaller mares (by 111.7 liters, $p<0.01$), which confirms the data obtained in the first experiment. It was found that the indicator of milk productivity of mares of the Novoolexandrivskii Draft Horse with different strength and value is reliably ($P<0.05$) associated with body measurements: height (r=0.511), body circumference (r=-0.033), chest circumference (r=0.200), metacarpal circumference (r=0.130) (table 4).

Table 4

Correlations between milk production, body measurements, and udder measurements

Indicators	milk productivity	udder girth	udder length	udder depth	height at the Withers	trunk circumference	Chest circumference	Metacarpal circumference	format Index	massiveness Index
Udder circumference	0,370									
Udder length	0,301	0,665								
Udder depth	-0,113	0,361	0,570							
Height at the withers	0,132	-0,175	-0,126	0,140						
Body circumference	0,232	0,352	0,145	0,232	0,661					
Chest circumference	0,280	0,228	0,310	0,224	0,465	0,761				
Metacarpal circumference	0,245	0,263	0,371	0,557	0,486	0,628	0,688			
Format index	0,175	0,654	0,323	0,157	-0,190	0,612	0,503	0,311		
Massiveness index	0,096	0,514	0,245	0,449	-0,006	0,420	0,434	0,510	0,556	
Bone dirth index	0,194	0,391	0,486	0,545	-0,010	0,343	0,515	0,868	0,461	0,570

It was found that large-type mares are also characterized by higher indicators of udder girth and length, while small-type mares predominated in udder depth (table 5).

It is proved that the milk productivity of the studied Mares of the Novoolexandrivskii Draft Horse is related to the measurements of their udder. positive correlation coefficients of the average strength of the relationship between the milk productivity of mares and the udder circumference (r=0.370) and the udder length (r=0.301) were revealed, with the udder depth the relationship is weak and negative (r=-0.113). A fairly strong relationship was found between udder measurements: girth × length (r=0.665), length × depth (r=0.570), girth × depth (r=0.361).

The udder girth index significantly and positively correlates with the indicators of body structure indices: format (r=0.654), massiveness (r=0.514), Bony (r=0.391). The udder length index is positively and significantly correlated with the bony index (r=0.486) and format index (r=0.323).

The milk productivity of mares of different breeds is poorly understood, however, this indicator is important in terms of the development of the market for organic food, medicines and cosmetics.

Table 5

Indicators of udder measurements and milk productivity of mares of the novoaleksandrovskaya heavy-duty breed

Groups of mares by type		Udder measurement, cm			Milk productivity, L
		girth	length	depth	
I (n=7)	M±m	65,22±1,71	19,44±0,41	18,56±0,52	2761,0±280,0
	Cv, %	7,85	6,36	8,46	26,83
II (n=9)	M±m	66,14±1,98	20,00±0,87	18,14±0,39	2872,7±191,26
	Cv, %	7,93	11,55	5,67	19,97
Together (n=16)	M±m	65,63±1,26	19,69±0,44	18,38±0,33	2761,3±161,77
	Cv, %	26,65	8,84	7,27	23,43

Discussion. According to the results of our research, the influence of morph functional parameters of mares of the Novoolexandrivskii Draft Horse on their milk productivity is proved.

Studies of the level of milk productivity of mares of large type (height 150 cm and above and chest circumference 190 cm and above) and small type (height at the withers less than 150 cm and chest circumference less than 190 cm) have established an advantage in terms of milk yield for lactation of large mares (by 111.7 l, $p<0.01$). At the same time, low positive correlations were established between the indicator of milk productivity and height at the withers (r=0.112) and oblique trunk length (r=0.109). Body measurements of dairy mares significantly correlate with each other: height at the withers × circumference of the body (r=0.811), circumference of the body × circumference of the metacarpus (r=0.573), chest circumference × circumference of the metacarpus (r=0.559), height at the withers × circumference of the metacarpus (r=0.520).

It should be noted that foals of both sexes obtained from large mares were also larger from birth to 18 months of age. The obtained data coincide with the research of T. A. Yusyuk (Yusyuk T. A., 2017) relatively high correlation rates between foal size and their mothers ' milk productivity (r=0.79, $p<0.05$).

Positive correlation coefficients of the average bond strength were found between the milk productivity of mares and udder circumference (r=0.370) and udder length (r=0.301), with udder depth the bond is weak and negative (r=-0.113). A fairly strong relationship was found between udder measurements: girth × length (r=0.665), length × depth (r=0.570), girth × depth (r=0.361). The udder girth index significantly and positively correlates with the indicators of body structure indices: format (r=0.654), massiveness (r=0.514), Bony (r=0.391). The udder length index is positively and significantly correlated with the bony index (r=0.486) and format index (r=0.323).

In most European countries, environmental issues are not yet considered important enough for stakeholders environmental issues are not yet considered important enough for horse breeding stakeholders. However, thanks to their "green" assets, horses can play an active role in the environmental transition and debate, both independently and as a supplement both independently and as an addition to other economic Productions and services (Rzekęć A. et all., 2020), which is the Economic key to preserving horses of local

populations. In this context, one of the most promising areas of horse use is the production of hypoallergenic mare's milk and processed products.

Conclusions. A higher level of milk productivity was established in mares of the novoaleksandrovskaya heavy-duty breed of large type (height at the withers 150 cm, chest circumference 190 cm) with a low level of correlation (r=0.112 with height at the withers, r=0.109 with the circumferential length of the body). Foals of both sexes obtained from large mares outnumbered peers obtained from small mares by live weight at all developmental periods (from birth to 18 months of age).

The influence of udder parameters on the indicator of milk productivity of mares is proved. Positive correlation coefficients of the average strength of the relationship between the milk productivity of mares and udder circumference (r=0.370) and udder length (r=0.301) were found.

Expanding the range of use of horses of heavy breeds as productive animals that produce hypoallergenic dietary milk is an economic guarantee for the preservation of horses of local populations.

References

Aguilera E., Guzmán G. I., González de Molina M., Soto D., Infante-Amate J. (2019). From animals to machines. The impact of mechanization on the carbon footprint of traction in Spanish agriculture: 1900–2014, *Journal of Cleaner Production*, Vol. 221, 295-305. doi:10.1016/j.jclepro.2019.02.247.

Asmare, B., Yayeh, Z. (2017). Assessment on the management of draft horses in selected areas of Awi Zone, Ethiopia. *Agric & Food Secur* 6, 69. doi:10.1186/s40066-017-0150-4

Balagura B. (2019). Ahrofirma z Kyyivshchyny vypuskatyme kumys pid vlasnym brendom – Kurkul [An agricultural company from the Kyiv region will produce koumiss under its own brand – Kurkul]. agri-gator.com.ua/2019/07/03/ahrofirma-z-kyivshchyny-vypuskatyme-kumys-pid-vlasnym-brendom-kurkul/

Burn, C. C., Dennison, T. L., & Whay, H. R. (2010). Relationships between behavior and health in working horses, donkeys, and mules in developing countries. *Applied Animal Behaviour Science*, *126*(3-4), 109-118.

Businco L., Giampietro P.G., Lucenti P., Lucaroni F., Pini C., Di Felice G., Iacovacci P., Curadi C., Orlandi M. (2000). Allergenicity of mare’s milk in children with cow’s milk allergy. *J Allergy Clin Immunol*, 105, 1031-1034.

Centoducati P., Maggiolino A., De Palo P., Tateo A. (2012). Application of Wood's model to lactation curve of Italian Draft horse mares. *J. Dairy Sci*.; 95: 5770-5. doi:10.3168/jds.2012-5513

Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific. (2010). Retrieved from https://www.fao.org/faolex/results/details/ru/c/LEXFAOC098296/

DSTU ISO/IEC 17025:2019 General requirements for the competence of testing and calibration laboratories. (2021). Retrieved from https://online.budstandart.com/ua/catalog/doc-page.html?id_doc=88724

Edmonds J. L. (1940). Draft Horse Type and the Breeder, *Journal of Animal Science***,** Vol. 1940, Iss. 1:87-89, doi:10.2527/jas1940.1940187x

Elcio P., Guimaraes E., Ruane J., Scherf B. D., Sonnino A., Dargie J. D. (2007). Marker-Assisted selection. *Current status and future perspectives in crops, livestock, forestry and fish*. FAO, Rome, 494 p.

Garre A. (2022). Farming with Draft Animals: Using Retro Innovations for Sustainable Agrarian Development. *A case study of organic small-scale farming in Northern*

Italy. Stockholm Resilience Centre Master's Thesis, 60 ECTS Social-ecological Resilience for Sustainable Development Master's programme 2020-2022, 120 ECTS Submitted May 20, 2022: 94

Hudson R. S., Cole C. L. (1939). Breeding and Development of Medium Weight Draft Horses, *Journal of Animal Science*, Vol. 1939, Iss. 1: 86-89, doi:10.1093/ansci/1939.1.86

Hurcombe S. D. A. (2014). Emergency Problems Unique to Draft Horses. *Equine Emergencies (Fourth Edition)*. https://www.sciencedirect.com/topics/pharmacology-toxicology-and-pharmaceutical-science/draft-horse

Iskhan, K. Zh., Akimbekov, A. R., Baimukanov, A. D., Aubakirov, Kh. A., Karynbayev, A. K., Rzabayev, T. S., Mukhatai Geminguli, Dzhunusova, R. Z., Apeev, K. B. (2019). Dairy productivity of the kazakh horse mares and their cross breeds with roadsters. *Bulletin of national academy of sciences of the republic of Kazakhstan. Vol. 3, N 379:22-35.* doi:10.32014/2019.2518-1467.65

Jastrzębska E., Wadas E, Daszkiewicz T., Pietrzak-Fiećko R. (2017). Nutritional Value and Health-Promoting Properties of Mare's Milk – a Review. Czech J Anim Sci.; 62(12): 511–518. doi:10.17221/61/2016-CJAS

Langlois, B. (2011). The history, ethnology and social importance of mare's milk consumption in Central Asia. *J. Life Sci*., 5, 863-872.

Law of Ukraine No. 249 «On the Procedure for Carrying out Experiments and Experiments on Animals by Scientific Institutions». (2012, March). Retrieved from https://zakon.rada.gov.ua/laws/show/z0416-12#Text

Liutykh S. V. (2002). Prospects for work with the Novoaleksandrovskaya heavy draft horse breed. *The Scientific and Technical Bulletin of Livestock farming institute of NAAS*, Kharkiv. 2002, № 82: 45-48

Mare's milk SPA Stud Hoher Odenwald Family Zollmann. Demeter: вебсайт. URL: https://www.demeter-bw.de/storage/1337

Miraglia, N. (2015). Sustainable development and equids in rural areas: An open challenge for the territory cohesion. *EAAP Scientific Series*; Vial, C., Evans, R., Eds.; Wageningen Academic Publishers: Wageningen, The Netherlands; Volume 136, pp. 167-176. ISBN 978-90-8686-279-5.

Miraglia, N., Salimei, E., & Fantuz, F. (2020). Equine Milk Production and Valorization of Marginal Areas: A Review. *Animals***,** *10*(2), 353. doi:10.3390/ani10020353

Pabat V.O., Goncharenko I.V. (2019). Technology of production and processing of mare's milk: practical manual. Kyiv: Publishing house Lira-K, 2019: 190 p.

Pieszka M., Łuszczyński J., Zamachowska M., Augustyn R., Długosz B., Hędrzak M. (2016). Is mare milk an appropriate food for people?–a review. Ann Anim Sci., 16(1): 33–51. doi:10.1515/aoas-2015-0041

Polupan Yu., Pryima S. (2024). Relative variability of heif growth and exterior of primary heifs with milk productivity of cows. *Bulletin of Agricultural Science*. 2024, № 5(854): 31-41. doi:10.31073/agrovisnyk202405-04

Program for breeding horses of the Novoaleksandrivskii draft breed until 2020 [Volkov D.A., Lyutikh S. V., Rossokha V.I., Tur G.M., Brovko O.V., of red. Tkachova I.V.], *Livestock farming institute of NAAS*, Kharkiv, 2014: 56 p.

Ranadheera C. S., Naumovski N., Ajlouni S. (2018). Non-bovine milk products as emerging probiotic carriers: recent developments and innovations. *Curr Opi. Food Sci*.; 22: 109–114. doi:10.1016/j.cofs.2018.02.010

Romaniuk, K.; Majszyk-S´wiatek, M.; Kryszak, K.; Danielewicz, A.; Andraszek, K. (2019). Alternative use of mare milk. Folia Pomer. Univ. Technol. Stetin., 348, 121-130. doi:10.21005/AAPZ2019.49.1.13

Rzekęć A., Vial C., Bigot G. (2020). Green assets of equines in the european context of the ecological transition of agriculture. *Animals,* 10 (1): 1-21. doi:10.3390/ani10010106ff.

Sansyzbaev B., Sydykov D., Kozhanov Zh., Akhmetov U. & Zhenishbekov A. (2024). Organization and analysis of the efficiency of production of horse breeding products by breeds and regions of the Republic of Kazakhstan. *Bulletin of Osh State University. Agriculture: agronomy, veterinary science and animal science*, 2(7), 219–226. doi:10.52754/16948696_2024_2(7)_24

Sawers L. (2003). U.S. army procurement of draft and pack animals in the civil war era. *Eastern Economic Journal*, Vol. 29, № 1, 59-67. https://core.ac.uk/download/pdf/6875145.pdf

Spugnoli P, Dainelli R. (2012). Environmental comparison ofdraught animal and tractor power. Sustain Sci. 8(1):61–72.doi: 10.1007/s11625-012-0171-7.

Stephens T. D., Splan R. K. (2013). Population history and genetic variability of the American Shire horse. *Animal Genetic Resources*. 2013. № 52. P.31-38.

Turabaev A. T. (2015). The role of the Kazakh horse in the formation of horse breeds in the Republic of Kazakhstan. *Ualikhanov readings - 19*: *collection of materials. int. scientific-practical. conf. Kokshetau*: 242.

Yakunin A. V., Sinyavskiy Y., Ibraimov Y. S. (2017). Assessment of the Nutritional Value of Mare's Milk and Fermented Mare's Milk Products and the Possibility of Their Use in Baby Food. *Current Pediatrics,* 16(3): 235–240. doi:10.15690/vsp.v16i3.1734

Yusyuk T. A. (2017). Growth dynamics of foals of the Novooleksandrivka heavy draft breed. Bulletin of the Dnipropetrovsk State Agrarian and Economic University, No. 4, 60–63.

Yusyuk T. A., Gopka B. M. (2018). The value of milk productivity in mares based on the measurements of the mares. *Abstracts of the International Conference. scientific-practical conf., priv. 120th edition of the National University of Bioresources and Natural History of Ukraine* (Kiev, May 23-25, 2018), 410–411.

Zollmann Aktiv-Kimis fermentierte Bio-Stutenmilch. Zollmann. https://www.stutenmilch.de/zollmann-aktiv (date of application 12.04.2024).

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INFLUENCE OF MICROBIOLOGICAL PREPARATION AND MINERAL FERTILIZER ON THE FORMATION OF CHICKPEAS PRODUCTIVITY

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Increasing the gross yield of chickpea seeds as a source of highly nutritious vegetable protein is one of the important aspects of ensuring national food security and nutrition of the population. The experiment to determine the effect of biological product based on nitrogen-fixing microorganisms, different doses of NPK, boron-containing fertilizer and their combinations on the formation of chickpea productivity was conducted in field conditions of a two-factor experiment in the conditions of the state enterprise "Experimental farm "Stepne" of the Institute of Pig Breeding and Agricultural Research of the NAAS" during 2023–2024. The results of the study showed that improving the nutritional regime of chickpea plants by inoculating seeds with a microbiological preparation based on nitrogen-fixing microorganisms, applying different doses of mineral fertilizers, foliar application of chickpea in the budding phase with microfertilizers and their combination improved the conditions for the formation of the leaf surface of plants and contributed to the extension of the duration of the period of its stay in an active state. Accordingly, the amount of absolutely dry aboveground mass accumulated by plants and the mass of seeds formed in beans increased. The most effective in this regard was the complex use of the microbiological preparation Anderiz (3.9 l/t) for pre-sowing inoculation of seeds and foliar application of crops with microfertilizer SmartGrow Bor-150 (1.5 l/ha) against the background of $N_{15}P_{60}K_{60}$ application, which, along with the highest values of the photosynthetic activity of plants in crops, ensured the yield of chickpea seeds at the level of 2.56 t/ha.

Keywords: chickpea, seed inoculation, foliar feeding, mineral fertilizers, photosynthesis, yield.

ВПЛИВ МІКРОБІОЛОГІЧНОГО ПРЕПАРАТУ ТА МІНЕРАЛЬНОГО УДОБРЕННЯ НА ФОРМУВАННЯ ПРОДУКТИВНОСТІ НУТУ

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Збільшення валових зборів насіння нуту, як джерела високопоживного рослинного білка є одним із вагомих аспектів забезпечення державної продовольчої безпеки та харчування населення. Дослідження із вивчення впливу біопрепарату на основі азотфіксуючих мікроорганізмів, різних доз NPK, боровмісного добрива та їх комбінацій на формування продуктивності нуту було проведено у польових умовах двохфакторного досліду в умовах державного підприємства "Дослідне господарство "Степне" Інституту свинарства і АПВ НААН" впродовж 2023–2024 рр. Результатами досліджень показали, що покращання поживного режиму рослин нуту за рахунок проведення інокуляції насіння мікробіологічним препаратом на основі азотфіксуючих мікроорганізмів, внесення різних доз мінеральних добрив, проведення позакореневого підживлення посівів нуту у фазі бутонізації мікродобоивом та їх поєднання покращувало умови формування листкової поверхні рослин та сприяло подовженню тривалості періоду перебування її у активному стані. Відповідно кількість накопиченої рослинами абсолютно сухої надземної маси та маси сформованого у бобах насіння збільшувалися. Найбільш ефективним у цьому відношенні виявилося комплексне застосування мікробіологічного препарату Андеріз (3,9 л/т) для передпосівного інокулювання насіння та позакореневого підживлення посівів мікродобривом SmartGrow бор-150 (1,5 л/га) на фоні внесення $N_{15}P_{60}K_{60}$, що поряд із найвищими значеннями показників фотосинтетичної діяльності рослин у посівах забезпечило отримання урожайності насіння нуту на рівні 2,56 т/га.

Ключові слова: *нут, інокулювання насіння, позакореневе підживлення, мінеральні добрива, фотосинтез, урожайність.*

Introduction. In solving the global problem of food security, the expansion of sown areas and the application of the latest agrotechnological aspects in the cultivation of leguminous crops as the main source of highly nutritious protein resources are of strategic importance. It is well known that protein is an important nutritional component necessary for ensuring vital functions of the body, such as body growth, tissue repair, strengthening the immune system, as well as regulating chemical and biochemical processes (Singh P., Krishnaswamy K., 2020; Çakor Ö. et al., 2019).

In the structure of protein resources used by the population for nutrition, animal proteins play a significant role (Pasiakos S.M. et al., 2015) However, as shown by the results of scientific research, the consumption of a significant amount of meat products causes the emergence and development of diseases of the cardiovascular and digestive systems, as well as the metabolic system. On the other hand, hidden hunger, caused by an insufficient amount of microelements in the daily diet, is the root cause of many health

problems, including growth retardation, underweight and the occurrence of cognitive disorders (Ibeanu V.N. et al., 2020). In this regard, an important element of the healthy nutrition system of the population of both developed countries and countries with a low standard of living can be highly nutritious products, the raw material of which is the seeds of leguminous crops (Yeremko L. et al., 2023).

A valuable representative of this group of crops is chickpeas, the main biological characteristics of which are drought resistance, heat resistance and, at the same time, resistance to the effects of low positive temperatures and short-term frosts (Karalija E. et al., 2022; Mir A.H. et al., 2021).

Due to the high content of proteins, fiber, mineral elements, as well as vitamins and biologically active compounds (Wang J. et al., 2021), the consumption of chickpea seeds normalizes the physiological processes of the human body and can be recognized as a potential candidate for the classification of "functional foods" to reduce the occurrence and development of various types of diseases (Begum N. et al., 2023; Jha U.C. et al., 2024).

Chickpea plants, by creating symbiotic relationships with nodule bacteria of the genus Mesorhizobium, such as Mesorhizobium ciceri and Mesorhizobium mediterraneum and converting or reducing molecular nitrogen from the air to ammonia, can provide about 70% of the need for this nutrient for metabolic reactions (Yeremko L. et al., 2024). At the same time, the interaction of plants with microorganisms provides them with biological control over the development of pathogens, increased resistance to the effects of adverse biotic and abiotic factors and better phosphorus availability (Monteoliva M. et al., 2022) due to the solubilization of phosphates (Sridevi M., Mallaiah K.V., 2009).

In this regard, a promising environmentally safe agrotechnological technique may be the use of biological preparations based on nitrogen-fixing bacteria for pre-sowing seed inoculation (O'Callaghan M., 2016). The introduction into the rhizosphere zone (root system formation) of highly active specific strains of nodule bacteria, which are characterized by higher competitiveness compared to aboriginal strains in the processes of infection and nodule formation and, accordingly, can increase the efficiency of legume-rhizobial symbiosis, and increase plant productivity (Pastor-Bueis R. et al., 2019; Sánchez-Navarro V. et al., 2020). At the same time, scientists note that despite the fact that atmospheric nitrogen is an unlimited resource of nitrogen nutrition, its symbiotic fixation by nodule bacteria usually cannot fully satisfy the plant's needs for this element. Thus, nitrogen uptake during legume-rhizobial symbiosis usually does not reach the same level as the uptake of NO_3^- and NH_4^+ by the root system, provided that they are sufficiently present in the soil. This phenomenon explains the inhibition of symbiotic nitrogen fixation by the application of high doses of mineral nitrogen, and plants satisfy their nitrogen needs by absorbing mineral nitrogen from the soil as non-symbiotic higher plants (Lepetit M., Brouquisse R., 2023). However, other researchers indicate the need to apply starting doses of nitrogen until the root system is sufficiently developed and the proper symbiotic apparatus is formed.

Phosphorus is the second most important element in plant nutrition. It plays a fundamental role in the regulation of various metabolic and physiological processes related to energy supply, cell division, DNA synthesis and phospholipid biosynthesis. This element is involved in the synthesis of sucrose, starch and cellulose and provides energy for the biosynthesis of phospholipids. Energy-rich phosphates, such as ATP, GTP, ADP, modulate the activity of enzymes through reverse phosphorylation (Isidra-Arellano M.S. et al., 2021).

Its sufficient presence in the soil ensures increased tolerance of plants to the effects

of increased average daily air temperature, drought, waterlogging, soil salinity and toxicity of heavy metals in it (Hawkesford M.J. et al., 2023).

Phosphorus deficiency, on the contrary, negatively affects the processes of root system development and plant productivity, which ultimately leads to a decrease in the number of fruits and their mass (Lambers H., 2022).

Potassium in the plants acts as an activator of more than 60 enzyme systems that catalyze numerous metabolic reactions. It maintains osmotic pressure and cell turgor, regulates their cation-anion balance and cytoplasmic pH, controls membrane polarization, cell expansion and stomatal movement, thus regulating the supply of CO_2 and moisture (Yeremko L. et al., 2024). This nutrient is involved in complex physiological processes of photosynthesis, synthesis of proteins, sugars and starch and their redistribution between plant organs.

Potassium is an important element that determines the adaptive capacity of plants to abiotic stresses and the level of crop quality indicators. In legumes, potassium, as the most common intracellular cation, plays an important role in the formation of a powerful root system and its absorption of moisture, the formation of root hairs, which, in turn, improves nodulation and N_2 fixation (Nakei M.D., 2022). At the same time, its presence ensures the maintenance of turgor pressure of bacterial cells, pH regulation, gene expression and activation of cellular enzymes (Domínguez-Ferreras A, 2009).

Microelements play a key role in increasing the yield of legume crops due to their influence on the physiological processes occurring in plants. They act as co-factors in the enzymatic system, and also participate in the key physiological processes of photosynthesis and respiration. These compounds contribute to the stabilization of cell walls of leguminous plants, membrane integrity, sugar transport and utilization of calcium and nitrogen (Flores R.A. et al., 2017). In addition, boron is an element that determines the reproductive ability of plants, due to its positive effect on the processes of pollination and fertilization and fruit formation. The role of this element is more pronounced during the reproductive stage of development. Scientists note that its deficiency during this period leads to pollen sterility and significantly reduces grain yield (Wang N., 2015).

At the same time, scientists note that boron deficiency indirectly affects photosynthesis by weakening vascular tissues responsible for ion transport (Goldbach H.E., Wimmer M.A., 2007). Thus, there is an assumption that disturbances in chloroplast membranes, the stomatal apparatus, the energy gradient across the membrane and thylakoid electron transport are the main reason for the decrease in photosynthesis under conditions of boron deficiency (El-FekyS.S. et al., 2012).

Thus, the review of scientific publications indicates the importance of chickpeas as a source of highly nutritious organic compounds in many areas of human life. Scientists note that the provision of plants with mineral nutrition elements is a significant factor in shaping the productivity of this crop. However, at present there is no consensus among researchers on the need to use mineral nitrogen in chickpea growing technology, and the level of mineral fertilizer is different for different growing regions. Thus, the scientific justification of the dose of mineral fertilizer for the Forest-Steppe zone of Ukraine is currently quite relevant. In this context, we conducted a study, the main goal of which was to determine the effect of different doses of mineral fertilizers in combination with the use of a biological preparation based on nitrogen-fixing microorganisms and boron microfertilizer on the formation of chickpea seed productivity and yield.

The purpose of the research is to determine the effect of different doses of mineral fertilizers in combination with the use of seed inoculation with a biological preparation based on nitrogen-fixing bacteria and foliar top dressing of crops with boron-containing fertilizer, on the formation of chickpea productivity.

Materials and methods. The study was conducted on the basis of the State Enterprise "Research Farm "Stepne" of the Institute of Pig Breeding and Agricultural Research of the NAAS".

The soil of the experimental site is a typical low-humus deep-boiling chernozem. By mechanical composition, the soil of the experimental site is a heavy loam with a content of coarse dust - 37–43%, silty particles - 25–38%. Colloidal particles are distributed in the profile to a small extent.

The value of the specific gravity of the arable soil layer (0–30 cm) is 2.63 g/cm^3, total porosity – 55.1–59.8%, moisture content of stable wilting – 8.9–9.4%, field moisture capacity – 29.7–30.5%. According to agrochemical indicators, the soil can be considered suitable for the purposes of production of agricultural crops available on the farm. Thus, the humus content in the 0–20 cm horizon is 4.9–5.2%, in the 35–45 cm horizon – 3.72–4.07%, in the 1.5 m horizon – 0.6–0.7%. In the arable soil layer, the cation absorption capacity is at the level of 33.0–35.0 mg-eq. per 100 g.

The reaction of the soil solution is slightly acidic, with a pH of saline extract at 6.3. The hydrolytic acidity of the soil is 1.6–1.9 mg-eq. per 100 g of soil. The content of basic elements in the arable layer of soil is at the level of: easily hydrolyzed nitrogen – 5.44–8.10 mg, (according to Tyurin and Kononova), mobile phosphorus – 10–15 mg (according to Chirikov), exchangeable potassium – 16–20 mg per 100 g of soil (according to Maslova).

The initial development of chickpea plants in 2023 took place with moderate air warming and a sufficient level of moisture reserves in the soil. The average air temperature in April was 9.8 °C, while the average long-term value of this indicator was at 9.3 °C. In total, 30.9 mm of precipitation fell during the month. May was characterized by moderate air temperatures and a rather uneven distribution of precipitation. The bulk of precipitation fell in the second decade of the month, while the first and third decades were dry.

Active development of the above-ground part of chickpeas occurred in June, which was characterized by a higher average daily air temperature by 0.9 °C compared to the long-term value. The amount of precipitation for the month was 33.8 mm, which is 27.9 mm less than the long-term value. Chickpeas ripened under hot, dry conditions in July and insufficient moisture supply to plants, which negatively affected the formation of crop productivity (Table 1).

Table 1

Air temperature and precipitation values for the growing season of 2023

Indicators	Months				
	April	May	June	July	August
Actual average daily air temperature, °C per month	9,8	15,0	20,3	24,3	20,0
Average daily temperature, norm per month	9,3	15,7	19,4	21,2	20,1
Absolute maximum t air, °C actual	25,8	30,6	33,8	35,3	33,8
norm	22,4	28,0	31,0	33,2	32,7
Precipitation, mm actual per month	30,9	27,3	34,6	25,2	22,9
Precipitation, mm multi-year norm per month	31,2	45,5	65,2	61,1	42,7

Weather conditions in 2024 were extremely unfavorable for the growth and development of plants and the formation of chickpea yields. In general, the growing season was characterized by a significant deficit of precipitation during the growing season of agricultural crops and chickpeas in particular. In turn, the air temperature during the growing season exceeded the average multi-year values to varying degrees. The combination of the complete absence of precipitation in the third decade of June and in July and high air temperatures led to a disruption of all physiological processes associated with the formation of the yield. (Table 2).

Table 2

Air temperature and precipitation values for the growing season of 2024

Indicators	Months				
	April	May	June	July	August
Actual average daily air temperature, °C per month	9,1	16,3	23,2	25,2	22,7
Average daily temperature, norm per month	9,3	15,7	19,4	21,2	20,1
Absolute maximum t air, °C					
actual	26,3	28,6	34,8	39,7	37,2
norm	22,4	28,0	31,0	33,2	32,7
Absolute minimum t air, °C					
actual	-4,2	3,4	8,1	13,0	12,0
norm	-3,7	2,1	6,8	9,9	8,5
Precipitation, mm actual per month	41,5	38,4	32,8	0,1	0,0
Precipitation, mm multi-year norm per month	31,2	45,5	65,2	61,1	42,7

The factors studied in the experiment were:

– seed inoculation with a biological preparation based on nodular nitrogen-fixing bacteria Mesorhizobium ciceri strain MC 285 (Anderiz 3.9 l/t) (factor A);

– mineral fertilizer control (without fertilizers), $N_{15}P_{30}K_{30}$, $N_{15}P_{60}K_{60}$, foliar application with SmartGrow boron-150, $N_{15}P_{30}K_{30}$+SmartGrow boron-150, $N_{15}P_{60}K_{60}$+SmartGrow boron-150. Fertilization with SmartGrow boron-150 microfertilizer at a dose of 1.5 l/ha was carried out in the budding phase (factor B).

The chickpea variety Budzhak was grown in the experiment. The placement of variants and repetitions was randomized. The repetition of the placement of variants was fourfold. The sowing and accounting area of the plot was 40 m^2. The chickpea growing technology in the experiment was generally accepted for the Forest-Steppe zone, with the exception of the studied elements.

During the research, the leaf surface area and dry weight of plants were determined according to the methods proposed by Z. M. Hrytsaienko and co-authors (2003), the net productivity of photosynthesis and photosynthetic potential were calculated according to the formulas proposed by A. A. Nychyporovych (1963). Before harvesting, sheaf samples were taken to determine the elements of individual plant productivity (number of beans, number of seeds in one bean, mass of seeds from one plant, mass of 1000 seeds) (Yeshchenko V. O. et al., 2005).

Research results. Plant productivity is determined as the result of the work of a holistic system. Thus, photosynthesis provides metabolic processes with carbon and

energy, on which the entire system relies, but this interaction is not linear, it is determined by the interrelation of several factors, such as development, structure of plant cover, size of leaf blades, ratio of source and sink, as well as intensity and productivity of photosynthesis. Leaf blades are the main organs of photosynthetic activity of plants, therefore their size is a key parameter of influence on various biological processes, for example, on plant growth and their reproduction. The development of the leaf surface is significantly influenced by environmental factors, among which the provision of plants with mineral nutrition elements plays a significant role.

The obtained research results indicate a positive effect of mineral fertilizer, the use of a microbiological preparation, microfertilizers, and their combination on the process of leaf surface formation of chickpea plants (Fig. 1).

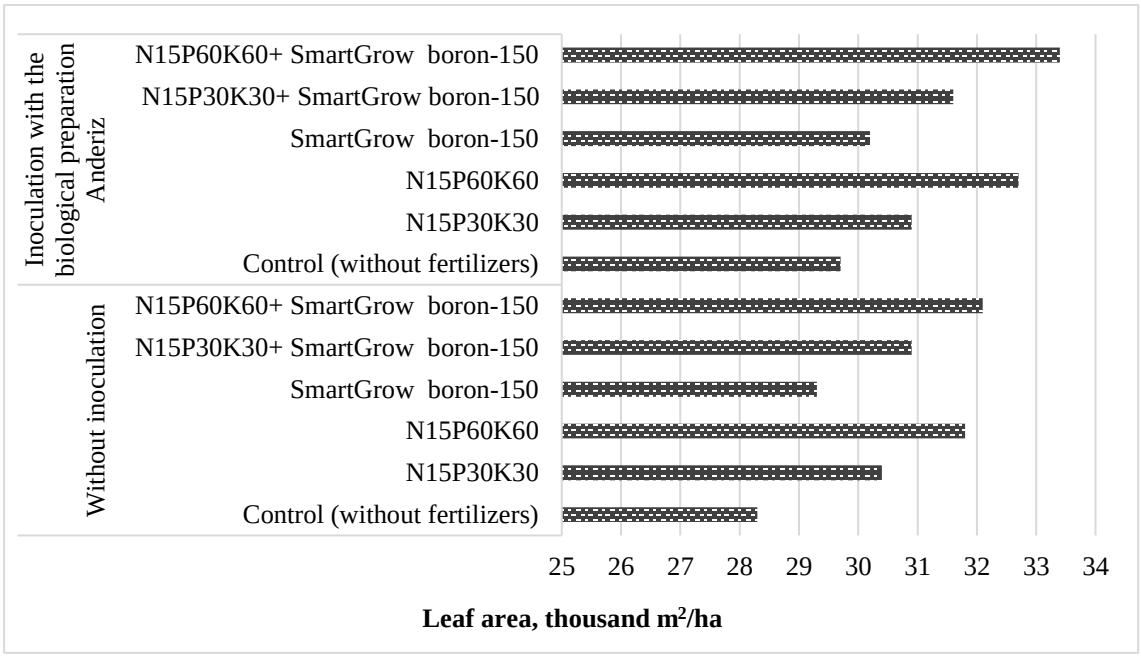

Fig. 1. **Leaf surface area of chickpea crops in the bean formation phase depending on seed bacterization and fertilization, thousand m^2/ha (2023-2024)**

In variants with application $N_{15}P_{30}K_{30}$, the leaf surface area of chickpea crops exceeded the control option in the bean formation phase by 2.1 thousand m^2/ha. The application of $N_{15}P_{50}K_{40}$ provided better development of the leaf surface than $N_{15}P_{25}K_{20}$.

Foliar fertilization of crops with boron contributed to an increase in the leaf surface of chickpea crops compared to the control by 1.0–3.8 thousand m^2/ha depending on the fertilization background. It should be noted that the values of this indicator increased as the plants' supply of mineral nutrition elements improved. Pre-sowing seed inoculation turned out to be less effective. This is indicated by the smaller values of the leaf surface area compared to the application of boron in the budding phase.

The combination of seed inoculation and foliar application of plants with boron-containing fertilizer contributed to an increase in the leaf surface area of chickpea crops in the bean formation phase, depending on the NPK dose, by 1.9–5.1 thousand m^2/ha relative to the control. The highest values of this indicator were noted in the variant of combining seed inoculation and foliar fertilization of crops with boron against the background of $N_{15}P_{60}K_{60}$ application.

In the process of photosynthesis, leaf blades absorb the energy of sunlight during the vegetation period and convert it into biomass. Thus, the amount of organic matter

synthesized by the plant is determined not only by the size of the leaf surface, but also by the duration of the period of its active functioning. The duration of the leaf surface in the active state determines the indicator of the photosynthetic potential of the crop.

The results of the studies indicate a positive effect of mineral fertilizer, seed inoculation, foliar fertilization of crops with boron and their combination on the duration of active photosynthetic work of the leaf surface of chickpea crops. The factors studied in the experiment had different effects on the duration of leaf blade functioning. In the variants of NPK application, the value of the photosynthetic potential of chickpea crops increased compared to the control by 0.064-0.166 million m^2×day/ha, and an increase in the level of mineral fertilizer extended the duration of photosynthetic work of the leaf surface of chickpea crops (Fig. 2).

Seed inoculation contributed to an increase in the value of the photosynthetic potential of chickpea crops by 0.028 million m^2×day/ha relative to the control. In the variants of combining seed inoculation and mineral fertilizer application, the excess of this indicator over the control was 4.91–10.8%, increasing as the supply of plants with mineral nutrition elements improved. The combination of seed inoculation and foliar fertilization of plants with boron contributed to an increase in the value of the photosynthetic potential of chickpea crops to 1.698 million m^2×day/ha, and it was highest in the variant where mineral fertilizers were applied at a dose of $N_{15}P_{60}K_{60}$, sowing was carried out with seeds pre-treated with the microbiological preparation Anderiz and fertilizing the crops with the microfertilizer SmartGrow boron-150.

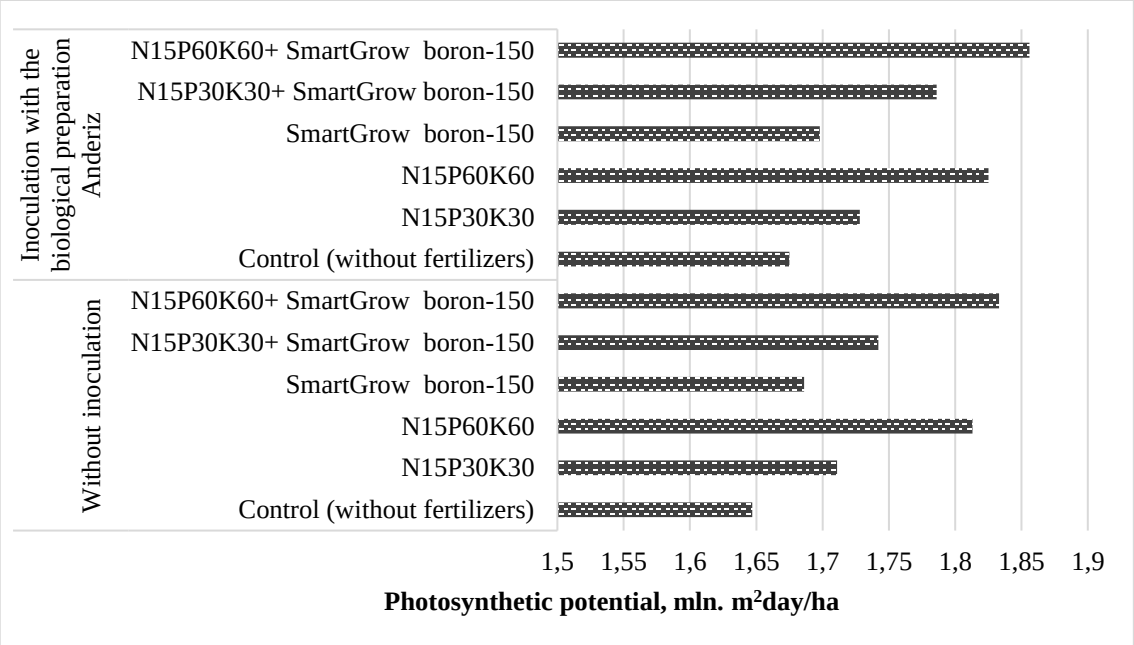

Fig. 2. **Photosynthetic potential of chickpea crops in the flowering-bean formation phase depending on seed bacterization and fertilization, million m^2×day/ha (2023-2024)**

Photosynthesis is the basis for biomass formation by plants. Thus, in the process of photosynthetic activity, plants synthesize about 95% of organic compounds, which ensures the passage of all vital processes.

In specialized structures, chloroplasts, plants use the energy of sunlight to convert carbon dioxide and water into glucose and oxygen. This process not only stimulates plant growth, but also leads to the accumulation of organic matter in the form of biomass. The

energy obtained in the process of photosynthesis accumulates in plant cells and can be used for various purposes, which makes plants an invaluable resource for both natural ecosystems and human activities.

The dynamics of the accumulation of organic biomass by plants is evidenced by the net productivity of photosynthesis. Its value expresses the amount of dry matter created in the process of photosynthesis per unit of leaf surface over a certain period of time.

In the experiment, the value of this indicator, depending on the influence of the factors studied, varied from 2.89 to 3.56 g/m^2 per day (Fig. 3). It should be noted that the influence of mineral fertilizers as a whole in the experiment on the net productivity of photosynthesis was the most pronounced. Thus, in the variants of NPK application, the excess of the values of net productivity of photosynthesis relative to the control was 11.1% for $N_{15}P_{30}K_{30}$ and 16.6% for $N_{15}P_{60}K_{60}$. Inoculation of seeds with the biological preparation Anderiz and fertilizing plants in the budding phase with the microfertilizer SmartGrow boron-150 ensured an increase in the value of this indicator by 6.50 and 7.60%, respectively, and in the variant of their combination, the net productivity of photosynthesis of chickpea crops increased compared to the control by 10.0%. The most effective in this regard was the complex application of a microbiological preparation, and $N_{15}P_{60}K_{60}$. In this variant, the excess of the net productivity of photosynthesis over the control was 23.2%.

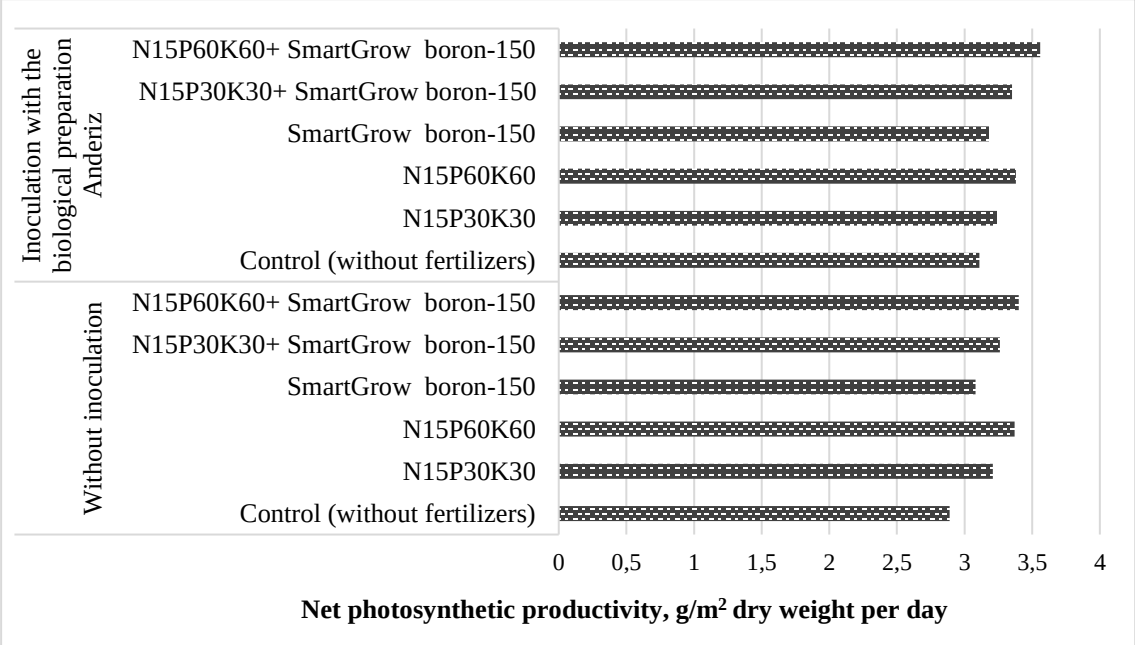

Fig. 3. **Net photosynthetic productivity of chickpeas in the flowering-bean formation phase depending on the use of the biological preparation and levels of mineral nutrition, g/m^2 per day (2023-2024)**

The intensity of accumulation of organic compounds determined the amount of dry aboveground biomass of plants. The results of the study showed a regular increase in it as the supply of plants with mineral nutrition elements improved (Table 3). Thus, the application of mineral fertilizers ensured an increase in the mass of plants in an absolutely dry state in the grain ripening phase by 0.81–1.66 g compared to the control. At the same time, it should be noted that the application of $N_{15}P_{60}K_{60}$ turned out to be more effective compared to $N_{15}P_{30}K_{30}$.

Seed inoculation contributed to an increase in the absolutely dry biomass of plants by 0.18 g compared to the control. Foliar application of plants with micronutrient SmartGrow boron-150 turned out to be more effective in this regard. The combination of seed inoculation and foliar feeding contributed to an increase in the absolute dry weight of plants relative to the control by 5.6%. The value of this indicator was maximum in the variant Anderiz + $N_{15}P_{60}K_{60}$ + SmartGrow boron-150.

Table 3

Formation of dry biomass of chickpea plants depending on seed inoculation and fertilizer levels, (2023-2024)

Seed inoculation	Fertilizer	Plant weight in a completely dry state, g
Without inoculation	Control (without fertilizers)	5,32
	$N_{15}P_{30}K_{30}$	6,13
	$N_{15}P_{60}K_{60}$	6,98
	SmartGrow boron-150	5,44
	$N_{15}P_{30}K_{30}$+ SmartGrow boron-150	6,34
	$N_{15}P_{60}K_{60}$+ SmartGrow boron-150	7,42
Inoculation with the biological preparation Anderiz	Control (without fertilizers)	5,50
	$N_{15}P_{30}K_{30}$	6,25
	$N_{15}P_{60}K_{60}$	7,34
	SmartGrow boron-150	5,62
	$N_{15}P_{30}K_{30}$+ SmartGrow boron-150	6,72
	$N_{15}P_{60}K_{60}$+ SmartGrow boron-150	8,13

The results obtained indicate a positive influence of the studied factors and their complex interaction on the mass of 1000 chickpea seeds. The studies have established an increase in the value of this indicator by 10.9–18.8% in the variants of NPK application relative to the control. At the same time, the highest values of the mass of 1000 chickpea seeds were also noted when applying maximum doses of mineral fertilizers.

In the variants of seed inoculation and foliar application of plants with boron-containing fertilizer, the mass of 1000 seeds increased by 1.1 and 2.6%, respectively, compared to the control. The most effective was the complex use of the studied factors, in particular the application of $N_{15}P_{60}K_{60}$ and the combination of seed inoculation and foliar application of plants, which ensured an increase in this indicator relative to the control by 8.4%.

Intensive growth of the above-ground part and root system of plants, sufficient development of the photosynthetic surface of plants and increased photosynthetic activity, contributed, in turn, to an increase in the amount of synthesized metabolites and the accumulation of dry matter by plants, as well as an increase in the average number of beans and seeds formed on plants, the mass of 1000 seeds. The values of these elements of plant productivity determined the increase in the seed yield (Table 4).

The highest values of this indicator were recorded in the variant of combining seed inoculation, foliar application of plants with microfertilizer SmartGrow boron-150 $N_{15}P_{60}K_{60}$ mineral fertilization. The average seed yield over the years of the study in this variant of the experiment was the highest and amounted to 2.56 t/ha. The increase in chickpea seed yield from the application of different doses of mineral fertilizers was at the level of 0.14–0.36 t/ha, relative to the variant without fertilizers.

Table 4

Chickpea seed yield depending on the use of biological product and mineral nutrition elements, t/ha

Seed inoculation	Fertilizer	Seed yield, t/ha		Average by years, t/ha
		2023	2024	
Without inoculation	control (without fertilizers)	2,24	1,68	1,96
	$N_{15}P_{30}K_{30}$	2,46	1,74	2,10
	$N_{15}P_{60}K_{60}$	2,78	1,86	2,32
	SmartGrow boron-150	2,31	1,77	2,04
	$N_{15}P_{30}K_{30}$+ SmartGrow boron-150	2,57	1,89	2,23
	$N_{15}P_{60}K_{60}$+ SmartGrow boron-150	2,91	1,95	2,43
Inoculation with the biological preparation Anderiz	control (without fertilizers)	2,28	1,71	2,00
	$N_{15}P_{30}K_{30}$	2,50	1,83	2,17
	$N_{15}P_{60}K_{60}$	2,83	1,98	2,41
	SmartGrow boron-150	2,35	1,83	2,09
	$N_{15}P_{30}K_{30}$+ SmartGrow boron-150	2,63	1,96	2,30
	$N_{15}P_{60}K_{60}$+ SmartGrow boron-150	3,08	2,04	2,56
$HIP_{0,95}$		factor A – 0.09; factor B – 0.12; interaction of factors AB – 0.15.		

Seed inoculation and foliar fertilization of crops contributed to an increase in seed yield compared to the control by 0.03 and 0.09 t/ha, respectively, and in the variant of their combination, the increase in yield was 0.15 t/ha.

Discussion. The magnitude of plant productivity is expressed by the totality of the effects of various environmental factors on a number of physiological and morphological processes occurring in plants during the growing season. Photosynthesis is a fundamental process during which the energy of solar radiation is used by plants to convert CO_2 into carbohydrates, which constitute about 90% of their biomass (Simkin et al., 2019; Muhie, 2022).

In turn, the productivity of photosynthetic activity of plants is largely determined by the size of the leaf surface, as the main assimilation organ of plants. Its size can serve as an indicator of the strategy of resource use by plants and have important consequences for the energy and water balance. The main factor in the formation of a developed leaf surface is the provision of plants with mineral nutrition elements. The results of our research indicate a positive effect of mineral fertilizer on the development of the leaf surface of plants. Its largest value was observed in the variant of the combination of factors studied. Along with this, the obtained data are consistent with the studies conducted by H. Ali, M.A. Khan and S.A. Randhawa (2004), where improving plant phosphorus nutrition by increasing the dose of P_2O_5 from 90 to 120 kg/ha increased the value of the leaf area index to 3.78. At the same time, scientists note that nitrogen deficiency leads to a decrease in the leaf area index, the productivity of photosynthetic activity and, accordingly, plant biomass (Chemining wa G. N., Vessey J. K., 2006).

In other studies, an increase in the level of nitrogen absorption by plants with improved phosphorus nutrition is noted. R.S. Jat, I.P.S. Ahalawat (2004) reported that the application of $P_{26.4}$ significantly increased the total absorption of nitrogen and phosphorus

by plants. In the studies of R. Meena et al. (2004), the introduction of P_{60} increased the amount of N, P, K absorbed by plants.

Other researchers agree with these statements, who found an increase in the intensity of chickpea growth processes due to the use of macronutrients, i.e. N, P and K (Goud V.V. et al., 2014; Shah T. et al., 2016), micronutrients (El-Habbasha S.F. et al., 2012), biofertilizers (Dutta D., Bandyopadhyay P., 2009). They note that the positive effect of microorganisms on plant growth and development may be associated not only with the improvement of their nitrogen nutrition due to legume-rhizobial interaction, but also due to the synthesis of phytohormones such as auxin, secondary metabolites in inoculated plants. They promote seed germination, root system development and at the same time increase the amount of moisture and nutrients absorbed by it, which in turn stimulates the growth of the leaf surface (Werner D., Newton W. E., 2005)

Scientific studies indicate an increase in the growth rate of the absolutely dry matter of plants, due to better development of the leaf surface, which in turn increases the accumulation of solar radiation by plants and the production of organic compounds by increasing the intensity and productivity of photosynthesis (McKenzie, B. A., Hill, G. D., 1995). This position was confirmed by the results of our studies, where an increase in the area of the leaf surface and an extension of the duration of its active functioning due to the complex use of seed inoculation, foliar application of plants with boron against the background of the application of various doses of mineral fertilizers contributed to the enhancement of the production of organic matter by plants, an increase in the mass of plants in a completely dry state and an increase in the level of seed yield up to 2.56 t/ha. Our results are consistent with the conclusions made by Reinprecht Y. With co-authors (2020) in the course of their studies. They note that the main source of nutrients entering the seed during its formation, filling and ripening is the process of photosynthesis and root nutrition, as well as the reuse of organic compounds from vegetative organs. Improving the conditions for the formation of plant productivity against the background of the application of mineral fertilizers may be associated not only with the positive effect of each nutrient on vital processes, but also with their synergistic effect. This is evidenced by the results of our studies, which are also consistent with the data obtained by other scientists (Meena R. et al., 2020., Kumar D. et al., 2014).

Scientists also note that a positive aspect of increasing plant productivity can be not only an increase in the number of doses of applied fertilizers, but also the ratio of mineral nutrition elements in them (Rashid A. et al., 2013). At the same time, the increase in leaf surface area, plant mass in a completely dry state and, accordingly, seed yield in the variant of foliar fertilizing of plants with boron and its combination with the application of mineral fertilizers, noted in our study, can be explained by the favorable effect of these nutrients on metabolism and biological activity, as well as their stimulating effect on the development of the leaf surface and an increase in the concentration of photosynthetic pigments, the activity of enzymes, which, in turn, have a stimulating effect on the vegetative growth of plants.

Conclusions:

1. The use of a microbiological preparation based on nitrogen-fixing bacteria for pre-sowing treatment of chickpea seeds, mineral fertilizers, foliar application with microfertilizer SmartGrow Bor-150 and their combination showed a positive effect on the formation of the leaf surface by chickpea plants. In this regard, the most effective was the combination of seed inoculation, foliar application of plants with boron and $N_{15}P_{60}K_{60}$ fertilization.

2. The size of the leaf surface, productivity and duration of its photosynthetic work, which increased as the supply of plants with mineral nutrition elements improved,

in turn determined the intensity of the accumulation of aboveground dry biomass by plants and the supply of organic compounds to seeds during their formation and filling. Accordingly, the dry weight of plants and the weight of 1000 seeds were the highest in the combination of seed inoculation foliar application of plants with boron and $N_{15}P_{60}K_{60}$ fertilization.

3. It was established that the introduction of NPK provided an increase in the yield of chickpea seeds relative to the control at the level of 0.14-0.36 t/ha. In the variants of using the microbiological preparation Anderiz based on nitrogen-fixing bacteria and the microfertilizer SmartGrow Bor-150, the seed productivity of chickpea crops increased compared to the control by 0.03 and 0.09 t/ha, respectively, and in the variant of their complex application, the yield increased by 0.15 t/ha. The most appropriate is the combination of seed inoculation, foliar application of plants with SmartGrow Bor-150 and $N_{15}P_{60}K_{60}$ fertilization, which makes it possible to increase the yield of chickpea seeds to 2.56 t/ha.

References

Abdel-Motagally, F.M.F., & El-Zohri, M. (2016). Improvement of wheat yield grown under drought stress by boron foliar application at different growth stages. *Journal of the Saudi Society of Agricultural Sciences,* 17 (2), 178-185. https://doi.org/10.1016/j.jssas.2016.03.005

Ali, H., Khan, M.A. & Randhawa, S.A. (2004). Interactive effect of seed inoculation and phosphorus application on growth and yield of chickpea (*Cicer arietinum* L.). *International Journal of Agriculture & Biology*, 6, 110-112.

Begum, N., Khan, Q.U., Liu, L.G., Li, W., Liu, D. & Haq, I.U. (2023) Nutritional composition, health benefits and bio-active compounds of chickpea (*Cicer arietinum* L.). *Frontiers in Nutrition*, 10, 1218468. https://doi.org/10.3389/fnut.2023.1218468

Çakor, Ö., Uçarlo, C., Tarhan, Ç., Pekmez, M., & Tyrgut-Kara, N. (2019). Nutritional and health benefits of legumes and their distinctive genomic properties. *Food Science and Technology*, 39(1), 1-12. https://doi.org/10.1590/fst.42117

Chemining wa, G. N., & Vessey, J. K. (2006). The abundance and efficacy of Rhizobium leguminosarum bv. *viciae* in cultivated soils of eastern Canadian prairie. *Soil Biology and Biochemistry*, 38, 294-302.

Domínguez-Ferreras, A., Muñoz, S., Olivares, J., Soto, M.J., & Sanjuan, J. (2009). Role of potassium uptake systems in *Sinorhizobium meliloty* osmoadaptation and symbiotic performance. *Journal of Bacteriology,* 191, 2133–2143. https://doi.org/10.1128/jb.01567-08

Dutta, D. & Bandyopadhyay, P. (2009) Performance of chickpea (*Cicer arietinum* L.) to application of phosphorus and bio-fertilizer in laterite soil. *Archives of Agronomy and Soil Science*, 55(2), 147-155.

El-Feky, S.S., El-Shintinawy, F., Shaker, E.M., & El-Din, H.A.S. (2012). Effect of elevated boron concentrations on the growth and yield of barley (*Hordeum vulgare* L.) and alleviation of its toxicity using different plant growth modulators. *Australian Journal of Crop Science*, 6, 1687–1695.

El-Habbasha, S.F., Ahmed, Amal G., & Magda M. H. (2012) Response of some chickpea varieties to compound foliar fertilizer under sandy soil conditions. *Journal of Applied Sciences Research,* 8 (10), 5177-5183.

Flores, R.A., da Silva, R.G., da Cunha, P.P., Damin, V., de Abdala, K.O., Arrud,a E.M., Rodrigues, R.A., & Maranhão, D.D.C. (2017). Economic viability of *Phaseolus vulgaris* (BRS Estilo) production in irrigated system in a function of application

of leaf boron. *Acta Agriculturae Scandinavica. Section B, Soil and Plant Science*, 67, 697–704. https://doi.org/10.1080/09064710.2017.1329454

George, T.S., Hinsinger, P., & Turner, B.L. (2016). Phosphorus in soils and plants – facing phosphorus scarcity. *Plant and Soil*, 401, 1–6. https://doi.org/10.1007/s11104-016-2846-9

Goldbach, H.E., & Wimmer, M.A. (2007). Boron in plants and animals: Is there a role beyond cell wall structure? *Journal of Plant Nutrition and Soil Science*, 170, 39–48. https://doi.org/10.1002/jpln.200625161

Goud, V.V., Konde, N.M., Mohod, P.V. & Kharche, V.K. (2014) Response of chickpea to potassium fertilization on yield, quality, soil fertility and economic in vertisols. *Legume Research*, 37 (3), 311-315.

Hawkesford, M.J., Cakmak, I., Coskun, D., De Kok, L.J., Lambers, H., Schjoerring, J.K., & White, P.J. (2023). Functions of macronutrients. *Marschner's Mineral Nutrition of Plants*; Academic Press: Cambridge, MA, USA, 201–281. https://doi.org/10.1016/B978-0-12-819773-8.00019-8

Hrytsaienko, Z. M., Hrytsaienko, A. O., & Karpenko, V. P. (2003) Metody biolohichnykh ta ahrokhimichnykh doslidzhen roslyn i gruntiv [Methods of biological and agrochemical studies of plants and soils] K.: ZAT "NIChLAVA". 320 (In Ukrainian).

Ibeanu, V. N., Edeh, C. G., & Ani, P. N. (2020). Evidence-based strategy for prevention of hidden hunger among adolescents in a suburb of Nigeria. *BMC Public Health*, 20(1), 1683. https://doi.org/10.1186/s12889-020-09729-8

Isidra-Arellano, M.C., Delaux, P.M., & Valds-Lpez, O. (2021). The phosphate starvation response system: its role in the regulation of plant-microbe interactions. *Plant and Cell Physiology*, 62, 392–400. https://doi.org/10.1093/pcp/pcab016

Jat, R.S., & Ahalawat I.P.S. (2004). Effect of vermicompost, biofertilizer and nutrient uptake by gram (*Cicer arietinum*) and their residual effect on fodder maize (*Zea mays*). *Indian journal of agricultural science*, 74, 359-361.

Jha, U.C., Nayyar, H., Thudi, M., Beena, R., Prasad, P.V.V. & Siddique, K.H.M. (2024) Unlocking the nutritional potential of chickpea: strategies for biofortification and enhanced multi-nutrient quality. *Frontiers in Plant Science*, 15, 1391496. https://doi.org/10.3389/fpls.2024.1391496

Karalija, E., Vergata, C., Basso, M.F., Negussu, M., Zaccai, M., Grossi-de-Sa, M.F.; & Martinelli, F. (2022). Chickpeas' tolerance of drought and heat: Current knowledge and next steps. *Agronomy,* 12, 2248. https://doi.org/10.3390/agronomy12102248

Kaur, N., Sharma, P. & Sharma, S. (2015). Co-inoculation of *Mesorhizobium sp.* and plant growth promoting rhizobacteria Pseudomonas sp. as bio-enhancer and bio-fertilizer in chickpea (*Cicer arietinum* L.). *Legume Researsh*, 38, 367-374. https://doi.org/10.5958/0976-0571.2015.00099.5

Kaushik, P., Pati, P.H., Khan, M.L., & Khare, P.K. (2021). A quick and simple method for estimating leaf area by leaf weight. *International Journal of Botany Studies*, 6, 1286–1288.

Koul, B., Sharma, K., Sehgal, V., Yadav, D., Mishra, M.; & Bharadwaj, C. (2022). Chickpea (*Cicer arietinum* L.) biology and biotechnology: from domestication to biofortification and biopharming. *Plants*, 11, 2926. https://doi.org/10.3390/plants11212926

Kumar, D., Arvadiya, L.K., Kumawat, A.K., Desai, K.L. & Patel, T. U. (2014). Yield, protein content, nutrient content and uptake of chickpea (*Cicer arietinum* L.) as influenced by graded levels of fertilizers and bio-fertilizers. *Research Journal of*

Chemical and Environmental Sciences, 2, 60-64.

Lambers, H. (2022). Phosphorus acquisition and utilization in plants. *Annu. Review of Plant Biology,* 73, 17–42. https://doi.org/10.1146/annurev-arplant-102720-125738

Lepetit, M. & Brouquisse, R. (2023). Control of the rhizobium–legume symbiosis by the plant nitrogen demand is tightly integrated at the whole plant level and requires interorgan systemic signaling. *Frontiers in Plant Science*, 14, 1114840. https://doi.org/10.3389/fpls.2023.1114840

McKenzie, B. A., & G. D. Hill. 1995. Growth and yield of two chickpea (*Cicer arietinum* L.) varieties in Canterbury, New Zealand. *New Zealand Journal of Crop and Horticultural Science,* 23, 467-474.

Meena, R., Meena M., Sharma, P.K., & Chetan Kumar, C. (2020). Effect of fertility levels and bio-fertilizers on growth and yield of chickpea (*Cicer arietinum* L.). *International Journal of Current Microbiology and Applied Sciences*, 9(2), 3098-3103. https://doi.org/10.20546/ijcmas.2020.902.357

Michail ,T., Walter, T., Astrid, W., Walter, G., Dieter, G., & Maria, S.J. (2004). A survey of foliar mineral nutrient concentrations of *Pinus canariensis* at field plots in Tenerife. *Forest Ecology and management*, 189, 49-55.

Mir, A.H., Bhat, M.A., Dar, S.A., Sofi, P.A., Bhat, N.A., & Mir, R.R. (2021). Assessment of cold tolerance in chickpea (*Cicer* spp.) grown under cold/freezing weather conditions of North-Western Himalayas of Jammu and Kashmir, India. *Physiology and Molecular Biology of Plants,* 27(5), 1105–1118. https://doi.org/10.1007/s12298-021-00997-1

Monteoliva, M., Valetti, L., Taurian, T., Crociara, C. S., & Guzzo, M. C. (2022). Synthetic communities of bacterial endophytes to improve the quality and yield of legume crops. *Intech Open.* 1–36. https://doi.org/10.5772/intechopen.102519

Muhie, S.H. (2022). Optimization of photosynthesis for sustainable crop production. *CABI Agriculture and Bioscience*, 3, 50. https://doi.org/10.1186/s43170-022-00117-3

Nakei, M.D., Venkataramana, P.B., & Ndakidemi, P.A. (2022). Soybean-nodulating rhizobia: ecology, characterization, diversity, and growth promoting functions. *Frontiers in Sustainaible Food System*, 6, 824444. https://doi.org/10.3389/fsufs.2022.824444

Nychyporovych, A. A. (1963). O putiakh povyshenyia produktyvnosty fotosynteza rastenyi v posevakh. V kn.: Fotosyntez y voprosy produktyvnosty rastenyi. M.: Yzd-vo AN SSSR. 5–36.

O'Callaghan, M. (2016). Microbial inoculation of seed for improved crop performance: issues and opportunities. *Applied Microbiology and Biotechnology*, 100, 5729–5746. https://doi.org/10.1007/s00253-016-7590-9

Oosterhuis, D.M., Loka, A.D., Kawakami, E.M., & William, T. (2014). Pettigrew chapter three - the physiology of potassium in crop production. *Advances in agronomy*, 126, 203-233. https://doi.org/10.1016/B978-0-12-800132-5.00003-1

Pasiakos, S. M., Agarwal, S., Lieberman, H. R., & Fulgoni, V. L. (2015). Sources and amounts of animal, dairy, and plant protein intake of US adults in 2007–2010. *Nutrients*, 7(8), 7058–7069. https://doi.org/10.3390/nu7085322

Pastor-Bueis, R., Sánchez-Cañizares, C., James, E.K. & González-Andrés, F. (2019). Formulation of a highly effective inoculant for common bean based on an autochthonous elite strain of *Rhizobium leguminosarum* bv. phaseoli, and genomic-based insights into its agronomic performance. *Frontiers in Microbiol*ogy, 10, 2724. https://doi.org/10.3389/fmicb.2019.02724

Prajapati, K., & Modi, H.A. (2012). The importance of potassium in plant growth – a review. *Indian Journal of Plant Sciences,* 1(02-03), 177-186.

Rashid, A., Ishaque, M., Hameed, K., Shabbirand, M. & Ahmad. (2013). Growth and yield response of three chickpea cultivars to varying NPK levels. *Asian Journal of Agriculture and Biol*ogy, 1, 95-99.

Reinprecht, Y., L. Schram, F. Marsolais, T.H. Smith, B. Hill, & K.P. Pauls. (2020). Effects of nitrogen application on nitrogen fixation in common bean production. *Frontiers in Plant Science,* 11, 1172. https://doi.org/10.3389/fpls.2020.01172

Sánchez-Navarro, V., Zornoza, R., Faz, Á., Egea-Gilabert, C., Ros, M., Pascual, J. A., & Fernández, J. A. (2020). Inoculation with different nitrogen-fixing bacteria and arbuscular mycorrhiza affects grain protein content and nodule bacterial communities of a Fava Bean Crop. *Agronomy*, 10 (6), 768. https://doi.org/10.3390/agronomy10060768

Shah, T., Fareed, A. & Nauman, M. (2016). Yield and quality response of chickpea cultivars to different NPK levels. *Austin Food Sci*ences, 1(4), 1-4.

Shen, J., Yuan, L., Zhang, J., Li, H., Bai, Z., Chen, X., Zhang, W., & Zhang, F. (2011). Phosphorus dynamics: from soil to plant. *Plant Physiol*ogy, 156, 997-1005. https://doi.org/10.1104/pp.111.175232

Simkin, A. J., Patricia, E. López-Calcagno, & Christine, A. (2019). Raines Feeding the world: improving photosynthetic efficiency for sustainable crop production. *Journal of Experimental Botany*, 70 (4), 1119–1140. https://doi.org/10.1093/jxb/ery445

Singh, P., & Krishnaswamy, K. (2022). Sustainable zero-waste processing system for soybeans and soy by-product valorization. *Trends in Food Science & Technology,* 128, 331–344. https://doi.org/10.1016/j.tifs.2022.08.015

Sridevi, M., & Mallaiah, K.V. (2009). Phosphate solubilization by Rhizobium strains. *Indian Journal of Microbiol*ogy, 49, 98. https://doi.org/10.1007/s12088-009-0005-1

Wang, J., Li, Y., Li, A., Liu, R.H., Gao, X., Li, D., Kou, X., & Xue, Z. (2021). Nutritional constituent and health benefits of chickpea (*Cicer arietinum L.*): A review. *Food Research International*, 150, 110790. https://doi.org/10.1016/j.foodres.2021.110790

Wang, N., Yang, C., Pan, Z., Liu, Y. & Peng, S. (2015). Boron deficiency in woody plants: Various responses and tolerance mechanism. *Frontiers in Plant Science*, 6, 916. https://doi.org/10.3389/fpls.2015.00916

Werner, D., & Newton, W. E. (2005). Nitrogen fixation in agriculture, forestry, ecology and environment. Netherlands, 2005. 347.

Yeremko, L., Hanhur, V., & Staniak, M. (2024) Effect of mineral fertilization and seed inoculation with microbial preparation on seed and protein yield of pea (*Pisum sativum* L.). *Agronomy*, 14, 1004. https://doi.org/10.3390/agronomy14051004

Yeremko, l., Hanhur, V., & Staniak, M. (2024). Effect of fertilization and microbial preparations on productivity of chickpea (*Cicer arietinum* L.). *Acta Agrobotanica*, 77, https://doi.org/10.5586/aa/182829

Yeshchenko, V. O., Kopytko, P. H., Opryshko, V. P., & Kostohryz, P. V. (2005). Osnovy naukovykh doslidzhen v ahronomii [fundamentals of scientific research in agronomy]. Diia. 288 (In Ukrainian).

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ANALYSIS OF INDIVIDUAL DYNAMICS OF PROGESTERONE LEVELS IN FEMALE DOGS DEPENDING ON AGE, BODY CONDITION, AND TEMPERAMENT

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The aim of this study was to examine the influence of individual characteristics of female dogs (Canis familiaris) of the Bull Terrier breed on the dynamics of progesterone levels in their blood throughout the estrous cycle. The experiment involved 25 clinically healthy female dogs divided into three age groups: young (1.4 years), middle-aged (4.8 years), and older dogs (8.6 years). The health status of the animals was assessed through clinical examinations and laboratory analyses, including the determination of reproductive status using folliculometry, vaginal cytology, and blood progesterone level analysis. Blood samples were collected from the jugular vein during key periods of the estrous cycle and analyzed using an enzyme-linked immunosorbent assay ("Progesterone – ELISA," HEMA, Ukraine). Additionally, the impact of body condition (evaluated using the BCS scale) and temperament (assessed using the C-BARQ questionnaire) on hormonal status was investigated. To this end, 12 groups of female dogs were formed according to behavioral characteristics (aggression, fear and anxiety, excitability, learning ability, and obedience). All experimental procedures complied with the requirements of the Ukrainian Law "On the Protection of Animals from Cruelty" and the principles of the European Convention for the Protection of Vertebrate Animals Used for Experimental and Scientific Purposes. The results of two-factor ANOVA revealed that, regardless of age, body weight, or temperament, the stage of the estrous cycle was the determining factor influencing blood progesterone levels ($F=79.8–173.0$; $P<0.001$). Among individual characteristics, body weight had the greatest impact ($F=16.3$; $P<0.001$), while age differences showed a statistically significant but less pronounced effect ($F=3.65$; $P<0.05$). The temperament of female dogs also significantly affected progesterone levels, particularly in terms of aggression ($F=10.68$; $P<0.001$), fear and

anxiety (F=9.21; P<0.001), and excitability (F=7.6; P<0.001). In contrast, learning ability and obedience did not show a significant effect on hormonal status (F=0.91; P=0.41). Analysis of interaction effects revealed a significant relationship between body weight, behavioral characteristics (aggression, fear), and the stage of the estrous cycle (F=2.24–2.92; P<0.005–0.001). This indicates that individual physiological and behavioral traits influence the endocrine regulation of reproductive function in female dogs. The findings expand the understanding of regulatory mechanisms in the estrous cycle of dogs and emphasize the need to consider individual characteristics when planning veterinary interventions and breeding programs.

Keywords: progesterone, estrous cycle, dogs, body condition, temperament, reproductive function, hormonal regulation.

АНАЛІЗ ІНДИВІДУАЛЬНОЇ ДИНАМІКИ РІВНЯ ПРОГЕСТЕРОНУ У СУК ЗАЛЕЖНО ВІД ВІКУ, СТАНУ ТІЛА ТА ТЕМПЕРАМЕНТУ

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Метою цього дослідження було вивчення впливу індивідуальних характеристик сук (Canis familiaris) породи бультер'єр на динаміку рівня прогестерону в їх крові протягом естрального циклу. В експерименті брали участь 25 клінічно здорових собак, розділених на три вікові групи: молоді (1,4 року), середнього віку (4,8 року) та старші (8,6 року). Стан здоров'я тварин оцінювали за допомогою клінічних та лабораторних досліджень, включаючи визначення репродуктивного статусу за допомогою фолікулометрії, вагінальної цитології та аналізу рівня прогестерону в крові. Зразки крові брали з яремної вени в ключові періоди естрального циклу та аналізували за допомогою імуноферментного аналізу («Прогестерон – ІФА», НЕМА, Україна). Додатково досліджували вплив стану тіла (оцінюваного за шкалою BCS) та темпераменту (оцінюваного за допомогою опитувальника C-BARQ) на гормональний статус сук. Для цього було сформовано 12 груп тварин за поведінковими характеристиками (агресія, страх і тривога, збудливість, здатність до навчання та слухняність). Усі експериментальні процедури відповідали вимогам Закону України «Про захист тварин від жорстокого поводження» та принципам Європейської конвенції про захист хребетних тварин, що використовуються в експериментальних та наукових цілях. Результати двофакторного дисперсійного аналізу (ANOVA) показали, що незалежно від віку, маси тіла чи темпераменту, стадія естрального циклу була визначальним фактором, що впливав на рівень прогестерону в крові (F=79,8–173,0; P<0,001). Серед індивідуальних характеристик найбільший вплив мала маса тіла (F=16,3; P<0,001), тоді як вікові відмінності показали

статистично значущий, але менш виражений вплив (F=3,65; P<0,05). Темперамент сук також суттєво впливав на рівень прогестерону, зокрема, щодо агресії (F=10,68; P<0,001), страху та тривоги (F=9,21; P<0,001) та збудливості (F=7,6; P<0,001). Натомість, здатність до навчання та слухняність не виявили значного впливу на гормональний статус (F=0,91; P=0,41). Аналіз ефектів взаємодії виявив значний зв'язок між масою тіла, поведінковими характеристиками (агресія, страх) та стадією естрального циклу (F=2,24–2,92; P<0,005–0,001). Це вказує на те, що індивідуальні фізіологічні та поведінкові риси впливають на ендокринну регуляцію репродуктивної функції у сук. Отримані результати розширюють розуміння регуляторних механізмів естрального циклу собак та підкреслюють необхідність врахування індивідуальних характеристик під час планування ветеринарних втручань та програм розведення.

Ключові слова: прогестерон, естральний цикл, собаки, стан тіла, темперамент, репродуктивна функція, гормональна регуляція.

Introduction. The reproductive function of dogs is a complex process that depends on numerous factors such as physiological condition, age, breed, temperament, and body condition. The reproductive cycles of dogs, in terms of physiological neurohumoral regulatory mechanisms, are unique among domestic animal species. To date, many physiological and clinical questions concerning the regulation of the estrous cycle in dogs remain unresolved (Concannon, 2009; Martin et al., 2009). Existing information on the individual characteristics of estrous cycle regulation in female dogs is fragmented and ambiguous. In particular, data on the impact of age, body condition, and temperament on the dynamics of sex hormones in the blood of Bull Terrier females are lacking. A better understanding of the mechanisms involved in the establishment and maintenance of pregnancy may be useful for elucidating the pathogenesis of some common disorders and contribute to the development of better clinical protocols, ensuring an individualized approach for each patient (de Carvalho Papa & Kowalewski, 2020).

The Corpus Luteum is a temporary endocrine gland formed after ovulation in the ovary at the site of the Graafian follicle. The corpus luteum produces the hormone progesterone, and its name derives from the characteristic yellow color of its content. In dogs, the corpus luteum (CL) is the sole source of progesterone (P4) and estradiol (E2) during diestrus (Papa & Hoffmann, 2011). The functional duration of the CL is determined by endocrine, paracrine, and autocrine factors (Sousa et al., 2016). Local growth factors, cytokines, and prostaglandins modulate CL function, creating a balance that leads to luteal regression in non-pregnant dogs or luteolysis during pregnancy (Mariusz P. Kowalewski, 2014). In pregnant dogs, the trophoblast acts as the fetomaternal compartment responsible for synthesizing prostaglandin F2α (PGF2a), which actively participates in prepartum luteolysis (Mariusz Pawel Kowalewski et al., 2010). Progesterone (P4) is a steroid hormone responsible for preparing the endometrium for implantation of the fertilized egg and for maintaining pregnancy. After implantation, the corpus luteum continues to produce progesterone during the early stages of pregnancy until the placenta develops and takes over progesterone production for the remainder of gestation (Holesh, Bass, & Lord, 2023). Progesterone plays a pivotal role in fertility. The expression and activation of the progesterone receptor (PGR) are essential for ovulation (Park et al., 2020). Additionally, progesterone is critical for preparing the uterine environment for implantation, embryonic development, and regulation of the estrous cycle (Pereira, Mainigi, & Strauss III, 2021).

Progesterone levels are widely used as a clinical biomarker in the reproductive management of dogs (Conley, Gonzales, Erb, & Christensen, 2023). The variability in

circulating P4 levels among dogs is associated with the number of ovulations and corpora lutea (Concannon, Butler, Hansel, Knight, & Hamilton, 1978). In many species, plasma progesterone concentrations increase with the rate of ovulation and the number of CL (Knox, Vatzias, Naber, & Zimmerman, 2003). Adult dogs exhibit higher efficiency in P4 synthesis compared to younger ones, indicating that luteal endocrine activity undergoes maturation as dogs transition from youth to adulthood (Marinelli, Rota, Carnier, Da Dalt, & Gabai, 2009). Beyond its primary function, progesterone also belongs to the group of neurosteroids. It is metabolized in all regions of the central nervous system (Hanukoglu, Karavolas, & Goy, 1977) and possesses neuromodulatory, neuroprotective, and neurogenic properties (Schumacher et al., 2004). These effects are mediated through interactions with non-nuclear progesterone receptors such as mPR and PGRMC1, as well as other receptors like σ1 and nACh (Singh, Su, & Ng, 2013).

Unlike other domestic animals, the reproductive system of female dogs is characterized by the absence of increased estrogen levels during pregnancy and before parturition (Concannon, 2011), while luteal regression occurs despite elevated pituitary hormone levels (Concannon, 2009). Elevated progesterone levels are also observed during pseudopregnancy (Feldman, Nelson, Reusch, & Scott-Moncrieff, 2014). Furthermore, significant individual variations in sex hormone levels have been observed in female dogs (Concannon, Castracane, Temple, & Montanez, 2009). Previous studies have indicated considerable variation in progesterone levels depending on the breed and physiological condition of dogs. For instance, Luz (2006) demonstrated significant individual fluctuations in P4 levels in the plasma of mixed-breed dogs, whereas these variations were less pronounced in Beagle females. Moreover, the impact of age, body weight, and temperament on reproductive efficiency has also attracted researchers' attention; however, the consistency of data in these aspects remains insufficient.

This study aimed to determine the effects and differences in progesterone dynamics in the plasma of female dogs depending on their individual characteristics such as age, body weight, and temperament. The objective of the research was to identify key factors influencing progesterone levels in female dogs and their interactions, which could improve the understanding of hormonal regulation mechanisms and provide a scientific basis for optimizing canine breeding programs.

Materials and methods. The experiment was conducted on 25 female dogs (Canis familiaris) of the Bull Terrier breed of various ages, body conditions, and temperaments. At the time of the study, all animals were clinically healthy, without signs of infectious or parasitic diseases. Health status was assessed through clinical examinations and laboratory analyses. To evaluate the humoral component of the reproductive status of the females, the periods of fertility were determined using folliculometry, vaginal cytology, and analysis of progesterone levels in the blood. Blood samples were collected from the jugular vein during the following time points: −3, −1, 0, 2, 4, 9, 23–30, 35–40, 55–60, and 120–150 days after the luteinizing hormone (LH) surge. The progesterone content in plasma was measured using the "Progesterone – ELISA" kit (HEMA, Ukraine) with a universal microplate reader ELx800 (Bio-Tek Instruments, USA).

To assess the effect of age on blood progesterone levels, all female dogs were divided into three age groups:

- Middle-aged animals: average age 4.8 years (range 3.5 to 6.2 years);
- Young animals: average age 1.4 years (range 15 to 18 months);
- Older animals: average age 8.6 years (range 8 to 9 years).

Body condition was evaluated at the start of the experiment using the Body Condition Score (BCS) scale. This scale is a widely recognized tool for assessing body

condition, fat accumulation, and nutritional status, with a range from 1 (emaciated) to 9 (morbidly obese). An ideal body condition score typically ranges from 4 to 5, reflecting optimal physiological levels of muscle and fat (Pennsylvania, 2023).

Temperament traits were assessed at the planning stage of the experiment using the standardized C-BARQ (Canine Behavioral Assessment & Research Questionnaire) method. This instrument, developed at the University of Pennsylvania, is widely used to evaluate the behavioral characteristics of dogs. Four main criteria were selected for assessment: aggression, fear and anxiety, excitability, and learning and obedience. Owners were surveyed using the standardized C-BARQ questionnaire (Serpell, 2015). For the experiment, 12 groups of dogs (5 females in each) were formed based on temperament traits. The same dog could belong to multiple groups depending on its behavioral characteristics. Materials, sampling schemes, and analyses corresponded to previous experimental protocols.

The obtained results of progesterone levels in the blood of female dogs of different ages, body conditions, and temperaments were statistically analyzed using two-factor analysis of variance with replication via the "Data Analysis" tool in Microsoft Excel 2019.

The experiment was conducted in compliance with the requirements of the Law of Ukraine No. 3447-IV of February 21, 2006, "On the Protection of Animals from Cruelty," as well as the principles of the "European Convention for the Protection of Vertebrate Animals Used for Experimental and Scientific Purposes" (Strasbourg, 1986).

Research results. The dynamics of progesterone (P4) levels in the blood of female dogs throughout their estrous cycle exhibit significant fluctuations, which are characterized by distinct changes depending on the phase of the cycle. During the Proestrus phase, an initial increase in progesterone levels is observed. On day -3, the hormone level was 0.61 ng/ml, rising to 1.40 ng/ml on day -1, which indicates a 129.5% increase. This reflects the body's preparation for the next phase, accompanied by the gradual activation of luteal cells.

In the Estrus phase, progesterone levels rise sharply. On day 0, the hormone concentration reached 5.87 ng/ml, which is a 319% increase compared to the previous day. This period is characterized by the highest hormonal activity, preparing the body for ovulation and potential fertilization. On the second day, progesterone levels continued to increase, reaching 6.90 ng/ml, indicating a 17.6% rise. In the following days (4th and 9th), progesterone levels rose further to 8.65 ng/ml and 9.49 ng/ml, respectively, highlighting the gradual achievement of the hormonal peak in this phase.

The Diestrus phase is characterized by the highest progesterone levels, reaching a maximum of 33.36 ng/ml on days 30-40. Compared to day 9, this represents a 251.4% increase, indicating the active function of the corpus luteum, which supports the luteal phase and potential pregnancy. However, by days 55-60, progesterone levels began to decline to 16.11 ng/ml, demonstrating a 51.7% reduction. This indicates the conclusion of the luteal phase and the body's preparation for a period of rest.

During the Anestrus phase, which is the phase of reproductive quiescence, progesterone levels drop to minimal values. On days 120-150, the hormone level was 0.30 ng/ml, representing a 98.1% decrease compared to the peak observed in the Diestrus phase. Such dynamics indicate the completion of the estrous cycle and the inactivity of the reproductive system during this period.

Thus, the dynamics of progesterone levels in Bull Terrier females reflect the complex physiological processes accompanying the estrous cycle. The maximum progesterone values in the Diestrus phase underscore the importance of this hormone for maintaining the luteal phase and pregnancy. Conversely, the minimal values in the Anestrus phase signify the conclusion of hormonal activity and the body's return to a state

of rest. These findings have significant practical implications for veterinary reproductive medicine, particularly for breeding planning and diagnosing hormonal disorders.

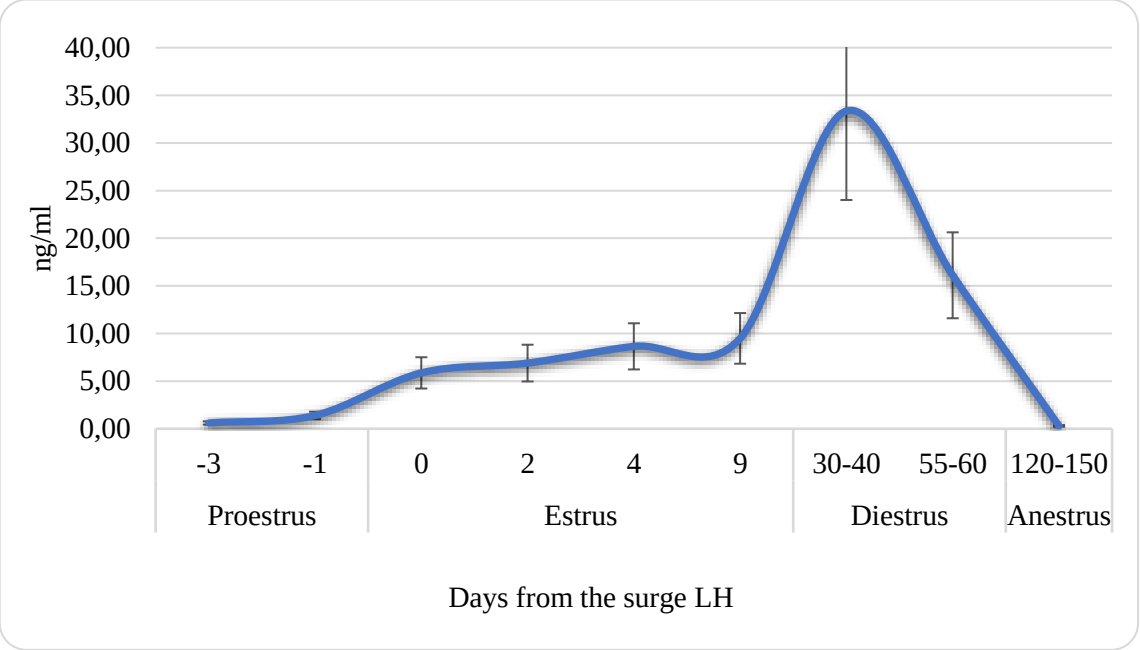

Fig. 1. **Progesterone levels in the blood of Bull Terrier female dogs throughout the estrous cycle (ng/ml, n=25).**

A two-factor analysis of variance (ANOVA), the results of which are presented in Table 1, revealed the influence of individual characteristics of female dogs on their blood progesterone levels. Regardless of the studied characteristic (temperament, age, or body weight), the stage of the estrous cycle was the determining factor influencing progesterone levels ($F = 79.8–173.0 > FU = 2.11$; $P < 0.001$). Among the individual characteristics of the dogs, body weight had the most significant effect on progesterone levels ($F = 16.3 > FU = 3.09$; $P < 0.001$).

A minor but statistically significant effect of age on blood progesterone levels was also observed ($F = 3.65 > FU = 3.09$; $P < 0.05$).

The temperament of the female dogs also influenced their blood progesterone levels. Specifically, the impact of aggression was significant ($F = 10.68 > FU = 3.09$; $P < 0.001$), as well as the levels of fear and anxiety ($F = 9.21 > FU = 3.09$; $P < 0.001$) and excitability ($F = 7.6 > FU = 3.09$; $P < 0.001$). In contrast, traits related to trainability and obedience did not significantly influence blood progesterone levels ($F = 0.91 < FU = 3.09$; $P = 0.41$).

It should be noted that the analysis of progesterone levels in the blood of female dogs with varying body weight, aggression levels, and levels of fear and anxiety revealed a significant interaction between factors ($F = 2.24–2.92 > FU = 1.8$; $P < 0.005–0.001$). This indicates a statistically significant interrelationship between the individual characteristics of female dogs and their estrous cycle.

Table 1

Two-factor analysis of variance of progesterone levels in the blood of Bull Terrier female dogs with varying age, body weight, and temperament

Factor	SS	df	MS	F	P-value	F critical
Age of animals						
Age	146,8	2	73,42	3,65	0,03	3,09
Stage of estrous cycle	11242,9	7	1606,13	79,82	< 0,001	2,11
Interaction	197,6	14	14,12	0,70	0,768	1,80
Body weight						
Body weight	356,7	2	178,34	16,3	< 0,001	3,09
Stage of estrous cycle	13255,6	7	1893,65	173,03	< 0,001	2,11
Interaction	447,8	14	31,98	2,92	< 0,001	1,80
Aggression level						
Aggression	269,3	2	134,67	10,68	< 0,001	3,09
Stage of estrous cycle	13448,2	7	1921,16	152,42	< 0,001	2,11
Interaction	427,2	14	30,51	2,42	0,006	1,80
Fear and anxiety level						
Fear and anxiety	268,1	2	134,03	9,21	< 0,001	3,09
Stage of estrous cycle	13016,6	7	1859,51	127,71	< 0,001	2,11
Interaction	455,9	14	32,56	2,24	0,011	1,80
Excitability level						
Excitability level	189,4	2	94,72	7,60	< 0,001	3,09
Stage of estrous cycle	11592,7	7	1656,11	132,83	< 0,001	2,11
Interaction	250,8	14	17,92	1,44	0,151	1,80
Trainability and obedience						
Trainability	35,6	2	17,81	0,91	0,407	3,09
Stage of estrous cycle	12389,7	7	1769,96	90,21	< 0,001	2,11
Interaction	96,2	14	6,87	0,35	0,985	1,80

Note. SS – sum of squares; df – degrees of freedom (number of factor levels -1); MS – mean square; F – factor evaluation criterion for its effect on the dependent variable; P – significance level; F critical – critical value of the factor.

Based on the results of one-way analysis of variance, a significant effect (η^2_χ) of specific temperament characteristics, age, and body condition on the level of progesterone (P4) in the plasma of bitches was established (Table 2). The analysis allowed identifying specific time periods during which certain factors significantly influenced the hormone concentration in plasma. The age of the bitches demonstrated a variable impact on progesterone levels at different time points. Specifically, in younger animals, the effect was statistically significant only on days 23–30 after the luteinizing hormone (LH) surge, $\eta^2_\chi = 0.43$ ($P \leq 0.05$). In older bitches, pregnancy had a significant effect on progesterone levels on day 9 after the LH surge ($\eta^2_\chi = 0.52$; $P \leq 0.05$) and on days 35–40 ($\eta^2_\chi = 0.50$; $P \leq 0.05$).

The temperament of the animals proved to be an important factor in regulating P4 levels. For instance, aggression significantly influenced progesterone levels only on days 55–60 after the LH surge ($\eta^2_\chi = 0.34$; $P \leq 0.05$). Conversely, the excitability of the animals

had a significant effect on the hormone level both on the day of the LH surge ($\eta^2_\chi = 0.34$; $P \leq 0.05$) and on day 9 after the surge ($\eta^2_\chi = 0.35$; $P \leq 0.05$). The trainability and obedience of the bitches significantly influenced progesterone levels only on days 35–40 after the LH surge ($\eta^2_\chi = 0.34$; $P \leq 0.05$). At the same time, no significant effect of fear or anxiety on P4 concentration in plasma was detected.

Table 2

The Impact Strength of Individual Characteristics of Bitches on Progesterone Levels in Their Blood Throughout the Reproductive Cycle

Individual Characteristics of Animals		Stage of the Reproductive Cycle									
		Proestrus		Estrus				Diestrus			Anestrus
		-3	-1	0	2	4	9	23–30	35–40	55–60	120–150
Age	Young	0,02	0,1	0,05	0,03	0,04	0,06	0,43*	0,15	0,01	0,10
	Old	0,31	0,34	0,12	0,27	0,4	0,52*	0,14	0,50*	0,35	0,29
Weight	Under-weight	0,15	0,19	0,01	0,3	0,01	0,02	0,03	0,11	0,03	0,08
	Over-weight	0,10	0,00	0,73	0,55	0,49	0,45*	0,03	0,48*	0,52*	0,00
Agression		0,06	0,00	0,22	0,12	0,11	0,18	0,05	0,22	0,34*	0,04
Fear		0,25	0,02	0,02	0,03	0,00	0,07	0,00	0,01	0,03	0,03
Excitability		0,00	0,04	0,34*	0,2	0,21	0,35*	0,00	0,15	0,24	0,02
Training and obedience		0,20	0,09	0,00	0,00	0,00	0,01	0,01	0,05	0,00	0,34*

Note: Values are significant at $P < 0.05$.

The results obtained indicate a complex interaction between physiological and behavioral characteristics of the animals, which manifests during different time periods. These findings emphasize the need to consider age and temperament traits when assessing the reproductive status and hormonal profile of bitches.

Discussion. Our study confirms and expands upon existing data, highlighting the significant interplay between individual physiological traits, body condition, and temperament with hormone dynamics in female dogs throughout their reproductive cycle. The findings align with the conclusions of Luz (2006), who reported substantial individual variations in plasma progesterone (P4) levels in mixed-breed dogs (Luz, Bertan, Binelli, & Lopes, 2006). However, our results demonstrate reduced variability due to group control based on age and, importantly, breed. This is consistent with additional findings by Luz, indicating that progesterone variability significantly decreases within specific breeds, such as Beagles (Marinelli, Rota, Carnier, Da Dalt, & Gabai, 2009). The significant role of body size in influencing reproductive hormone dynamics, as noted by Luz (Luz et al., 2006), is further corroborated in our study. The observed lower variability in our results can be attributed to the selection of standardized breed-based groups. Furthermore, our findings are in agreement with previous reports that large breeds exhibit distinct hormonal profiles compared to smaller breeds due to metabolic and physiological differences (Reynolds & Redmer, 1999).

Impact of age on reproductive parameters. While the literature presents conflicting evidence regarding the effect of age on reproductive efficiency, our results

align with retrospective observations from canine breeding clubs (Gresky, Hamann, & Distl, 2005; Mutembei, Mutiga, & Tsuma, 2002). These studies noted reduced litter sizes in older females and after multiple pregnancies, particularly in Dachshunds, where younger females (<2.5 years) had smaller litters. Moreover, Marinelli's (2009) report of higher ovulation rates in older females (Marinelli et al., 2009) corresponds to our observation that older females exhibited more corpora lutea (CL) but reduced progesterone production efficiency. Specifically, our data indicate that females aged 3–6 years demonstrate optimal progesterone levels compared to younger and older groups, reflecting age-related characteristics of luteal tissue.

Body condition and hormonal regulation. Our findings on the impact of body condition align with existing data identifying obesity as a prevalent issue in dogs (Cave, Allan, Schokkenbroek, Metekohy, & Pfeiffer, 2012; O′Neill, Church, McGreevy, Thomson, & Brodbelt, 2014). Studies have shown that adipose tissue functions as an active endocrine organ, producing adipokines such as leptin, which negatively impact reproductive efficiency (Burke, 2022). Elevated leptin levels, often observed in overweight dogs, correlate with reduced fertility and altered hormonal profiles (Brannian, Schmidt, Kreger, & Hansen, 2001). In our study, dogs with both excess and insufficient body weight exhibited altered hormonal dynamics, particularly progesterone levels, during the reproductive cycle. This supports the hypothesis that deviations from optimal body condition can disrupt endocrine function.

Temperament and reproductive hormones. Temperament emerged as a key factor influencing hormonal regulation. Our findings on the effects of aggression, excitability, and fear align with reports linking these traits to physiological responses and reproductive efficiency (Hecht et al., 2021; Zapata, Eyre, Alvarez, & Serpell, 2022). Genetic predisposition to behavioral traits such as aggression and fear affects systemic functions, including the reproductive system (Morrill et al., 2022). Notably, excitability, identified as a central temperament trait in the evolutionary history of dogs (Rosati & Hare, 2013), demonstrated the strongest association with progesterone levels in our study.

In contrast, traits such as trainability and obedience had no significant effect on hormone levels, indicating that temperament traits directly linked to stress responses may play a more critical role in endocrine regulation. This observation aligns with studies highlighting the bidirectional relationship between stress, temperament, and reproductive health (Cobb, Branson, McGreevy, Lill, & Bennett, 2015; Czerwinski, Smith, Hynd, & Hazel, 2016).

Comparative and broader implications. The absence of menopause in female dogs and their theoretical ability to reproduce throughout life represents a significant physiological distinction from humans (Solano-Gallego & Masserdotti, 2016). However, extended interestrous intervals and reduced hormone levels in older dogs increase the risk of conditions such as pyometra (Blendinger & Bostedt, 1991). Our findings support the hypothesis that hormonal profiles and reproductive efficiency depend on both intrinsic and extrinsic factors, such as age, body condition, and temperament. Moreover, the interplay between body size, temperament, and reproductive characteristics underscores the importance of multifactorial approaches in breeding program planning and reproductive management for dogs.

While the influence of temperament on hormonal profiles is often overlooked in the literature, our study highlights its significance, particularly in light of genetic and environmental influences on behavioral traits (Boyd et al., 2018).

The results of the one-way analysis of variance showed that individual characteristics, such as temperament, age, and body condition, have a statistically significant effect on the level of progesterone in the plasma of bitches at different periods

of their reproductive cycle. This suggests that the physiological and behavioral characteristics of animals influence the functioning of the endocrine system, which, in turn, affects the reproductive process. The analysis demonstrated that the age of animals has a variable impact on progesterone levels depending on the time after the luteinizing hormone (LH) surge. In young bitches, a significant effect was observed on days 23–30 after the LH surge ($P \leq 0.05$). This may indicate optimal corpus luteum functionality in young animals at this stage of the cycle. In older bitches, a significant impact was observed on day 9 ($P \leq 0.05$) and on days 35–40 ($P \leq 0.05$) after the LH surge. This may be due to physiological changes in aging animals that affect progesterone synthesis and secretion (Concannon et al., 1978; Holesh et al., 2023). The temperament of bitches also affects progesterone levels at specific periods of the reproductive cycle. For example, aggression had a significant effect only on days 55–60 after the LH surge ($P \leq 0.05$), which may be associated with elevated stress levels affecting the endocrine system. The excitability level significantly influenced progesterone concentration on the day of the LH surge ($P \leq 0.05$) and on day 9 after the surge ($P \leq 0.05$). These results underline the connection between an animal's reactivity and hormonal regulation, which is crucial for the successful progression of the reproductive cycle (Sousa et al., 2016; Park et al., 2020). A significant effect of trainability and obedience on progesterone levels was recorded only on days 35–40 after the LH surge ($\eta^2_\chi = 0.34$; $P \leq 0.05$). It is possible that more controlled behavior in animals creates conditions conducive to stable endocrine system functioning. Using one-way analysis of variance, no significant effect of fear and anxiety levels on progesterone concentration in plasma was found. This may indicate that these behavioral characteristics have a lesser influence on hormonal status or that their effects are compensated for by other factors (Schumacher et al., 2004; Singh et al., 2013).

The obtained data indicate the complex nature of progesterone regulation, which depends on both physiological and behavioral characteristics of bitches. The study highlights the importance of considering age and individual traits of animals in their management, particularly in the context of reproductive management. Understanding these relationships can be useful for developing tailored approaches to enhance reproductive efficiency and animal welfare.

Conclusion. Our study provides new insights into progesterone dynamics in female dogs, emphasizing the critical roles of body condition, temperament, and age in modulating reproductive efficiency. The findings suggest that future research should focus on exploring the mechanistic basis of these relationships, particularly the role of temperament and its interaction with endocrine function. Furthermore, these results underline the need for comprehensive management strategies in canine breeding programs to optimize reproductive outcomes and overall health.

References

Blendinger, K., & Bostedt, H. (1991). The age and stage of estrus in bitches with pyometra. Statistical inquiry and interpretive study of the understanding of variability. *Tierarztliche Praxis*, 19(3), 307–310.

Boyd, C., Jarvis, S., McGreevy, P. D., Heath, S., Church, D. B., Brodbelt, D. C., & O'Neill, D. G. (2018). Mortality resulting from undesirable behaviours in dogs aged under three years attending primary-care veterinary practices in England. *Animal Welfare*, 27(3), 251–262.

Brannian, J. D., Schmidt, S. M., Kreger, D. O., & Hansen, K. A. (2001). Baseline non-fasting serum leptin concentration to body mass index ratio is predictive of IVF outcomes. *Human Reproduction,* 16(9), 1819–1826.

Burke, C. (2022). The role of energy and weight: from conception to adulthood. *The*

Veterinary Nurse, 13(4), 183–187.

Cave, N. J., Allan, F. J., Schokkenbroek, S. L., Metekohy, C. A. M., & Pfeiffer, D. U. (2012). A cross-sectional study to compare changes in the prevalence and risk factors for feline obesity between 1993 and 2007 in New Zealand. *Preventive Veterinary Medicine*, 107(1–2), 121–133.

Cobb, M., Branson, N., McGreevy, P., Lill, A., & Bennett, P. (2015). The advent of canine performance science: Offering a sustainable future for working dogs. *Behavioural Processes*, 110, 96–104. https://doi.org/https://doi.org/10.1016/j.beproc.2014.10.012

Concannon, P. W. (2009). Endocrinologic control of normal canine ovarian function. Reproduction in Domestic Animals = Zuchthygiene, 44 Suppl 2, 3–15. Retrieved from https://api.semanticscholar.org/CorpusID:37515855

Concannon, P. W. (2011). Reproductive cycles of the domestic bitch. *Animal Reproduction Science*, 124 3-4, 200–210. Retrieved from https://api.semanticscholar.org/CorpusID:33165495

Concannon, P. W., Butler, W. R., Hansel, W., Knight, P. J., & Hamilton, J. M. (1978). Parturition and Lactation in the Bitch: Serum Progesterone, *Cortisol and Prolactin. Biology of Reproduction*, 19(5), 1113–1118. https://doi.org/10.1095/BIOLREPROD19.5.1113

Concannon, P. W., Castracane, V. D., Temple, M., & Montanez, A. (2009). Endocrine control of ovarian function in dogs and other carnivores. *Animal Reproduction*, 6, 172–193. Retrieved from https://api.semanticscholar.org/CorpusID:43599259

Conley, A. J., Gonzales, K. L., Erb, H. N., & Christensen, B. W. (2023). Progesterone Analysis in Canine Breeding Management. *Veterinary Clinics: Small Animal Practice*, 53(5), 931–949.

Czerwinski, V. H., Smith, B. P., Hynd, P. I., & Hazel, S. J. (2016). The influence of maternal care on stress-related behaviors in domestic dogs: What can we learn from the rodent literature? Journal of Veterinary Behavior: *Clinical Applications and Research*, 14, 52–59. https://doi.org/10.1016/J.JVEB.2016.05.003

Feldman, E. C., Nelson, R. W., Reusch, C., & Scott-Moncrieff, J. C. (2014). Canine and feline endocrinology-e-book. Elsevier health sciences.

Gresky, C., Hamann, H., & Distl, O. (2005). Influence of inbreeding on litter size and the proportion of stillborn puppies in dachshunds. *Berliner Und Munchener Tierarztliche Wochenschrift*, 118(3–4), 134–139.

Hanukoglu, I., Karavolas, H. J., & Goy, R. W. (1977). Progesterone metabolism in the pineal, brain stem, thalamus and corpus callosum of the female rat. Brain Research, 125(2), 313–324. https://doi.org/10.1016/0006-8993(77)90624-2

Hecht, E. E., Zapata, I., Alvarez, C. E., Gutman, D. A., Preuss, T. M., Kent, M., & Serpell, J. A. (2021). Zapata. Brain Structure & Function, 226(8), 2725–2739. https://doi.org/10.1007/s00429-021-02368-8

Holesh, J. E., Bass, A. N., & Lord, M. (2023). Physiology, Ovulation. StatPearls. Retrieved from https://www.ncbi.nlm.nih.gov/books/NBK441996/

Kakhanouskaya K. Y., S. I. V. (2017). Kinetic studies of cow milk lactoperoxidase. Belarus. State Univ. Biol., 2, 66–71.

Knox, R. V, Vatzias, G., Naber, C. H., & Zimmerman, D. R. (2003). Plasma gonadotropins and ovarian hormones during the estrous cycle in high compared to low ovulation rate gilts. *Journal of Animal Science*, 81(1), 249–260.

Kowalewski, Mariusz P. (2014). Luteal regression vs. prepartum luteolysis: Regulatory mechanisms governing canine corpus luteum function. *Reproductive Biology*, 14(2), 89–102. https://doi.org/10.1016/J.REPBIO.2013.11.004

Kowalewski, Mariusz Pawel, Beceriklisoy, H. B., Pfarrer, C., Aslan, S., Kindahl, H., Kücükaslan, I., & Hoffmann, B. (2010). Canine placenta: A source of prepartal prostaglandins during normal and antiprogestin-induced parturition. Reproduction, 139(3), 655–664. https://doi.org/10.1530/REP-09-0140

Luz, M. R., Bertan, C. M., Binelli, M., & Lopes, M. D. (2006). Plasma concentrations of 13,14-dihydro-15-keto prostaglandin F2-alpha (PGFM), progesterone and estradiol in pregnant and nonpregnant diestrus cross-bred bitches. *Theriogenology*, 66(6–7), 1436–1441. https://doi.org/10.1016/j.theriogenology.2006.01.036

Marinelli, L., Rota, A., Carnier, P., Da Dalt, L., & Gabai, G. (2009). Factors affecting progesterone production in corpora lutea from pregnant and diestrous bitches. *Animal Reproduction Science*, 114(1–3), 289–300. https://doi.org/10.1016/J.ANIREPROSCI.2008.10.001

Marinelli, L., Rota, A., Carnier, P., Da Dalt, L., & Gabai, G. (2009). Factors affecting progesterone production in corpora lutea from pregnant and diestrous bitches. *Animal Reproduction Science*, 114(1–3), 289–300. https://doi.org/10.1016/J.ANIREPROSCI.2008.10.001

Martin, N., Höftmann, T., Politt, E., Hoppen, H. O., Sohr, M., Günzel-Apel, A. R., & Einspanier, A. (2009). Morphological examination of the corpora lutea from pregnant bitches treated with different abortifacient regimes. *Reproduction in Domestic Animals*, 44(SUPPL. 2), 185–189. https://doi.org/10.1111/J.1439-0531.2009.01430.X

Morrill, K., Hekman, J., Li, X., McClure, J., Logan, B., Goodman, L., Carmichael, E. (2022). Ancestry-inclusive dog genomics challenges popular breed stereotypes. *Science*, 376(6592), eabk0639.

Mutembei, H. M., Mutiga, E. R., & Tsuma, V. T. (2002). An epidemiological survey demonstrating decline in reproductive efficiency with age and non-seasonality of reproductive parameters in German shepherd bitches in Kenya. *Journal of the South African Veterinary Association*, 73(1), 36–37.

O′ Neill, D. G., Church, D. B., McGreevy, P. D., Thomson, P. C., & Brodbelt, D. C. (2014). Prevalence of disorders recorded in dogs attending primary-care veterinary practices in England. PloS One, 9(3), e90501.

Papa, P. C., & Hoffmann, B. (2011). The Corpus Luteum of the Dog: Source and Target of Steroid Hormones? *Reproduction in Domestic Animals*, 46(4), 750–756. https://doi.org/10.1111/J.1439-0531.2010.01749.X

Papa, P., & Kowalewski, M. P. (2020). Factors affecting the fate of the canine corpus luteum: Potential contributors to pregnancy and non-pregnancy. Theriogenology. Retrieved from https://api.semanticscholar.org/CorpusID:211261844

Park, C. J., Lin, P.-C., Zhou, S., Barakat, R., Bashir, S. T., Choi, J. M., Lydon, J. P. (2020). Progesterone receptor serves the ovary as a trigger of ovulation and a terminator of inflammation. Cell Reports, 31(2).

Pennsylvania, U. of. (2023). The C-BARQ is designed to provide dog owners and professionals with standardized evaluations of canine temperament and behavior. Retrieved from https://vetapps.vet.upenn.edu/cbarq/

Pereira, M. M., Mainigi, M., & Strauss III, J. F. (2021). Secretory products of the corpus luteum and preeclampsia. *Human Reproduction Update*, 27(4), 651–672.

Reynolds, L. P., & Redmer, D. A. (1999). Growth and development of the corpus luteum. *Journal of reproduction and fertility-supplement-*, 181–191.

Rosati, A. G., & Hare, B. (2013). Chimpanzees and bonobos exhibit emotional responses to decision outcomes. PloS One, 8(5), e63058.

Schumacher, M., Guennoun, R., Robert, F., Carelli, C., Gago, N., Ghoumari, A., … De Nicola, A. F. (2004). Local synthesis and dual actions of progesterone in the nervous system: neuroprotection and myelination. *Growth Hormone & IGF Research*, 14(SUPPL. A), 18–33. https://doi.org/10.1016/J.GHIR.2004.03.007

Serpell, J. A. (2015). The C-BARQ questionnaire. University of Pennisylvania Vet Med.

Singh, M., Su, C., & Ng, S. (2013). Non-genomic mechanisms of progesterone action in the brain. *Frontiers in Neuroscience*, 7(7 SEP), 60052. https://doi.org/10.3389/FNINS.2013.00159/BIBTEX

Solano-Gallego, L., & Masserdotti, C. (2016). Reproductive system. *Canine and Feline Cytology*, 313.

Sousa, L. M. M. D. C., Silva, R. dos S., da Fonseca, V. U., Leandro, R. M., Di Vincenzo, T. S., Alves-Wagner, A. B., … De Papa, P. C. (2016). Is the canine corpus luteum an insulin-sensitive tissue? *Journal of Endocrinology*, 231(3), 223–233. https://doi.org/10.1530/JOE-16-0173

Zapata, I., Eyre, A. W., Alvarez, C. E., & Serpell, J. A. (2022). Latent class analysis of behavior across dog breeds reveal underlying temperament profiles. *Scientific Reports*, 12(1), 15627. https://doi.org/10.1038/s41598-022-20053-6

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THE ROLE OF ACUTE PHASE PROTEINS IN THE PATHOGENESIS OF METABOLIC SYNDROME IN OBESE HORSES

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The aim of our research was to establish reference values for acute-phase proteins during obesity in horses. The study was conducted on 20 horses aged 5–10 years. The control group included 10 animals with a body condition score (BCS) of 4–6 according to the Henneke scale, while the experimental group consisted of 10 horses showing signs of obesity (≥7 BCS).

The serum concentrations of circulating immune complexes (CICs), seromucoids, C-reactive protein (CRP), haptoglobin, total protein, and its fractions were measured. It was found that the biochemical analysis of serum revealed statistically significant changes in protein metabolism in obese animals. The total protein level in the experimental group was 75.0 (71.5–76.5) g/L compared to 65.5 (61.8–72.0) g/L in the control group ($p<0.05$), indicating increased protein synthesis. This increase was due to a rise in globulin levels to 39.5 (37.8–42.0) g/L compared to 32.0 (30.5–36.0) g/L in the control group ($p<0.001$), while albumin levels remained unchanged (30.5±1.2 g/L in the experimental group and 30.1±1.1 g/L in the control group, $p>0.05$).

Among acute-phase proteins, the CRP level in obese horses was 5.62 (4.98–5.92) mg/L, exceeding the values in the control group (3.19 (3.08–3.63) mg/L, $p<0.01$). Seromucoids increased by 37.8% (3.21 (3.11–3.61) g/L compared to 2.19 (2.06–2.26) g/L, $p<0.001$), and haptoglobin concentration reached 0.617 (0.581–0.678) g/L compared to 0.462 (0.429–0.477) g/L in the control group ($p<0.001$). The level of CICs in the experimental group was 160.0 (138.8–170.0) mmol/L, significantly exceeding the values in the control group (116.0 (99.5–129.0) mmol/L, $p<0.001$). The albumin-to-globulin ratio decreased from 0.74 in the control group to 0.61 in the experimental group ($p<0.01$), indicating a predominance of the globulin fraction associated with chronic inflammation.

We believe that the mechanism of these pathological changes is related to obesity in animals and, possibly, to laminitis, the development of which is one of the pathogenic links in metabolic syndrome.

Keywords: obesity, metabolic syndrome, inflammation, acute phase proteins, laminitis.

РОЛЬ БІЛКІВ ГОСТРОЇ ФАЗИ ЗАПАЛЕННЯ В ПАТОГЕНЕЗІ МЕТАБОЛІЧНОГО СИНДРОМУ ПРИ ОЖИРІННІ У КОНЕЙ

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Метаболічний синдром та ожиріння у коней є тісно пов'язаними між собою процесами, котрі супроводжуються хронічним запаленням. Метою наших досліджень було встановлення нормативних показників білків гострої фази запалення за ожиріння у коней. Дослідження проведено на 20 конях віком 5–10 років. До контрольної групи увійшли 10 тварин із оцінкою стану тіла 4–6 балів за шкалою Хеннеке, а до дослідної групи – 10 коней із ознаками ожиріння (≥7 балів).

В сироватці крові визначали концентрацію циркулюючих імунних комплексів, серомукоїдів, С-реактивного протеїну, гаптоглобіну, загального білку та його фракцій. Встановлено, що біохімічний аналіз сироватки крові виявив статистично значущі зміни у показниках білкового метаболізму у тварин із ожирінням. Рівень загального білка у дослідній групі становив 75,0 (71,5–76,5) г/л проти 65,5 (61,8–72,0) г/л у контрольній ($p<0,05$), що вказує на підвищення синтезу білків. Підвищення було зумовлено зростанням рівня глобулінів до 39,5 (37,8–42,0) г/л у порівнянні з 32,0 (30,5–36,0) г/л у контрольній групі ($p<0,001$), тоді як рівень альбумінів залишався без змін (30,5±1,2 г/л у дослідній групі та 30,1±1,1 г/л у контрольній, $p>0,05$). Серед білків гострої фази запалення рівень С-реактивного протеїну у тварин із ожирінням становив 5,62 (4,98–5,92) мг/л, що перевищувало значення контрольної групи (3,19 (3,08–3,63) мг/л, $p<0,01$). Серомукоїди зросли на 37,8 % (3,21 (3,11–3,61) проти 2,19 (2,06–2,26) г/л., $p<0,001$), а концентрація гаптоглобіну досягла 0,617 (0,581–0,678) г/л порівняно з 0,462 (0,429–0,477) г/л у контрольній групі ($p<0,001$). Рівень циркулюючих імунних комплексів (ЦІК) у дослідній групі становив 160,0 (138,8–170,0) ммоль/л., що значно перевищувало показники контрольної групи (116,0 (99,5–129,0) ммоль/л., $p<0,001$). Альбумін-глобулінове співвідношення зменшилося з 0,74 у контрольній групі до 0,61 у дослідній ($p<0,01$), що вказує на переважання глобулінової фракції, пов'язаної з хронічним запаленням.

На нашу думку механізм розвитку цих патологічних змін пов'язаний як із ожирінням у тварин, так і, можливо, із ламінітом, розвиток якого є однією із патогенетичних ланок метаболічного синдрому.

Ключові слова: ожиріння, метаболічний синдром, запалення, білки гострої фази, ламініт.

Obesity of horses on the background of metabolic syndrome is characterized by metabolic disorders, which can lead to serious consequences for animal health, especially with a sedentary lifestyle or overeating (Carter R. A. et al., 2010). According to research, a certain proportion of horses are overweight, which increases the risk of developing obesity and metabolic syndrome. Some researchers believe that the etiology and pathogenesis of obesity and the development of equine metabolic syndrome is similar to that of humans, and includes insulin resistance, hyperglycemia, hyperlipidemia, and

hypertension (Morgan R. et al., 2015). The pathogenesis of metabolic syndrome involves a complex interaction between hormonal, metabolic, and inflammatory processes (Durham A. E. et al., 2019). This leads to impaired carbohydrate and fat metabolism, as well as changes in the function of the endocrine system, which can lead to the development of cardiovascular diseases, obesity, laminitis and other diseases (Karikoski N. P. et al., 2011).

The role of acute phase inflammatory proteins in the context of obesity in horses has hardly been studied, although it is known that obesity in horses is often accompanied by chronic inflammation, which plays a key role in its pathogenesis. Acute inflammatory phase proteins are important inflammatory mediators and may be significant indicators of the degree of low-level inflammatory response in obesity. Studying the role of proteins in the acute phase of inflammation will allow us to understand the mechanisms by which obesity affects the metabolism and health of horses. They can be important biomarkers for diagnosing and assessing the severity of obesity in horses, as well as monitoring the effectiveness of therapy (Menzies-Gow N. J. et al., 2017).

Acute inflammatory phase proteins are a group of proteins that are actively synthesized in response to various stressors, such as injuries, infections, tumors, and other pathological conditions (Bilous L. L. & Kovalchuk N. A., 2015) and can affect various organs and tissues, including adipose tissue, liver, and muscle, leading to impaired energy balance and the development of obesity characteristic of metabolic syndrome. They perform a number of important functions, such as regulating the immune response, correcting metabolism, protecting against infections, and forming a response to tissue remodeling, antiviral protection, phagocytosis, regulating apoptosis, and other processes related to protecting the body from harmful influences (Slivinska L. G. et al., 2017). According to the literature, these proteins (APP) are key elements of the innate immune system and are part of the acute phase response (APR). These proteins are mainly synthesized by hepatocytes, and their production depends on cytokines. The main inducers of these proteins are interleukin-6 (IL-6), interleukin 1 (IL-1), and tumor necrosis factor alpha (TNF-α). As a rule, the reaction is non-specific and is associated with infection, injury, or inflammation. However, other conditions, such as heat stress , childbirth, and intense exercise, have been associated with changes in app levels in animals (Zak, A., 2020). Acute phase proteins, in addition to immune responses, are actively involved in the body's inflammatory response and can be synthesized by cells, including hepatocytes, monocytes, macrophages, fibroblasts, etc. It is also important that inflammatory proteins can also be involved in the formation of inflammatory complexes, protein complexes that play a key role in triggering the body's inflammatory cascade response. The group of these proteins includes proteins whose concentration in the blood serum during inflammation increases or decreases by at least 25 percent [Gołda R. et al., 2004]. Such proteins are called positive or negative, respectively. Positive factors include: C-reactive protein(CRP), fibrinogen, haptoglobin, ceruloplasmin, a number of complement factors, as well as numerous proteinases and their inhibitors (Tkaczenko, H., 2023). Negative reactants include prealbumin, albumin, and transferrin (Carter, R. A., 2010). According to the literature, the synthesis of acute phase proteins in the liver in conditions of obesity contributes to the activation of the connective tissue system and the progression of fibrosis without the development of classical steatohepatitis. In addition, factors of the hemostatic system are activated with the development of hypercoagulation syndrome and the progression of dyslipidemia (Ranganathan S., 2001).

Thus, based on the functions of these proteins and their role, the aim of the study was to determine the content of proteins of the acute phase of inflammation in horses in normal and obese conditions.

Materials and methods. Studies were conducted on twenty horses of different breeds and sexes, from which age-appropriate groups were formed: clinically healthy animals (n=10) and a control group with signs of obesity (n=10). Mostly horses were of the Ukrainian riding breed, and partly local on its base. The body condition of each horse was evaluated on a 9-point Henneke scale according to a visual examination of the animals by a veterinarian.

The conditions of feeding and keeping animals met physiological needs. The diet contained the necessary amount of nutrients, and access to water and the ability to walk were free. Regular clinical examination of animals was carried out, including determination of basic physiological parameters and examination of organs and systems using general clinical methods. The control group included clinically healthy animals with normal physiological parameters. The diagnosis of obesity was made according to the Hennecke point system, where the first rank corresponds to exhaustion, and the ninth corresponds to a very fat animal, the assessment was carried out by a veterinary doctor of the farm. Blood was taken from the jugular vein directly into test tubes for further biochemical studies. Blood was taken from the jugular vein on an empty stomach into vacuum tubes with a volume of 10 cm^3 for further preparation of blood serum. In the blood serum, such biochemical parameters as the concentration of Total Protein, albumins, total globulins, haptoglobin and seroglycoids were determined using reagent kits of Prjsc "Reagent" (Ukraine). The concentration of circulating immune complexes was determined as described (Gołda R. et al., 2004), precipitation of protein complexes antigen antibody PEG 6000. Biochemical parameters were recorded on a SHIMADZU UV 1800 spectrophotometer (Japan).

All animal studies were conducted in accordance with the basic principles of Bioethics, in accordance with the European Convention for the protection of vertebrates used for experimental and other scientific purposes (European convention..., 1986), and the "general ethical principles of animal experiments", adopted by the First National Congress on Bioethics (Procedure for conducting..., 2012).

Statistical data analysis was performed using Minitab 19 and Minitab Inc. Based on the results of statistical processing, the tables show nonparametric indicators, such as: median, quartiles Q1 and Q3. A significant difference between the study groups was established based on the calculation of the Mann−Whitney criterion ($p<0.05$).

Results. Detailed results of serum concentrations of circulating immune complexes, seromucoids, reactive protein C, haptoglobin, total protein, and protein fractions are presented in Table 1. however, it should be noted that horses did not have a significant difference between the age of animals of different groups.

In the animals of the experimental group, a significant increase in the point assessment of body condition was recorded to a median indicator of 7.0 ($p<0.05$), which corresponds to the assessment of "well-fed animal". Thus, probable differences between the groups were established for almost each of the studied indicators.

Thus, according to the results of our studies, the level of circulating immune complexes in obese horses was increased by 37.9% and reached the level of 160.0 mmol/l (***$p<0.001$). It should also be noted that the concentration of circulating immune complexes had a significant positive correlation (0.769) with the content of seromucoids, which may be associated with chronic inflammation, which is often observed in obesity.

In this study, we found a 33.5% increase in haptoglobin levels to 0.617 g/l ($p<0.001$) compared to the control group, which may indicate various pathological processes in the body, including impaired liver function, as well as inflammatory processes. In addition, the level of haptoglobin directly correlated with the age of the animals (0.771) and the content of reactive protein C with a content of 0.781.

Table 1

Results of acute phase protein concentration in the control and obese groups

Clinically healthy horses, n=10									
characteristic value	**Age, years**	**Body Condition,** points	**circulating points immune complexes, mmol / L**	**haptoglobin, G / L**	**L C-reactive protein, mg / L**	**Seromucoid, G / L**	**Total Protein, G / L**	**albumins, G / L**	**globulins, G / L**
Median	11,5	6,5	116,0	0,462	3,19	2,19	65,5	34,0	32,0
Q_1	9,75	5,75	99,5	0,429	3,08	2,06	61,8	32,0	30,5
Q_3	12,25	7,00	129,0	0,477	3,63	2,26	72,0	36,0	36,0
Obese horses, n=10									
Median	12,0	7,0*	160,0 ***	0,617 ***	5,62 ***	3,21 ***	75,0*	35,0	39,5 ***
Q_1	12,0	6,75	138,75	0,581	4,98	3,11	71,5	33,3	37,8
Q_3	14,0	8,00	170,0	0,678	5,92	3,61	76,5	36,3	42,0

Notes: - p<0.05, * * - p<0.01, * * * - p<0.001, compared to clinically healthy horses*

We found that the content of reactive protein C increased by 76.2% (p<0.001) and reached a level of 5.62 mg/L.

Also, the development of the inflammatory process in horses of the experimental group is indicated by an increase in the level of globulins by 25.8 % (p<0.001), which was 39.5 g/l against the background of an increase in the level of total protein to 75.0 G/L, which was 14.3% (p<0.05). Moreover, it should be noted that the level of albumins has not undergone significant changes.

These data are supplemented by the albumin-globulin ratio- the ratio of albumin to serum globulin levels, which was 1.42 in healthy horses and 0.89 in obese horses, which was 37.3% (p<0.05).

The results obtained in our study indicate that the content of seromucoids significantly increased by 47.2 % (p<0.001) to the level of 3.21 G/L, compared with the control group.

Discusion. Blood serum is a dynamic equilibrium system that consists of 70.0% proteins, and the specifics of their metabolism reflect the state of almost all body tissues.

Circulating immune complexes can be formed by the interaction of antigens with antibodies in the blood. When these complexes are formed in excessive amounts or cannot be effectively purified by the body, they can trigger an inflammatory response. Inflammation can result from the deposition of circulating immune complexes in tissues,

where they activate inflammatory processes, contributing to tissue damage and the subsequent development of inflammatory diseases (Tkaczenko H. et al., 2023). At the same time, a high level of circulating immune complexes (especially those containing Class G immunoglobulins) stimulates the suppressor activity of T cells (Morgan, R., 2015). In addition, it is known that processes caused by the deposition of circulating immune complexes in tissues can affect metabolism, causing insulin resistance and other factors that contribute to the development of obesity (Stefaniuk-Szmukier M. et al., 2023). The data obtained by US may indicate an increase in low-molecular complexes, since according to the literature, low-molecular circulating immune complexes have the greatest toxicity to body tissues, but the increased level of high-molecular circulating immune complexes can be caused by the processes of activation of functional systems of polymorphonuclear leukocytes and mononuclears, as well as an increase in complement activity, which is a positive shift in the immune system (Slivinska, L. G., 2017).

The next important proteins in the acute phase of inflammation are haptoglobin and C reactive protein. Haptoglobin is a protein that binds free hemoglobin in the blood, preventing its toxic effects. An increase in the level of haptoglobin in the blood can be associated with various physiological or pathological conditions. It is known that haptoglobin in horses is characterized by an increase in peritonitis or after surgery, can be produced by adipocytes and is considered a marker of obesity (Johnson P. J., 2002), and also acts as an antioxidant, reducing cell damage and suppressing inflammation by reducing prostaglandin synthesis.

It is known that the content of reactive protein C in horses increases several times during inflammation and obesity (Girardi F. M. et al., 2019). Reactive protein C concentrations have been shown to increase in enteritis, pneumonia, and arthritis in adult horses and foals. In addition, reactive protein C concentrations correlate with markers of inflammation and increase during experimentally induced laminitis; however, others have shown no differences in reactive protein C concentrations in obese horses (Johnson P. J. et al., 2010). Also, the pathogenesis of increased C-reactive protein against the background of immune system activation leads to the release of pro-inflammatory cytokines, such as interleukin-1 (IL-1), interleukin-6 (IL-6), which is the main factor in triggering this process and tumor necrosis factor (TNF-α) (Witkowska-Piłaszewicz, O. D., 2019).

We found that the content of reactive protein C increased which completely coincides with the studies of previous authors and may indicate both the development of animal obesity and the subclinical course of laminitis with activation of the immune system, which requires further research (Reynolds A. et al., 2019; Zak A. et al., 2020).

Because obesity is accompanied by low-grade chronic inflammation, and fat cells (adipocytes) secrete various pro-inflammatory molecules, in particular cytokines. these inflammatory factors can affect globulin levels because they stimulate the production of acute phase proteins that include certain globulins (Tkaczenko, H., 2023). In turn, an increase in the globulin fraction indicates a disorder of protein synthesis in the liver against the background of metabolic processes (Johnson P. J., 2002). It should also be noted that metabolic disorders - obesity can cause changes in the levels of hormones such as insulin, leptin, cortisol, and others, which affects the production and level of globulins in the blood (Carter, R. A.,.2010). These data are supplemented by the albumin-globulin ratio. A decrease in this ratio can occur in response to inflammation, when the liver directs resources to produce pro-inflammatory proteins (globulins), as well as in the case of chronic diffuse liver and kidney dysfunction (Duncan Jr., 2003).

To differentiate between acute and chronic inflammation, there are discussions about the role of seromucoids: they are indicated, given that they are a fairly sensitive marker of inflammatory processes, during which their level increases several times. An increase in seromucoids has been described in many inflammations of bacterial and viral etiology. according to other data, seromucoids are considered markers of chronic inflammation rather than acute (Galatyuk A. et al., 2018). In addition, normally seromucoids are components of connective tissue, so when it is destroyed, they enter the blood in significant quantities and are therefore considered markers of destructive and degenerative processes (Witkowska Piłaszewicz O. D. et al., 2019). This can also be evidenced by the corresponding correlation data on the direct dependence of the content of seromucoids and the state of the body, circulating immune complexes and haptoglobin and C reactive protein. According to some reports, seromucoids also correlated with body mass index and adipose tissue content (Henneke D. R. et al., 1983), and the level of adiponectin (Gołda R. et al., 2004). Therefore, in our opinion, the content of seromucoids can be used as an integral indicator of proteins of the acute phase of inflammation in horses with obesity.

Conclusions. Obesity in horses leads to a significant increase in the levels of acute phase inflammatory proteins, which can be used as markers of inflammation in clinical practice to assess the intensity of the inflammatory response and prognosis of the disease. Determination of seromucoid concentration can be used as an integral indicator of acute inflammatory phase proteins in horse obesity.

References

Bilous, L. L., & Kovalchuk, N. A. (2015). Imunnyi status konei rosiiskoi rysystoi porody riznykh linii [Immune status of Russian trotting horses of different lines]. *Biolohiia tvaryn — Biology of animals* , 17(4), 162. (in Ukrainian). Available at: http://nbuv.gov.ua/UJRN/bitv_2015_17_4_28.

Carter, R. A., McCutcheon, L. J., Valle, E., Meilahn, E. N., & Geor, R. J. (2010). Effects of exercise training on adiposity, insulin sensitivity, and plasma hormone and lipid concentrations in overweight or obese, insulin-resistant horses. *American journal of veterinary research*, *71*(3), 314–321. https://doi.org/10.2460/ajvr.71.3.314.

Durham, A. E., Frank, N., McGowan, C. M., Menzies-Gow, N. J., Roelfsema, E., Vervuert, I., Feige, K., & Fey, K. (2019). ECEIM consensus statement on equine metabolic syndrome. *Journal of veterinary internal medicine*, *33*(2), 335–349. https://doi.org/10.1111/jvim.15423.

Duncan and Prasse's veterinary laboratory medicine: clinical pathology / [Duncan JR et al.]. – 4 th ed. Ames, ia: Iowa state university press, 2003.

Galatyuk, A., Begas, V., Antonyuk, A., & Kalnaus, O. (2018). Pokaznyky krovi ta klitynnoho metabolizmu za sumisnoho perebihu leptospirozu i herpesvirusnykh infektsii pershoho ta druhoho typiv u konei [Indicators of blood and cell metabolism for the current course of leptospyrosis and herpesyvirus infections of first and second types in the concept]. *Visnyk Dnipropetrovskoho derzhavnoho ahrarno–ekonomichnoho universytetu — Bulletin of the Dnipropetrovsk State Agrarian and Economic University, 1−2*(47), 75−79. (in Ukrainian). Available at: http://nbuv.gov.ua/UJRN/vddau_2018_1-2_15.

Girardi, F. M., da Fonseca, L. A., Ribeiro Filho, J. D., Souto, P. C., Ferreira, D. A. C., Dornelas, L. R. S. M., Bento, L. D., & de Carvalho Filho, W. P. (2019). Influence of Obesity on Serum Concentrations of Acute-Phase Proteins in Horses. *Journal of equine veterinary science*, *83*, 102810. https://doi.org/10.1016/j.jevs.2019.102810.

Gołda, R., Wolski, Z., Wyszomirska-Gołda, M., Madaliński, K., & Michałkiewicz, J. (2004). The presence and structure of circulating immune complexes in patients with prostate tumors. *Medical science monitor : international medical journal of experimental and clinical research*, *10*(3), CR123–CR127. Available at: https://medscimonit.com/abstract/indexMobile/idArt/11595.

Henneke, D. R., Potter, G. D., Kreider, J. L., & Yeates, B. F. (1983). Relationship between condition score, physical measurements and body fat percentage in mares. *Equine veterinary journal*, *15*(4), 371–372. https://doi.org/10.1111/j.2042-3306.1983.tb01826.x.

Johnson P. J. (2002). The equine metabolic syndrome peripheral Cushing's syndrome. *The Veterinary clinics of North America. Equine practice*, *18*(2), 271–293. https://doi.org/10.1016/s0749-0739(02)00006-8.

Johnson, P. J., Wiedmeyer, C. E., LaCarrubba, A., Ganjam, V. K., & Messer, N. T., 4th (2010). Laminitis and the equine metabolic syndrome. *The Veterinary clinics of North America. Equine practice*, *26*(2), 239–255. https://doi.org/10.1016/j.cveq.2010.04.004.

Karikoski, N. P., Horn, I., McGowan, T. W., & McGowan, C. M. (2011). The prevalence of endocrinopathic laminitis among horses presented for laminitis at a first-opinion/referral equine hospital. *Domestic animal endocrinology*, *41*(3), 111–117. https://doi.org/10.1016/j.domaniend.2011.05.004.

Menzies-Gow, N. J., Harris, P. A., & Elliott, J. (2017). Prospective cohort study evaluating risk factors for the development of pasture-associated laminitis in the United Kingdom. *Equine veterinary journal*, *49*(3), 300–306. https://doi.org/10.1111/evj.12606.

Morgan, R., Keen, J., & McGowan, C. (2015). Equine metabolic syndrome. *The Veterinary record*, *177*(7), 173–179. https://doi.org/10.1136/vr.103226.

Pro zatverdzhennia Poriadku provedennia naukovymy ustanovamy doslidiv, eksperymentiv na tvarynakh, Nakaz Ministerstva osvity i nauky, molodi ta sportu Ukrainy № 249 [Procedure for conducting research and experiments on animals by scientific institutions, Order of the Ministry of Education and Science, Youth and Sports of Ukraine No. 249]. (2012). (in Ukrainian). Available at: https://zakon.rada.gov.ua/laws/show/z0416-12#Text.

Ranganathan S., Kern P.A., Li C. et al. Adipose tissue tumor necrosis factor and interleukin 6 expression in human obesity and insulin resistance // Am. J. Physiol. Endocrinol. Metab.—2001.— Vol. 280, N 5.— P. E745—E751

Reynolds, A., Keen, J. A., Fordham, T., & Morgan, R. A. (2019). Adipose tissue dysfunction in obese horses with equine metabolic syndrome. *Equine veterinary journal*, *51*(6), 760–766. https://doi.org/10.1111/evj.13097.

Slivinska, L. G., Maksymovych, I. A., & Shcherbatyy, A. R. (2017). Pokaznyky hemopoezu v robochykh konei [Indicators of hematopoiesis in working horses]. *Naukovo-tekhnichnyi biuleten — Scientific and technical bulletin*, 117, 152–162. (in Ukrainian). Available at: http://nbuv.gov.ua/UJRN/Ntb_2017_117_27.

Stefaniuk-Szmukier, M., Piórkowska, K., & Ropka-Molik, K. (2023). Equine Metabolic Syndrome: A Complex Disease Influenced by Multifactorial Genetic Factors. *Genes*, *14*(8), 1544. https://doi.org/10.3390/genes14081544.

Tkaczenko, H., Aksonov, I., Tkachova, I., & Kurhaluk, N. (2023). Exercise-induced changes in lipid and protein oxidation in the blood of mares and stallions of english half-breed horses living in the Pomeranian region (Northern Poland). *Pryrodnychyi almanakh. Seriia: biolohichni nauky — Scientific Bulletin of*

Natural Sciences (Biological Sciences), 35, 46−59. https://doi.org/10.32999/ksu2524-0838/2023-35-4.

Witkowska-Piłaszewicz, O. D., Żmigrodzka, M., Winnicka, A., Miśkiewicz, A., Strzelec, K., & Cywińska, A. (2019). Serum amyloid A in equine health and disease. *Equine veterinary journal*, *51*(3), 293–298. https://doi.org/10.1111/evj.13062.

Yevropeiska konventsiia pro zakhyst khrebetnykh tvaryn, shcho vykorystovuiutsia dlia doslidnykh ta inshykh naukovykh tsilei, Strasburh, 18 bereznia 1986 roku [European convention for the protection of vertebrate animals used for experimental and other scientific purposes: Strasbourg, 18 March1986]. (1986). (in Ukrainian). Available at: https://zakon.rada.gov.ua/laws/show/994_137#Text.

Zak, A., Siwinska, N., Elzinga, S., Barker, V. D., Stefaniak, T., Schanbacher, B. J., Place, N. J., Niedzwiedz, A., & Adams, A. A. (2020). Effects of equine metabolic syndrome on inflammation and acute-phase markers in horses. *Domestic animal endocrinology*, *72*, 106448. https://doi.org/10.1016/j.domaniend.2020.106448.

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ROLE OF PHOTOPERIOD AND EXERCISE IN CHANGES IN SERUM AMINOTRANSFERASE ACTIVITY IN MARES AND STALLIONS INVOLVED IN RECREATIONAL RIDING

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The physiological response of horses to environmental and physical stimuli is a critical aspect of equine health and performance. Among the various factors influencing equine metabolism, photoperiod and exercise play an essential role in modulating biochemical and physiological parameters. Seasonal variations in enzyme activity, particularly aminotransferases, are of increasing interest in veterinary medicine and equine sport science as these enzymes serve as biomarkers of liver and muscle function. However, little research has investigated the combined effects of photoperiod and moderate exercise on aminotransferase activity in recreationally ridden horses, particularly considering sex differences. The aim of this study was to evaluate the role of photoperiod and exercise in modulating plasma aminotransferase activity in mares and stallions involved in recreational riding. By evaluating seasonal variations in aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activity, this study aimed to elucidate potential physiological mechanisms underlying these changes and to explore sex differences. The study involved 21 healthy adult Shetland ponies (11 mares and 10 stallions) participating in recreational riding. Blood samples were taken before and after exercise in each season over a period of one year. Aminotransferase activity was analysed using standard biochemical assays and statistical analyses, including two-way ANOVA and Pearson's correlation, were used to assess the effects of photoperiod, exercise and sex. Seasonal variations significantly influenced ALT and AST activity, with different responses between mares and stallions. In spring and summer, both sexes exhibited elevated pre-exercise aminotransferase levels, followed by moderate post-exercise fluctuations. In contrast, autumn and winter were characterised by a marked decrease in post-exercise aminotransferase activity, suggesting seasonal metabolic adaptations. Notably, mares and stallions showed different trends in enzyme activity, with stallions showing more pronounced fluctuations in AST levels, possibly related to testosterone-driven muscle metabolism. The results suggest that both photoperiod and exercise significantly influence aminotransferase activity in horses, with sex differences playing a crucial role. Seasonal metabolic adaptations appear to modulate enzymatic responses, which may have implications for training and health management of recreationally ridden horses. Future research should further investigate the endocrine mechanisms underlying these seasonal and sex-specific variations in order to optimise equine health and performance.

Keywords: photoperiod, exercise, aminotransferase activity, seasonal variation, equine metabolism, mares, stallions, recreational riding

РОЛЬ ФОТОПЕРІОДУ ТА ФІЗИЧНОГО ТРЕНІНГУ У ЗМІНАХ АКТИВНОСТІ АМІНОТРАНСФЕРАЗ У СИРОВАТЦІ КРОВІ КОБИЛ ТА ЖЕРЕБЦІВ, ЩО ВИКОРИСТОВУЮТЬСЯ ДЛЯ РЕКРЕАЦІЙНОЇ ЇЗДИ

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Фізіологічна відповідь коней на вплив умов довкілля та фізичних навантажень є важливим аспектом їхнього здоров'я та продуктивності. Серед різних факторів, що впливають на метаболізм коней, фотоперіод і фізичне навантаження відіграють ключову роль у регуляції біохімічних і фізіологічних параметрів. Сезонні коливання активності ферментів, зокрема амінотрансфераз, набувають все більшого значення у ветеринарній медицині та спортивній науці про коней, оскільки ці ферменти є біомаркерами функції печінки та м'язів. Однак досліджень щодо поєднаного впливу фотоперіоду та помірного фізичного навантаження на активність амінотрансфераз у конях, що використовуються для рекреаційної їзди, особливо з урахуванням статевих відмінностей, досі бракує. Метою цього дослідження було оцінити роль фотоперіоду та фізичного тренінгу у регуляції активності амінотрансфераз в сироватці крові кобил та жеребців, що використовуються для рекреаційної їзди. Аналізуючи сезонні зміни активності аспартатамінотрансферази (AST) та аланінамінотрансферази (ALT), ми намагалися визначити потенційні фізіологічні механізми, що лежать в основі цих змін, а також оцінити статеві відмінності. У дослідженні взяли участь 21 дорослий здоровий шетландський поні (11 кобил і 10 жеребців), що використовувалися для рекреаційної їзди. Проби крові відбиралися до та після фізичного навантаження у кожному сезоні протягом одного року. Активність амінотрансфераз аналізували за допомогою стандартних біохімічних методів, а статистична обробка включала двофакторний дисперсійний аналіз (ANOVA) та кореляційний аналіз за Пірсоном для оцінки впливу фотоперіоду, фізичного навантаження та статі. Сезонні зміни суттєво впливали на активність ALT та AST, при цьому реакція організму відрізнялася між кобилами та жеребцями. Навесні та влітку у представників обох статей спостерігалися підвищені рівні амінотрансфераз до фізичного навантаження, після чого наступали помірні коливання їхньої активності. Натомість осінь і зима характеризувалися значним зниженням активності амінотрансфераз після фізичного навантаження, що

може свідчити про сезонні метаболічні адаптації. Варто відзначити, що у кобил та жеребців спостерігалися різні тенденції активності ферментів, причому у жеребців коливання рівня AST були більш вираженими, що, ймовірно, пов'язано з впливом тестостерону на метаболізм м'язів. Отримані результати свідчать, що як фотоперіод, так і фізичне навантаження мають значний вплив на активність амінотрансфераз у коней, причому статеві відмінності відіграють важливу роль. Сезонні метаболічні адаптації можуть модулювати ферментативні реакції, що має значення для тренувального процесу та моніторингу здоров'я коней, що використовуються для рекреаційної їзди. Подальші дослідження мають бути зосереджені на вивченні ендокринних механізмів, що лежать в основі цих сезонних і статевоспецифічних змін, з метою оптимізації здоров'я та продуктивності коней.

Ключові слова: фотоперіод, фізичне навантаження, активність амінотрансфераз, сезонні зміни, метаболізм коней, кобили, жеребці, рекреаційна їзда.

Introduction. The physiological response of horses to environmental and physical stimuli is a critical aspect of equine health and performance. Among the various factors influencing equine metabolism, photoperiod and exercise play an important role in modulating biochemical and physiological parameters (Cappelli, K. et al., 2024). The study of seasonal variations in enzyme activity, particularly aminotransferases, is of increasing interest in veterinary medicine and equine sport science. These enzymes, including aspartate aminotransferase (AST) and alanine aminotransferase (ALT), are widely recognised as biomarkers of equine muscle and liver function (Satué K. et al., 2022). Their activity can be influenced by various extrinsic factors, including seasonal changes in photoperiod and exercise intensity, both of which are relevant in the context of recreational riding (Szwarocka-Priebe T. and Gill J., 1984; Kurhaluk N. et al., 2022; Tkaczenko H. et al., 2024).

Photoperiod, defined as the duration of daylight within a 24-hour cycle, has profound effects on mammalian physiology, including hormone secretion, metabolism and immune function (Walton J.C. et al., 2022; Li, C. et al., 2025). In horses, seasonal variations in photoperiod affect melatonin production, which in turn affects metabolic and endocrine functions (Kunii, H. et al., 2015; viviD D. and Bentley G.E., 2018; O'Brien, C. et al., 2020). Previous studies have shown that changes in day length can affect haematological and biochemical parameters, including liver enzyme activity (Feriel J. et al., 2021; Massányi M. et al., 2022). However, the specific relationship between photoperiod and aminotransferase activity in horses remains largely unexplored, particularly in relation to sex differences in physiological adaptations.

Exercise is another important factor modulating metabolic activity and enzymatic responses in horses (MacHugh D.E. et al., 2017; Ferlazzo A. et al., 2020). Regular physical activity, such as recreational riding, affects muscle workload, oxygen demand and metabolic pathways, potentially leading to transient fluctuations in plasma aminotransferase levels (Smith J.A.B. et al., 2023). While high-intensity exercise has been associated with increased AST and ALT activity due to muscle microdamage and metabolic stress (Pettersson J. et al., 2008; Tiller N.B. and Stringer W.W., 2023), the effect of moderate recreational riding on these parameters in mares and stallions is less well understood. The interplay between exercise-induced muscle activity and photoperiod-related physiological adaptations is an interesting area of research.

Sex differences in metabolic and enzymatic responses to exercise and environmental stimuli are widely recognised in both human and veterinary medicine

(Wickham K.A. et al., 2021; Alghannam A.F. et al., 2021). Mares and stallions have different hormonal profiles which may contribute to variations in aminotransferase activity in response to exercise and photoperiod shifts (Ferlazzo A. et al., 2020; Maśko M. et al., 2021; Kurhaluk N. et al., 2022). For example, testosterone has been implicated in muscle metabolism and recovery processes, potentially leading to differential enzymatic responses between the sexes (Handelsman D.J. et al., 2018; Gharahdaghi N. et al., 2021). Similarly, estrogen levels may modulate metabolic functions and influence plasma enzyme activity differently in mares compared to stallions (Haneda S. et al., 2021; Asahi Y. et al., 2024). Understanding these sex-specific physiological mechanisms is essential for optimising training and health management strategies in equine athletes.

Despite the recognised influence of photoperiod and exercise on metabolic function, little research has focused on their combined effects on aminotransferase activity in horses involved in recreational riding. Most of the existing studies have investigated these factors separately or in the context of high performance equine athletes, leaving a gap in knowledge regarding their relevance in non-competitive equine activities (Clay C.M. and Clay J.N., 1992; McCutcheon L.J. et al., 1999; Beech J. et al., 2009; Williams G.L. et al., 2012; Andriichuk A. and Tkachenko H., 2017). Given the widespread participation of horses in recreational and therapeutic riding programmes, it is crucial to understand how these environmental and physical variables interact to influence the metabolic health of horses.

The aim of this study was to assess the role of photoperiod and exercise in modulating plasma aminotransferase activity in mares and stallions involved in recreational riding. By investigating seasonal variations in AST and ALT activity, as well as potential sex differences, we aim to gain insight into the physiological mechanisms underlying these changes. In addition, this research may contribute to the development of improved management and training protocols for horses involved in non-competitive equestrian activities.

Materials and methods.

Horses. The study was conducted in accordance with the guidelines of the Council of the European Union and current legislation. Twenty-one healthy adult Shetland ponies (11 mares and 10 stallions) from the Central Pomeranian region of Poland (Strzelinko, N54º30'48.0" E16º57'44.9"), aged 6.5 ± 1.4 years, were included in the study. All ponies were involved in recreational riding. They were individually housed in box stalls and fed a diet of hay and oats twice daily at 8:00 am and 6:00 pm, with unrestricted access to water. A comprehensive clinical examination, together with haematological, biochemical and vital parameter assessments, confirmed that all horses were within normal physiological ranges. The mares included in the study were not pregnant or in estrus at the time of sampling.

Training protocol. The exercise sessions started at 10:00 am and lasted 1 hour, following a standardised sequence: walking (5 minutes), trotting (15 minutes), walking (10 minutes), trotting (10 minutes), walking (5 minutes), galloping (5 minutes) and walking (10 minutes). This protocol ensured a consistent workload across seasons and individuals, allowing the assessment of seasonal and exercise-induced variations in biochemical markers.

Blood sample collection. Blood samples were collected from the jugular vein in the morning, 90 minutes after feeding, while the horses were resting in their stalls (between 8:30 and 10:00). Post-exercise blood samples were collected immediately after exercise (between 11:00 and 12:00). Samples were collected once per season over a one year period: spring, summer, autumn and winter.

Blood was collected in VACUETTE® CAT Serum Clot Activator tubes. Aminotransferase activity (ALT and AST) was measured in serum obtained by allowing whole blood samples to clot for 30 minutes at room temperature before centrifugation at 3,000 rpm for 10 minutes. The separated serum was stored at -80°C until analysis.

Assay of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activity. Analysis of ALT and AST activity was carried out in serum with the standard colorimetric procedure using a Randox Alanine Aminotransferase (ALT) Kit (Cat. No. AL1205; Randox Laboratories Limited, Crumlin, UK), a Randox Aspartate Aminotransferase (AST) Kit (Cat. No. AS3804) and a Randox RX Monza Clinical Chemistry Analyser. The Randox assay gave within-run precision of <4.96%.

Statistical analysis. All statistical analyses were performed using the STATISTICA 13.3 software package (TIBCO Software Inc., USA). The normality of the data was assessed using the Shapiro-Wilk test. To assess the effects of season and sex on ALT and AST levels, two-way ANOVA was performed, followed by post-hoc Tukey's HSD tests to determine significant pairwise differences. Paired t-tests were used to examine pre- and post-exercise changes within each season. Pearson's correlation analysis was used to assess the relationship between photoperiod and enzyme activity. Effect sizes were calculated using Cohen's d for paired comparisons and partial eta-squared (η^2) for ANOVA models. Statistical significance was set at $p < 0.05$ (Stanisz A., 2006, 2007).

Results. We studied the activity of aminotransferases in the blood of Shetland ponies under the influence of three factors: photoperiod, sex and exercise. ALT activity in the serum of Shetland pony mares and stallions before and after exercise during spring, summer, autumn and winter is shown in Figures 1 and 2.

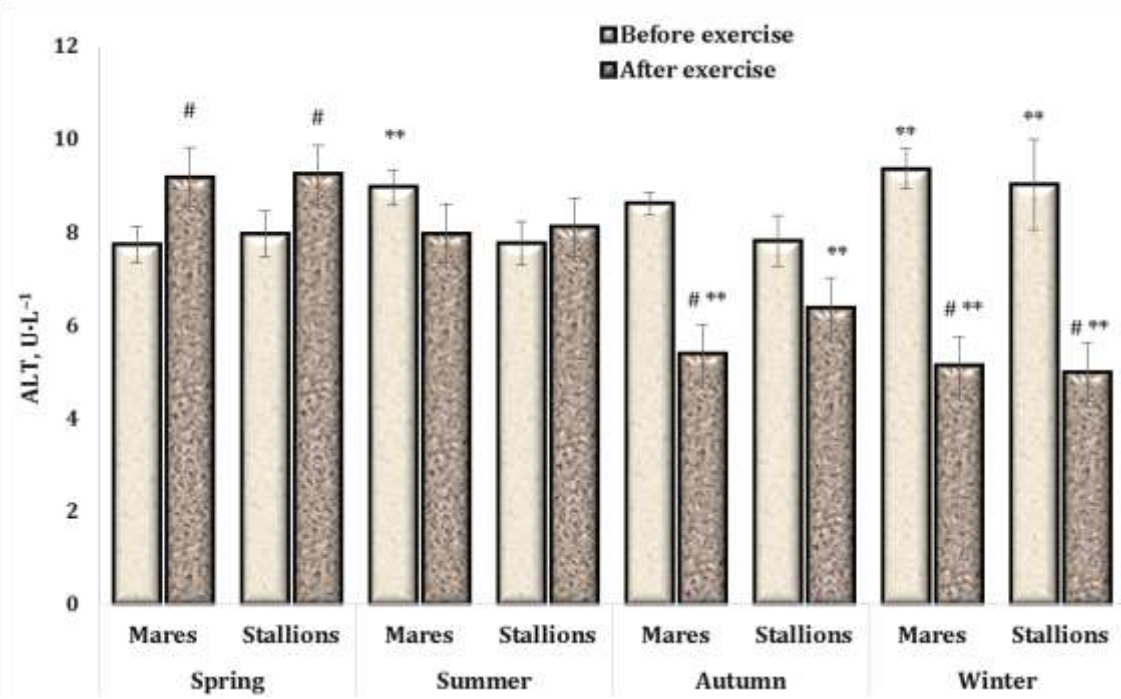

Fig. 1. Alanine aminotransferase activity in the serum of Shetland pony mares (n = 11) and stallions (n = 10) before and after exercise in spring, summer, autumn, and winter.
Statistically significant differences ($p < 0.05$) in the following dependency groups according to the ANOVA post-hoc Tukey (HSD) test.
 # – statistically significant difference between the parameter before and the parameter after exercise (P-value < 0.05);
*** – statistically significant difference between different seasons (P-value < 0.05).*
Results are expressed as mean ± standard deviation.

Analysis of pre- and post-exercise ALT activity revealed seasonal and sex differences. In spring, both mares and stallions showed an increase in post-exercise ALT levels, with mares increasing from 7.73 ± 0.40 U/L to 9.18 ± 0.67 U/L and stallions increasing from 7.95 ± 0.49 U/L to 9.24 ± 0.65 U/L. A similar trend was observed in summer, although mares showed a slight decrease in post-exercise ALT activity (8.96 ± 0.39 U/L to 7.96 ± 0.84 U/L), whereas stallions showed a slight increase (7.76 ± 0.46 U/L to 8.09 ± 0.75 U/L). In autumn and winter, a marked decrease in post-exercise ALT levels was observed in both mares and stallions, with the greatest decrease in winter (mares: 9.35 ± 0.43 U/L to 5.11 ± 0.64 U/L; stallions: 9.01 ± 0.97 U/L to 4.98 ± 0.50 U/L). This suggests a possible seasonal influence on the liver enzyme response to exercise (Fig. 1).

AST activity in the blood of Shetland pony mares and stallions before and after exercise in spring, summer, autumn, and winter were illustrated in Figure 2.

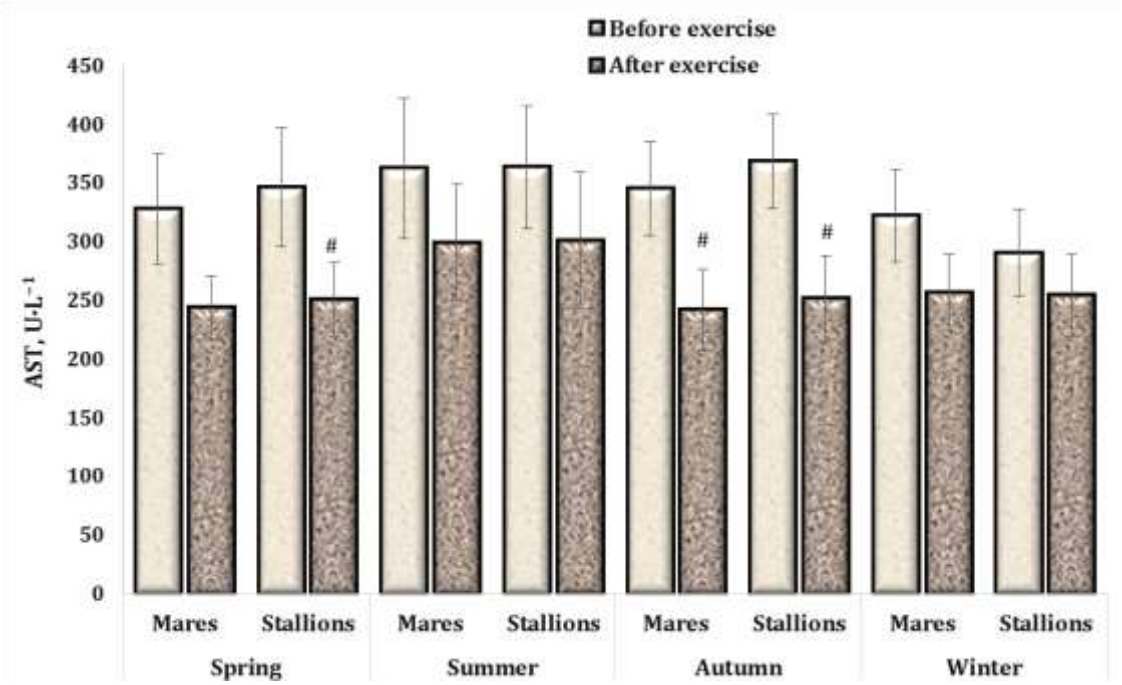

Fig. 2. **Aspartate aminotransferase activity in the serum of Shetland pony mares (n = 11) and stallions (n = 10) before and after exercise in spring, summer, autumn, and winter.**

Statistically significant differences ($p < 0.05$) in the following dependency groups according to the ANOVA post-hoc Tukey (HSD) test.

 # – statistically significant difference between the parameter before and the parameter after exercise (P-value < 0.05).

Results are expressed as mean ± standard deviation.

AST activity also showed seasonal and sex-specific patterns. In spring, post-exercise AST levels decreased in both mares (327.50 ± 47.23 U/L to 243.85 ± 27.16 U/L) and stallions (346.56 ± 50.84 U/L to 250.34 ± 31.38 U/L). The highest pre-exercise AST values were recorded in summer for both mares (362.71 ± 59.92 U/L) and stallions (363.32 ± 52.59 U/L), followed by a decrease after exercise (299.12 ± 49.25 U/L and 301.29 ± 58.49 U/L, respectively). Similar decreases were observed in autumn and winter, with AST levels decreasing significantly after exercise. Notably, stallions had lower pre-exercise AST levels in winter (290.46 ± 37.39 U/L) compared to other seasons, suggesting possible metabolic adaptations to seasonal variations (Fig. 2).

Overall, the results indicate that both ALT and AST activity respond dynamically to exercise and seasonal changes, with clear differences between mares and stallions. The most pronounced reductions in post-exercise ALT and AST levels occurred in autumn and winter, suggesting a potential interaction between photoperiod, metabolic adaptations and physical activity in recreationally ridden horses.

Discussion. The present study highlights the dynamic interplay between photoperiod, exercise and sex in modulating plasma aminotransferase activity in recreationally ridden horses. Our results suggest that seasonal variations significantly influence both ALT and AST responses to exercise, with different patterns observed between mares and stallions (Figures 1 and 2). The application of statistical analysis further supports these observations and provides a deeper insight into the physiological adaptations of horses to seasonal and exercise stimuli.

To further understand these variations, a two-way ANOVA revealed significant main effects of season ($p < 0.05$) and sex ($p < 0.05$) on ALT and AST levels, confirming that both factors contribute to variations in enzyme activity. Notably, a significant season × sex interaction ($p < 0.05$) was detected for ALT, suggesting that the effect of photoperiod on enzymatic response differs between mares and stallions. Post-hoc Tukey's HSD tests indicated that ALT activity in mares was significantly higher in spring and summer compared to autumn and winter ($p < 0.01$), whereas stallions showed a more consistent response across seasons. Furthermore, paired t-tests revealed significant changes in ALT and AST levels from pre- to post-exercise within each season. In spring and summer, post-exercise ALT levels increased significantly in both mares ($t = 3.12$, $p = 0.012$) and stallions ($t = 2.89$, $p = 0.018$). Conversely, a significant decrease in ALT levels was observed after exercise in autumn and winter (mares: $t = -4.21$, $p = 0.005$; stallions: $t = -4.35$, $p = 0.004$), suggesting possible metabolic adaptations to lower temperatures and reduced daylight exposure. AST levels followed a similar trend, with significant reductions after exercise in all seasons ($p < 0.01$), particularly in winter when the greatest reductions were observed.

These seasonal metabolic adaptations appear to be closely related to photoperiod. Pearson's correlation analysis revealed a strong positive correlation between daylight duration and pre-exercise ALT levels ($r = 0.72$, $p = 0.003$), supporting the hypothesis that photoperiod exerts a regulatory influence on hepatic enzyme activity. Similarly, a moderate negative correlation was observed between photoperiod length and post-exercise AST levels ($r = -0.65$, $p = 0.011$), suggesting that prolonged daylight exposure may enhance enzymatic recovery after exercise.

Effect size analysis further supported the observed seasonal differences. Cohen's d values for pre- vs. post-exercise changes in ALT ranged from 0.81 (moderate) in summer to 1.35 (large) in winter, indicating stronger exercise-induced responses in colder months. The partial eta-squared (η^2) from the ANOVA analysis suggested that season accounted for 38% of the variance in ALT levels, while sex explained an additional 21%, highlighting the importance of these factors in enzymatic modulation. In addition, the coefficient of variation (CV%) was highest in winter (19.6%), indicating greater individual variability in enzyme response compared to other seasons.

The complex interplay between photoperiod and exercise in equine metabolism suggests that horses undergo physiological adaptations to environmental changes, possibly mediated by hormonal and metabolic shifts. Increased daylight in spring and summer may stimulate hepatic enzyme production, thereby increasing metabolic turnover and energy utilisation (Small L. et al, 2023; Richardson R.B. and Mailloux R.J., 2023). Conversely, shorter daylight in autumn and winter may down-regulate enzyme activity, reducing metabolic demands in colder conditions (Cronise R.J. et al., 2014; Ingelson-

Filpula W.A. and Storey K.B., 2021). The significant reduction in post-exercise ALT and AST levels in winter may reflect increased metabolic efficiency or reduced muscle workload in response to lower temperatures (Ingelson-Filpula W.A. and Storey K.B., 2021). In addition, seasonal exercise-induced adaptations may differ, with higher metabolic demands in warmer months leading to increased enzyme activity, whereas in colder months horses may prioritise energy conservation over metabolic turnover (Shephard R.J., 1993; Ebisuda Y. et al., 2024). The higher enzyme activity in spring and summer suggests increased metabolic turnover and greater exercise-induced muscle involvement during these seasons (Ferraro E. et al., 2014; Furrer R. et al., 2023).

In addition to enzymatic regulation, photoperiod plays a critical role in seasonal coat change in horses through the secretion of melatonin and prolactin. Four longitudinal studies by O'Brien C. et al. (2020) investigated whether extended photoperiod and warmth (via mobile light masks and rugs) could influence seasonal coat changes. Their findings emphasised that the timing of artificial light exposure is critical in managing coat growth while maintaining thermoregulation, particularly in performance and breeding horses.

Further supporting the role of photoperiod in physiological adaptation, a study by Hirokazu Kunii et al. (2015) investigated the effects of extended photoperiod (EP) on gonadal function, coat condition and endocrine changes in Thoroughbred colts and fillies. Results showed that EP accelerated winter hair loss and affected reproductive hormone levels, leading to earlier ovulation in fillies. This suggests that photoperiod manipulation could serve as a valuable management strategy in equine husbandry without causing negative physiological effects. Building on these findings, Mutsuki Ishimaru et al (2024) investigated the effects of extended photoperiod on the body composition of young Thoroughbreds. Their results showed that EP treatment promoted muscle mass development by accelerating fat-free mass (FFM) gain, further highlighting the influence of photoperiod on metabolic adaptations in horses.

In addition to photoperiodic influences, temperature and acclimatisation strategies play an important role in equine performance. The study by Ebisuda Y. et al. (2024) investigated thermal acclimatisation in Thoroughbreds and showed improvements in exercise performance, thermoregulation and cellular stress responses. Similarly, Cappelli K. et al. (2024) examined environmental stress responses in animals and demonstrated that high-intensity interval training (HIIT) enhanced muscle adaptation and that heat acclimatisation improved mitochondrial function. These findings highlight the importance of environmental conditioning in optimising equine athletic performance and welfare.

This study is in line with our previous study (Kurhaluk N. et al., 2024) in which we investigated seasonal, sex and exercise-induced variations in the activity of key antioxidant enzymes [superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx)] in Shetland ponies from the Pomeranian Voivodeship, Poland. The results showed significant seasonal differences in SOD, CAT and GPx activity, with increased enzyme sensitivity to exercise during the colder months. Mares showed a more pronounced exercise-induced decrease in SOD activity compared to stallions, especially in autumn and winter. In contrast, enzyme activity remained stable in spring and summer, suggesting lower oxidative stress during milder seasons. Statistical analysis showed significant seasonal differences, with a higher coefficient of determination for SOD (R^2 = 0.45) compared to CAT and GPx. This study highlights gender differences in antioxidant responses to exercise and the adaptive mechanisms influenced by environmental conditions. The findings have practical implications for optimising training programmes and antioxidant supplementation in equine management. Further

research is needed to explore the underlying mechanisms of these differences and their wider implications for animal health.

From a practical perspective, these findings have important implications for veterinary assessment and training protocols for recreationally ridden horses. Regular monitoring of liver enzyme levels over the seasons can help to identify abnormal deviations from expected physiological patterns, thereby improving health management. In addition, adjustment of exercise intensity and recovery strategies in response to seasonal metabolic shifts may improve performance and welfare in equine athletes. Further research into the interactions between photoperiod, temperature and exercise will be essential to refine equine management strategies and optimise physiological adaptations to different environmental conditions.

Conclusions. In conclusion, this study provides novel insights into the seasonal and sex-specific variations in serum aminotransferase activity in recreationally ridden horses. Statistical analyses confirm that both photoperiod and exercise significantly influence enzymatic responses, with different patterns observed between mares and stallions. The interplay between photoperiod-induced metabolic changes and exercise-induced enzyme fluctuations highlights the need for seasonally adapted training regimes to optimise equine health and performance. These findings contribute to a broader understanding of equine metabolic adaptations and highlight the importance of considering seasonal factors in equine health monitoring and management strategies. Future research should explore the underlying hormonal mechanisms driving these variations and investigate the potential implications for optimising performance in different equestrian disciplines.

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References

Alghannam, A. F., Ghaith, M. M., & Alhussain, M. H. (2021). Regulation of Energy Substrate Metabolism in Endurance Exercise. *International Journal of Environmental Research and Public Health*, 18(9), 4963. https://doi.org/10.3390/ijerph18094963.

Andriichuk, A., & Tkachenko, H. (2017). Effect of gender and exercise on haematological and biochemical parameters in Holsteiner horses. *Journal of animal physiology and animal nutrition*, 101(5), e404–e413. https://doi.org/10.1111/jpn.12620.

Asahi, Y., Arai, T., & Tanaka, Y. (2024). Changes in plasma metabolite concentrations and enzyme activities in aging riding horses. *Frontiers in veterinary science*, 11, 1345548. https://doi.org/10.3389/fvets.2024.1345548.

Beech, J., Boston, R. C., McFarlane, D., & Lindborg, S. (2009). Evaluation of plasma ACTH, alpha-melanocyte-stimulating hormone, and insulin concentrations during various photoperiods in clinically normal horses and ponies and those with pituitary pars intermedia dysfunction. *Journal of the American Veterinary Medical Association*, 235(6), 715–722. https://doi.org/10.2460/javma.235.6.715.

Bryan, K., McGivney, B. A., Farries, G., McGettigan, P. A., McGivney, C. L., Gough, K. F., MacHugh, D. E., Katz, L. M., & Hill, E. W. (2017). Equine skeletal muscle adaptations to exercise and training: evidence of differential regulation of autophagosomal and mitochondrial components. *BMC genomics*, 18(1), 595. https://doi.org/10.1186/s12864-017-4007-9.

Cappelli, K., Hosseini-Ghaffari, M., Lopreiato, V., & Mecocci, S. (2024). Editorial: Physiological response to exercise-induced stress and stressful environmental

stimuli: insights from systems biology. *Frontiers in veterinary science*, 11, 1369154. https://doi.org/10.3389/fvets.2024.1369154.

Clay, C. M., & Clay, J. N. (1992). Endocrine and testicular changes associated with season, artificial photoperiod, and the peri-pubertal period in stallions. *The Veterinary clinics of North America. Equine practice*, 8(1), 31–56. https://doi.org/10.1016/s0749-0739(17)30465-0.

Cronise, R. J., Sinclair, D. A., & Bremer, A. A. (2014). The "metabolic winter" hypothesis: a cause of the current epidemics of obesity and cardiometabolic disease. *Metabolic syndrome and related disorders*, 12(7), 355–361. https://doi.org/10.1089/met.2014.0027.

Ebisuda, Y., Mukai, K., Takahashi, Y., Yoshida, T., Matsuhashi, T., Kawano, A., Miyata, H., Kuwahara, M., & Ohmura, H. (2024). Heat acclimation improves exercise performance in hot conditions and increases heat shock protein 70 and 90 of skeletal muscles in Thoroughbred horses. *Physiological reports*, 12(10), e16083. https://doi.org/10.14814/phy2.16083.

Feriel, J., Tchipeva, D., & Depasse, F. (2021). Effects of circadian variation, lifestyle and environment on hematological parameters: A narrative review. *International journal of laboratory hematology*, 43(5), 917–926. https://doi.org/10.1111/ijlh.13590.

Ferlazzo, A., Cravana, C., Fazio, E., & Medica, P. (2020). The different hormonal system during exercise stress coping in horses. *Veterinary world*, 13(5), 847–859. https://doi.org/10.14202/vetworld.2020.847-859.

Ferraro, E., Giammarioli, A. M., Chiandotto, S., Spoletini, I., & Rosano, G. (2014). Exercise-induced skeletal muscle remodeling and metabolic adaptation: redox signaling and role of autophagy. *Antioxidants & redox signaling*, 21(1), 154–176. https://doi.org/10.1089/ars.2013.5773.

Furrer, R., Hawley, J. A., & Handschin, C. (2023). The molecular athlete: exercise physiology from mechanisms to medals. *Physiological reviews*, 103(3), 1693–1787. https://doi.org/10.1152/physrev.00017.2022.

Gharahdaghi, N., Phillips, B. E., Szewczyk, N. J., Smith, K., Wilkinson, D. J., & Atherton, P. J. (2021). Links Between Testosterone, Oestrogen, and the Growth Hormone/Insulin-Like Growth Factor Axis and Resistance Exercise Muscle Adaptations. *Frontiers in physiology*, 11, 621226. https://doi.org/10.3389/fphys.2020.621226.

Handelsman, D. J., Hirschberg, A. L., & Bermon, S. (2018). Circulating Testosterone as the Hormonal Basis of Sex Differences in Athletic Performance. *Endocrine reviews*, 39(5), 803–829. https://doi.org/10.1210/er.2018-00020.

Haneda, S., Dini, P., Esteller-Vico, A., Scoggin, K. E., Squires, E. L., Troedsson, M. H., Daels, P., Nambo, Y., & Ball, B. A. (2021). Estrogens Regulate Placental Angiogenesis in Horses. *International journal of molecular sciences*, 22(22), 12116. https://doi.org/10.3390/ijms222212116.

Ingelson-Filpula, W. A., & Storey, K. B. (2021). Muscles in Winter: The Epigenetics of Metabolic Arrest. *Epigenomes*, 5(4), 28. https://doi.org/10.3390/epigenomes5040028.

Ishimaru, M., Okano, A., Matsui, A., Murase, H., Korosue, K., Akiyama, K., & Taya, K. (2024). Effects of an extended photoperiod on body composition of young Thoroughbreds in training. *The Journal of veterinary medical science*, 86(1), 58–65. https://doi.org/10.1292/jvms.23-0349.

Kunii, H., Nambo, Y., Okano, A., Matsui, A., Ishimaru, M., Asai, Y., Sato, F., Fujii, K., Nagaoka, K., Watanabe, G., & Taya, K. (2015). Effects of an extended

photoperiod on gonadal function and condition of hair coats in Thoroughbred colts and fillies. *Journal of equine science*, 26(2), 57–66. https://doi.org/10.1294/jes.26.57.

Kurhaluk, N., Lukash, O., & Tkachenko, H. (2022). Photoperiod-dependent changes in oxidative stress markers in the blood of Shetland pony mares and stallions involved in recreational horseback riding. *Chronobiology international*, 39(11), 1419–1434. https://doi.org/10.1080/07420528.2022.2115922.

Kurhaluk, N., Tkaczenko, H., Tkachova, I., Lukash, O. (2024). Activity of antioxidant enzymes in the blood of Shetland pony mares and stallions involved in recreational horseback riding: role of photoperiod and exercise. *Scientific and technical bulletin of Livestock farming Institute of NAAS of Ukraine*, 132, 74-86. https://doi.org/10.32900/2312-8402-2024-132-74-86.

Li, C., Shu, H., & Gu, X. (2025). Photoperiod Management in Farm Animal Husbandry: A Review. *Animals*, 15(4), 591. https://doi.org/10.3390/ani15040591.

Maśko, M., Domino, M., Jasiński, T., & Witkowska-Piłaszewicz, O. (2021). The Physical Activity-Dependent Hematological and Biochemical Changes in School Horses in Comparison to Blood Profiles in Endurance and Race Horses. *Animals*, 11(4), 1128. https://doi.org/10.3390/ani11041128.

Massányi, M., Halo, M., Jr, Massányi, P., Mlyneková, E., Greń, A., Formicki, G., & Halo, M. (2022). Changes in haematological and biochemical parameters in blood serum of horses during exposition to workload stress. *Heliyon*, 8(12), e12241. https://doi.org/10.1016/j.heliyon.2022.e12241.

McCutcheon, L. J., Geor, R. J., & Hinchcliff, K. W. (1999). Effects of prior exercise on muscle metabolism during sprint exercise in horses. *Journal of applied physiology (Bethesda, Md.: 1985)*, 87(5), 1914–1922. https://doi.org/10.1152/jappl.1999.87.5.1914.

O'Brien, C., Darcy-Dunne, M. R., & Murphy, B. A. (2020). The effects of extended photoperiod and warmth on hair growth in ponies and horses at different times of year. *PloS one*, 15(1), e0227115. https://doi.org/10.1371/journal.pone.0227115.

Pettersson, J., Hindorf, U., Persson, P., Bengtsson, T., Malmqvist, U., Werkström, V., & Ekelund, M. (2008). Muscular exercise can cause highly pathological liver function tests in healthy men. *British journal of clinical pharmacology*, 65(2), 253–259. https://doi.org/10.1111/j.1365-2125.2007.03001.x.

Richardson, R. B., & Mailloux, R. J. (2023). Mitochondria Need Their Sleep: Redox, Bioenergetics, and Temperature Regulation of Circadian Rhythms and the Role of Cysteine-Mediated Redox Signaling, Uncoupling Proteins, and Substrate Cycles. *Antioxidants (Basel, Switzerland)*, 12(3), 674. https://doi.org/10.3390/antiox12030674.

Satué, K., Miguel-Pastor, L., Chicharro, D., & Gardón, J. C. (2022). Hepatic Enzyme Profile in Horses. *Animals: an open access journal from MDPI*, 12(7), 861. https://doi.org/10.3390/ani12070861.

Shephard R. J. (1993). Metabolic adaptations to exercise in the cold. An update. *Sports medicine (Auckland, N.Z.)*, 16(4), 266–289. https://doi.org/10.2165/00007256-199316040-00005.

Small, L., Lundell, L. S., Iversen, J., Ehrlich, A. M., Dall, M., Basse, A. L., Dalbram, E., Hansen, A. N., Treebak, J. T., Barrès, R., & Zierath, J. R. (2023). Seasonal light hours modulate peripheral clocks and energy metabolism in mice. *Cell metabolism*, 35(10), 1722–1735.e5. https://doi.org/10.1016/j.cmet.2023.08.005.

Smith, J. A. B., Murach, K. A., Dyar, K. A., & Zierath, J. R. (2023). Exercise metabolism and adaptation in skeletal muscle. Nature reviews. *Molecular cell biology*, 24(9), 607–632. https://doi.org/10.1038/s41580-023-00606-x.

Stanisz A. 2006, 2007. *An affordable course of statistics using STATISTICA PL on examples from medicine*. Vol. 1-3. Basic Statistics. StatSoft Polska, Krakow, 2006, 2007. – 532 p., ISBN 83-88724-18-5.

Szwarocka-Priebe, T., & Gill, J. (1984). Seasonal enzyme activity changes in two aminotransferases AspAT and AlAT, acid and alkaline phosphatases and aldolase in the serum of Thoroughbred horses during a racing season. *Acta physiologica Polonica*, 35(3), 249–256.

Tiller, N. B., & Stringer, W. W. (2023). Exercise-induced increases in "liver function tests" in a healthy adult male: Is there a knowledge gap in primary care?. *Journal of family medicine and primary care*, 12(1), 177–180. https://doi.org/10.4103/jfmpc.jfmpc_1923_22.

Tkaczenko, H., Lukash, O., & Kurhaluk, N. (2024). Analysis of the season-dependent component in the evaluation of morphological and biochemical blood parameters in Shetland ponies of both sexes during exercise. *Journal of veterinary research*, 68(1), 155–166. https://doi.org/10.2478/jvetres-2024-0017.

viviD, D., & Bentley, G. E. (2018). Seasonal Reproduction in Vertebrates: Melatonin Synthesis, Binding, and Functionality Using Tinbergen's Four Questions. *Molecules (Basel, Switzerland)*, 23(3), 652. https://doi.org/10.3390/molecules23030652.

Walton, J. C., Weil, Z. M., & Nelson, R. J. (2011). Influence of photoperiod on hormones, behavior, and immune function. *Frontiers in neuroendocrinology*, 32(3), 303–319. https://doi.org/10.1016/j.yfrne.2010.12.003.

Wickham, K. A., McCarthy, D. G., Spriet, L. L., & Cheung, S. S. (2021). Sex differences in the physiological responses to exercise-induced dehydration: consequences and mechanisms. *Journal of applied physiology (Bethesda, Md.: 1985)*, 131(2), 504–510. https://doi.org/10.1152/japplphysiol.00266.2021.

Williams, G. L., Thorson, J. F., Prezotto, L. D., Velez, I. C., Cardoso, R. C., & Amstalden, M. (2012). Reproductive seasonality in the mare: neuroendocrine basis and pharmacologic control. *Domestic animal endocrinology*, 43(2), 103–115. https://doi.org/10.1016/j.domaniend.2012.04.001.

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AUTOPHAGIC FUNCTION OF THE LIVER OF VACCINATED RAINBOW TROUT (*ONCORHYNCHUS MYKISS* WALBAUM) FOLLOWING *YERSINIA RUCKERI* INFECTION

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The liver plays a critical role in maintaining metabolic homeostasis and immune defence in fish, particularly in response to bacterial infections. Autophagy, a conserved cellular process essential for homeostasis and pathogen clearance, has been implicated in host defence mechanisms. However, the role of autophagy in the liver of vaccinated fish following pathogen exposure remains largely unexplored. Yersinia ruckeri, the causative agent of enteric redmouth disease (ERM), poses a significant threat to rainbow trout (Oncorhynchus mykiss Walbaum) aquaculture, primarily affecting the liver, spleen and kidneys. Vaccination is a widely used preventive strategy, but its effect on autophagic activity during infection is not well understood. The aim of this study was to evaluate the autophagic response in the liver of vaccinated rainbow trout following Y. ruckeri infection by assessing the activity of four lysosomal enzymes: alanyl aminopeptidase (AAP), leucyl aminopeptidase (LAP), acid phosphatase (AcP) and β-N-acetylglucosaminidase (NAG). Rainbow trout were divided into experimental groups: unvaccinated control, vaccinated uninfected, unvaccinated infected and vaccinated infected. The fish were orally immunised with a Y. ruckeri vaccine and challenged with a virulent strain of Y. ruckeri. The results showed significant differences in lysosomal enzyme activity between groups, indicating that vaccination modulated the hepatic autophagic response during bacterial infection. AAP and LAP activity peaked in unvaccinated infected fish, whereas vaccinated fish exhibited a blunted enzymatic response, suggesting that vaccination attenuated excessive autophagic activation. Similarly, AcP and NAG activity patterns indicated an infection-induced autophagic response that was partially attenuated in vaccinated fish. These results suggest that vaccination influences autophagy-related enzymatic activity in the liver of rainbow trout, potentially enhancing pathogen clearance while preventing excessive cellular stress. Understanding the interplay between vaccination, infection and autophagy may provide valuable insights to optimise vaccination strategies and improve disease management in aquaculture.

Keywords: autophagy, liver, rainbow trout, *Yersinia ruckeri*, lysosomal enzymes, vaccination, bacterial infection, metabolic homeostasis

АУТОФАГІЧНА ФУНКЦІЯ ПЕЧІНКИ ВАКЦИНОВАНОЇ РАЙДУЖНОЇ ФОРЕЛІ (*ONCORHYNCHUS MYKISS* WALBAUM) ПІСЛЯ ІНФЕКУВАННЯ ШТАМОМ *YERSINIA RUCKERI*

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Печінка відіграє важливу роль у підтриманні метаболічної гомеостазу та імунного захисту у риб, особливо у відповідь на бактеріальні інфекції. Аутофагія, консервативний клітинний процес, необхідний для гомеостазу та очищення від патогенів, пов'язана з механізмами захисту організму. Однак роль аутофагії у печінці вакцинованих риб після впливу патогенів залишається в основному не дослідженою. Yersinia ruckeri, збудник кишкового єрсиніозу, становить серйозну загрозу для розведення райдужної форелі (Oncorhynchus mykiss Walbaum), впливаючи насамперед на печінку, селезінку та нирки. Вакцинація є широко використовуваною профілактичною стратегією, але її вплив на аутофагічну активність під час інфекції не до кінця зрозумілий. Метою цього дослідження було оцінити аутофагічну відповідь у печінці вакцинованої райдужної форелі після інфекції Y. ruckeri, оцінюючи активність чотирьох лізосомальних ферментів: аланіламіноептидази (AAP), лейциламіноептидази (LAP), кислотної фосфатази (AcP) та β-N-ацетилглюкозамінідази (NAG). Райдужну форель поділили на експериментальні групи: невакциновану контрольну, вакциновану неінфековану, невакциновану заражену та вакциновану заражену. Рибу орально імунізували вакциною від Y. ruckeri та інфікували штамом Y. ruckeri. Результати показали значні відмінності в активності лізосомальних ферментів між групами, що вказує на те, що вакцинація модулює аутофагічну відповідь печінки під час бактеріальної інфекції. Активність AAP і LAP досягла піку у невакцинованих заражених риб, в той час як вакциновані риби показали знижену ензиматичну відповідь, що вказує на те, що вакцинація послабила надмірну аутофагічну активацію. Аналогічно, патерни активності AcP і NAG вказують на аутофагічну відповідь, індуковану інфекцією, яка частково була ослаблена у вакцинованих риб. Ці результати свідчать про те, що вакцинація впливає на активність аутофагії у печінці райдужної форелі, потенційно покращуючи очищення від патогенів і запобігаючи надмірному клітинному стресу. Розуміння взаємодії між вакцинацією, інфекцією та аутофагією може надати важливі відомості для оптимізації стратегій вакцинації та покращення управління захворюваннями у аквакультурі.

Ключові слова: аутофагія, печінка, райдужна форель, *Yersinia ruckeri*, лізосомальні ферменти, вакцинація, бактеріальна інфекція, метаболічний гомеостаз.

Introduction. The liver plays a crucial role in maintaining homeostasis and immune defence in fish, acting as a central metabolic organ and a key component of the innate immune system (Mokhtar D.M. et al., 2023). Autophagy, a highly conserved cellular degradation and recycling process, is essential for cellular homeostasis, pathogen clearance and immune regulation (Chun Y. and Kim J., 2018; Gómez-Virgilio L. et al., 2022). In teleost fish, the autophagic process has been implicated in response to various stressors, including infectious diseases (Johnstone C. and Chaves-Pozo E., 2022; Zhou Z. et al., 2022). However, the precise role of autophagy in the liver of vaccinated fish following pathogen exposure remains largely unexplored.

Yersinia ruckeri, the causative agent of enteric redmouth disease (ERM), is a major bacterial pathogen affecting rainbow trout (*Oncorhynchus mykiss* Walbaum) aquaculture worldwide (Kumar G. et al., 2015). This Gram-negative bacterium primarily affects the liver, spleen and kidneys, resulting in severe economic losses in aquaculture (Wrobel A. et al., 2019). Vaccination has been widely used as a preventive strategy against ERM, significantly reducing mortality and improving disease resistance (Villumsen K.R. et al., 2014; Wangkahart E. et al., 2019). However, the effect of vaccination on cellular processes such as autophagy in the liver during *Y. ruckeri* infection remains poorly understood.

Recent studies suggest that autophagy plays a dual role in bacterial infections, acting both as a host defence mechanism and as a potential pathway for pathogen survival (Desai M. et al., 2015; Zhou Z. et al., 2022). In mammals, autophagy has been shown to eliminate intracellular pathogens, regulate inflammation and modulate immune responses (Deretic V., 2021; Gan T. et al., 2023). Similar mechanisms are thought to exist in fish, but empirical evidence remains limited (Zhou Z. et al., 2022). Understanding how vaccination affects the autophagic response in infected fish may provide valuable insights into the protective mechanisms underlying vaccine efficacy.

The liver, as a major immune and metabolic organ, is particularly sensitive to bacterial infection (Tarasenko T.N. and McGuire P.J., 2017). During *Y. ruckeri* infection, hepatocytes undergo significant cellular stress, which may induce autophagy as an adaptive response. Activation of autophagy-related genes and proteins in response to bacterial invasion may enhance the host's ability to eliminate pathogens and mitigate tissue damage (Kroemer G. et al., 2010; Rahman M.A. et al., 2024). However, excessive or dysregulated autophagy can also contribute to cellular dysfunction and a balanced response is required for optimal liver function (Ke P.Y., 2019).

Studies in model organisms have shown that vaccination can modulate autophagic pathways, potentially enhancing pathogen clearance and immune responses (Jang Y.J. et al., 2019). In fish, the role of autophagy in vaccinated individuals remains an emerging area of research (Kurhaluk N. and Tkachenko H., 2021). Investigating autophagic activity in the liver of vaccinated and infected rainbow trout may provide new insights into how vaccination affects cellular defence mechanisms against bacterial pathogens.

This study aims to assess the autophagic function of the liver in vaccinated rainbow trout following infection with *Y. ruckeri*. By evaluating the activity of four lysosomal enzymes – alanyl aminopeptidase (AAP), leucyl aminopeptidase (LAP), acid phosphatase (AcP) and β-N-acetylglucosaminidase (NAG) – we aim to determine whether vaccination enhances or suppresses autophagic responses during bacterial challenge. A deeper understanding of this process may contribute to the development of improved vaccination strategies and disease management practices in aquaculture. Elucidating the interplay between vaccination, infection and autophagy in rainbow trout may have wider implications for fish health and the sustainability of aquaculture.

Materials and methods.

Fish. Rainbow trout (*Oncorhynchus mykiss* Walbaum) with an average weight of 310-320 g were used in the experiment. The study was conducted at the Salmonid Research Department of the Stanislaw Sakowicz Institute of Inland Fisheries in Olsztyn, Poland. The fish were kept under controlled environmental conditions to ensure stable water parameters: temperature of 14.5 ± 0.5°C, pH of 7.5 and dissolved oxygen of approximately 12 ppm. Supplemental oxygen was provided by aeration at a rate of 25 litres per minute under a 12 h light/12 h dark photoperiod. Fish were fed a commercial pelleted diet using 12-hour fish belt feeders to ensure optimal food intake. Daily food allowances were calculated according to established dietary guidelines.

All enzymatic assays were performed at the Department of Zoology and Department of Animal Physiology, Institute of Biology, University of Pomerania, Słupsk, Poland, using standardised protocols to ensure consistency and reliability of results.

Study groups. Fish were divided into two experimental groups: (I) an unvaccinated control group and (II) a group orally vaccinated against *Yersinia ruckeri*. The fish were housed in 1000 L square tanks, with 150 fish per tank, under identical environmental conditions. The vaccine was developed at the Department of Fish Diseases, National Veterinary Research Institute in Puławy, Poland, according to a patented process (Patent No. P.428259). The oral vaccine contained *Y. ruckeri* at a concentration of 1×10^9 cells/ml, which was incorporated into the feed and administered over three consecutive days. After vaccination, the fish were maintained in water at 14.5 ± 0.5°C and pH 7.5 for 60 days. The challenge test was performed on day 61 after vaccination.

For the challenge test, 40 fish were divided into four subgroups (10 fish per group): (I) uninfected control, (II) infected control, (III) vaccinated uninfected and (IV) vaccinated infected. Experimental infection was induced with *Y. ruckeri* serotype O1, biotype 2, isolated from an outbreak on a rainbow trout farm. Prior to infection, *Y. ruckeri* was cultured on tryptone soy agar supplemented with 5% horse blood (Oxoid®) at 25 ± 1°C for 24 h. Fish were infected intraperitoneally with *Y. ruckeri* at a dose of 1×10^7 CFU/mL suspension.

The experiment lasted 10 days, during which time fish were observed three times daily for behavioural changes, clinical signs and mortality. Cumulative survival was assessed based on mortality counts. Swabs from the head kidney of deceased fish were analysed to confirm *Y. ruckeri* as the cause of death. Mortality rates were expressed as percentages, with n = 10 considered as 100%.

Sampling procedures. Fish were euthanised 10 days after challenge. The liver was excised *in situ*. For lysosomal enzyme assays, liver tissue was rinsed with 0.15 M KCl cold isolation buffer to remove blood before homogenisation on ice using a Potter-Elvehjem glass homogeniser with a Teflon motorised pestle. The isolation buffer contained 0.25 M sucrose and 2 mM EDTA, adjusted to pH 7.0 with KOH. Homogenates (20% w/v) were prepared for differential centrifugation according to the method of DeMartino and Goldberg (1978). After centrifugation, the supernatant fractions were resuspended in 50 mM acetic acid/sodium acetate buffer (pH 5.0) before storage and further analysis. Isolated fractions were subjected to two freeze-thaw cycles to ensure enzyme activation. Protein concentration was determined by the Bradford method using bovine serum albumin as standard. The absorbance was measured at 595 nm. All assays were performed in duplicate at 22 ± 0.5°C, with enzymatic reactions initiated by the addition of tissue supernatant.

Lysosomal enzyme assay. The activities of alanyl aminopeptidase and leucyl aminopeptidase were determined spectrophotometrically according to the method of

DeMartino and Goldberg (1978). The reaction was initiated by incubating 50 μL of sample with 500 μL substrate incubation medium containing DMF (Serva, Germany) at 37°C, pH 6.0, for 60 min. The reaction was terminated by the addition of 500 μL stop buffer containing Fast Blue BB salt dissolved in 2% Tween 20 (Sigma, USA). The absorbance was measured at 540 nm. Alanyl aminopeptidase activity was determined using L-alanyl-2-naphthylamine in 0.1 M PBS buffer, while leucyl aminopeptidase activity was measured using L-leucyl-2-naphthylamine in 0.1 M PBS buffer (pH 7.0).

Acid phosphatase and β-N-acetylglucosaminidase activities were measured at 420 nm using 4-nitrophenyl derivatives as substrates according to the method of Barrett and Heath (1977). Enzyme activities were expressed as nmol per hour per mg protein.

Statistical analysis. Results are expressed as mean ± S.D. Each data set was processed separately using Statistica 13.3 (TIBCO Software Inc., USA). Normality of data was assessed using the Kolmogorov-Smirnov test ($p > 0.05$), while homogeneity of variance was assessed using Levene's test. Significant differences within and between groups were determined using unequal sample size one-way analysis of variance (ANOVA) followed by Tukey's post-hoc test. Correlation and regression analyses were also performed to assess relationships between parameters. Differences were considered statistically significant at $p < 0.05$.

Results. The activity of key lysosomal enzymes in the hepatic tissue of rainbow trout of rainbow trout orally immunised against *Y. ruckeri* and challenged with *Y. ruckeri* is shown in Fig. 1.

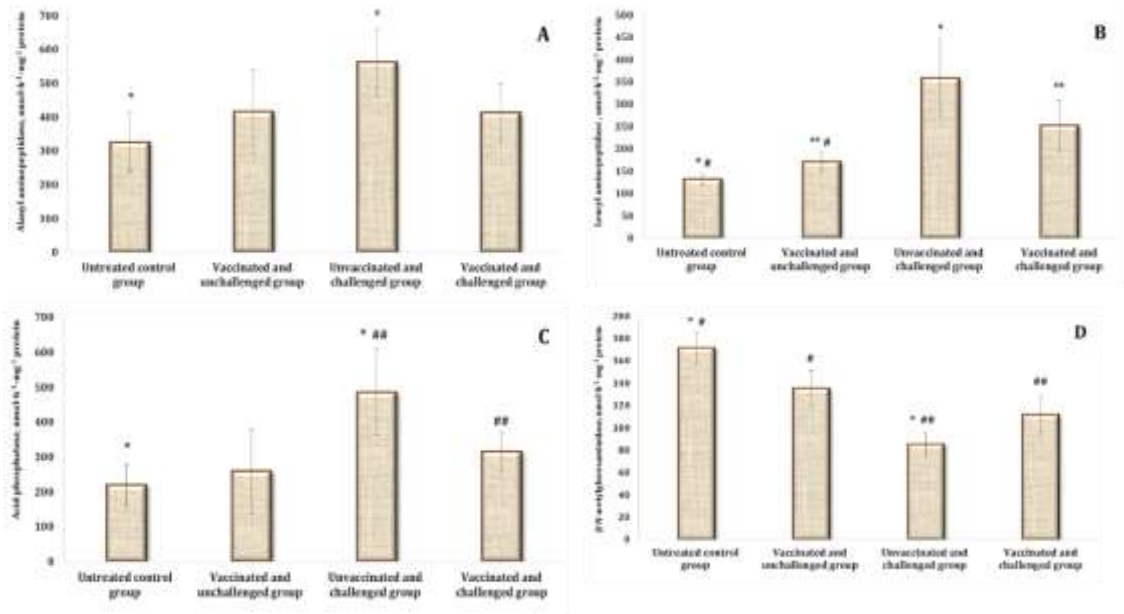

Fig. 1. **The activity of key lysosomal enzymes [alanyl aminopeptidase (AAP), leucyl aminopeptidase (LAP), acid phosphatase (AcP) and β-N-acetylglucosaminidase (NAG)] in the hepatic tissues of rainbow trout orally immunised against *Y. ruckeri* and challenged with *Y. ruckeri*.**

Data are presented as means ± S.D. (n = 10).
** Significant differences ($p < 0.05$) between the untreated control group and the group challenged with Y. ruckeri;*
*** Significant differences ($p < 0.05$) between the vaccinated group subjected to the Y. ruckeri challenge and the vaccinated group;*
 # Significant differences ($p < 0.05$) between the untreated control group and the vaccinated group;
Significant differences ($p < 0.05$) between the vaccinated group subjected to the Y. ruckeri challenge and the group challenged with Y. ruckeri.

The activity of four lysosomal enzymes – alanyl aminopeptidase (AAP), leucyl aminopeptidase (LAP), acid phosphatase (AcP) and β-N-acetylglucosaminidase (NAG) – varied significantly among the experimental groups, reflecting the influence of vaccination and *Y. ruckeri* infection on liver function in rainbow trout.

AAP activity had the highest mean value in the unvaccinated and challenged group (562.11 ± 98.78 nmol·h^{-1}·mg^{-1} protein), indicating a pronounced enzymatic response to bacterial infection. The vaccinated and unchallenged group showed increased AAP activity (415.54 ± 124.1 nmol·h^{-1}·mg^{-1} protein) compared to the untreated control group (325.21 ± 87.11 nmol·h^{-1}·mg^{-1} protein), indicating a possible vaccine-induced priming effect. However, the vaccinated and challenged group showed AAP activity (411.89 ± 85.47 nmol·h^{-1}·mg^{-1} protein) comparable to the vaccinated and unchallenged group, suggesting that vaccination attenuated the infection-induced increase in AAP activity (Fig. 1A).

LAP activity followed a similar trend, with the highest value observed in the unvaccinated and challenged group (358.01 ± 87.58 nmol·h^{-1}·mg^{-1} protein). This significant increase suggests that infection resulted in an enhanced proteolytic response. The vaccinated and challenged group had a lower LAP activity (252.12 ± 56.04 nmol·h^{-1}·mg^{-1} protein) than the unvaccinated and challenged group, but it remained higher than in the vaccinated and unchallenged group (170.69 ± 22.11 nmol·h^{-1}·mg^{-1} protein). The untreated control group had the lowest LAP activity (132.33 ± 10.2 nmol·h^{-1}·mg^{-1} protein), highlighting the baseline enzyme levels in unstressed fish (Fig. 1B).

AcP activity was significantly increased in the unvaccinated and challenged group (485.47 ± 124.1 nmol·h^{-1}·mg^{-1} protein), indicating increased lysosomal degradation processes in response to infection. The vaccinated and challenged group had intermediate AcP activity (314.25 ± 56.2 nmol·h^{-1}·mg^{-1} protein), suggesting that vaccination attenuated the infection-induced increase in lysosomal activity. The vaccinated and unchallenged group had slightly higher AcP activity (258.21 ± 121.2 nmol·h^{-1}·mg^{-1} protein) than the untreated control group (219.45 ± 58.33 nmol·h^{-1}·mg^{-1} protein), possibly due to the immune stimulation associated with vaccination (Fig. 1C).

In contrast to the other enzymes, NAG activity showed an inverse trend, with the lowest activity observed in the unvaccinated and challenged group (85.16 ± 10.2 nmol·h^{-1}·mg^{-1} protein). This decrease suggests a possible exhaustion of lysosomal function under intense infection stress. The vaccinated and challenged group showed higher NAG activity (111.5 ± 16.45 nmol·h^{-1}·mg^{-1} protein), suggesting a partial restoration of enzymatic function by vaccination. The highest NAG activity was observed in the untreated control group (171.25 ± 14.22 nmol·h^{-1}·mg^{-1} protein), followed by the vaccinated and unchallenged group (135.2 ± 16.25 nmol·h^{-1}·mg^{-1} protein), suggesting that both infection and vaccination affect NAG activity differently from the other lysosomal enzymes (Fig. 1D).

Collectively, these results indicate that bacterial infection significantly alters lysosomal enzyme activity in the liver of rainbow trout, with vaccination playing a modulatory role in attenuating infection-induced enzymatic changes. The observed differences in enzyme activity between groups highlight the potential impact of vaccination in regulating lysosomal function during pathogenic challenges.

Discussion. The observed variation in lysosomal enzyme activity between experimental groups highlights the complex interplay between bacterial infection and vaccination in modulating liver function in rainbow trout. The significant increase in AAP, LAP and AcP activities in the unvaccinated and challenged groups is consistent with previous studies indicating that bacterial infection triggers lysosomal activation as part of the immune response (van der Vaart M. et al., 2012; Pérez-Stuardo D. et al., 2020;

Chiang Y.R. et al., 2022). The marked elevation of these enzymes suggests an enhanced proteolytic and degradative response to *Y. ruckeri* infection, likely associated with the breakdown of damaged cellular components and pathogen clearance (Menanteau-Ledouble S. et al., 2020).

Vaccination appeared to attenuate the infection-induced increase in lysosomal enzyme activity. The vaccinated and challenged group had lower AAP, LAP and AcP activities than the unvaccinated and challenged group, suggesting that prior immunisation reduced the inflammatory response and cellular damage caused by the pathogen. This finding is consistent with reports indicating that effective vaccination limits excessive lysosomal activation by enhancing adaptive immune mechanisms, thereby reducing reliance on lysosomal degradation pathways (Zwack E.E. et al., 2015; Osterloh A., 2022).

Interestingly, NAG activity followed a distinct pattern, with the lowest levels observed in the unvaccinated and challenged groups (Fig. 1D). This decrease suggests potential lysosomal dysfunction under severe infection stress, possibly due to depletion of cellular resources or enzyme inactivation caused by prolonged immune activation. The partial restoration of NAG activity in the vaccinated and challenged groups further supports the protective role of vaccination in maintaining lysosomal function. These results corroborate previous findings highlighting the immunomodulatory effects of vaccination in fish species, where vaccine-induced immune priming prevents excessive metabolic stress on lysosomal pathways (Du et al., 2022).

The moderate increase in AAP, LAP and AcP activities in the vaccinated and unvaccinated groups compared to the untreated control group suggests that vaccination alone affects lysosomal function, probably through the stimulation of immune-related metabolic processes. Such effects have been documented in other teleost species, where activation of the immune system following vaccination leads to transient metabolic adjustments in hepatocytes (Aluru N. and Vijayan M.M., 2009; Mussap M. et al., 2024).

In our previous study (Kurhaluk N. et al., 2024), we investigated oxidative stress biomarkers, antioxidant and lysosomal enzyme activity, and biochemical parameters in the liver of rainbow trout vaccinated against enteric redmouth disease and challenged with *Y. ruckeri*. The results showed that in unvaccinated fish, *Y. ruckeri* infection disrupted the oxidative balance, increasing lipid peroxidation, oxidative protein modification and lysosomal enzyme activity, while reducing total antioxidant capacity. In contrast, vaccinated fish showed increased glutathione-related enzyme activity, reduced lipid peroxidation and lower lysosomal enzyme activity after infection compared to unvaccinated and challenged fish. These results suggest that vaccination mitigates oxidative damage and modulates enzymatic responses in fish exposed to *Y. ruckeri* (Kurhaluk N. et al., 2024).

We also evaluated the time-dependent effects of *Y. ruckeri* vaccination on oxidative mechanisms by assessing key biomarkers of lipid peroxidation [2-thiobarbituric acid reactive substances (TBARS)] and protein oxidation [aldehyde and ketone derivatives of oxidatively modified proteins (OMP)], antioxidant defences [superoxide dismutase (SOD), catalase (CAT), glutathione reductase (GR), glutathione peroxidase (GPx), total antioxidant capacity (TAC)] in rainbow trout liver tissue (Tkaczenko H. et al., 2023). A concentrated *Y. ruckeri* vaccine was incorporated into the fish feed and administered three times at two-day intervals. Fish were euthanised at 31, 61 and 181 days post vaccination for liver tissue analysis. Vaccination against *Y. ruckeri* did not significantly alter TBARS levels, but reduced aldehyde and ketonic OMP derivatives, particularly in the first and second months after immunisation. Glutathione-dependent enzyme activity increased, particularly at one and six months post-vaccination, with the highest TAC levels observed at two and six months. The results suggest that vaccination-

induced oxidative stress in liver tissue triggers adaptive responses through transient activation of antioxidant and lysosomal enzymes. In addition, vaccination affected lysosomal membrane permeability, particularly for carbohydrate cleavage, following activation of the immune system against *Y. ruckeri*. Antioxidant defences were generally preserved, as evidenced by the maintenance or increase in CAT, GR and GPx activity after vaccination. These results highlight the role of oxidative mechanisms in the immune response and the potential of vaccination to modulate lysosomal and antioxidant enzyme activity in rainbow trout (Tkaczenko H. et al., 2023).

Collectively, these findings highlight the critical role of lysosomal enzymes in the immune response to bacterial infection and provide further evidence for the beneficial effects of vaccination in modulating lysosomal activity. Further research is needed to elucidate the long-term effects of vaccination on lysosomal function and to explore the potential metabolic trade-offs associated with immune stimulation in fish.

Conclusions. The present study demonstrates that *Y. ruckeri* infection significantly alters lysosomal enzyme activity in the liver of rainbow trout, reflecting an enhanced proteolytic and degradative response to bacterial challenge. Vaccination played a modulatory role by attenuating the infection-induced increase in AAP, LAP and AcP activities, while partially restoring NAG activity, suggesting its protective effect in maintaining lysosomal function. The moderate increase in lysosomal enzyme activity in the vaccinated and unvaccinated groups further suggests that vaccination alone affects metabolic and immune processes in hepatocytes.

These findings highlight the importance of lysosomal enzymes as potential biomarkers for assessing infection severity and vaccine efficacy in fish. Future research should investigate the long-term metabolic consequences of vaccination and its impact on immune homeostasis in aquaculture species. Understanding these mechanisms could help to optimise vaccination strategies and improve disease resistance in farmed fish populations.

References

Aluru, N., & Vijayan, M. M. (2009). Stress transcriptomics in fish: a role for genomic cortisol signaling. *General and comparative endocrinology*, 164(2-3), 142–150. https://doi.org/10.1016/j.ygcen.2009.03.020.

Barrett, A.J., & Heath, M.F. Lysosomal enzymes. In: *Lysosomes, a Laboratory Handbook* (ed. Dingle, J. T.), North Holland, 1977, pp. 19–146.

Bradford M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical biochemistry*, 72, 248–254. https://doi.org/10.1016/0003-2697(76)90527-3.

Chiang, Y. R., Wang, L. C., Lin, H. T., & Lin, J. H. (2022). Bioactivity of orange-spotted grouper (*Epinephelus coioides*) cathepsin L: Proteolysis of bacteria and regulation of the innate immune response. *Fish & shellfish immunology*, 122, 399–408. https://doi.org/10.1016/j.fsi.2022.02.003.

Chun, Y., & Kim, J. (2018). Autophagy: An Essential Degradation Program for Cellular Homeostasis and Life. *Cells*, 7(12), 278. https://doi.org/10.3390/cells7120278.

DeMartino, G. N., & Goldberg, A. L. (1978). Thyroid hormones control lysosomal enzyme activities in liver and skeletal muscle. *Proceedings of the National Academy of Sciences of the United States of America*, 75(3), 1369–1373. https://doi.org/10.1073/pnas.75.3.1369.

Deretic V. (2021). Autophagy in inflammation, infection, and immunometabolism. *Immunity*, 54(3), 437–453. https://doi.org/10.1016/j.immuni.2021.01.018.

Desai, M., Fang, R., & Sun, J. (2015). The role of autophagy in microbial infection and immunity. *ImmunoTargets and therapy*, 4, 13–26. https://doi.org/10.2147/ITT.S76720.

Du, Y., Hu, X., Miao, L., & Chen, J. (2022). Current status and development prospects of aquatic vaccines. *Frontiers in immunology*, 13, 1040336. https://doi.org/10.3389/fimmu.2022.1040336.

Gan, T., Qu, S., Zhang, H., & Zhou, X. J. (2023). Modulation of the immunity and inflammation by autophagy. *MedComm*, 4(4), e311. https://doi.org/10.1002/mco2.311.

Gómez-Virgilio, L., Silva-Lucero, M. D., Flores-Morelos, D. S., Gallardo-Nieto, J., Lopez-Toledo, G., Abarca-Fernandez, A. M., Zacapala-Gómez, A. E., Luna-Muñoz, J., Montiel-Sosa, F., Soto-Rojas, L. O., Pacheco-Herrero, M., & Cardenas-Aguayo, M. D. (2022). Autophagy: A Key Regulator of Homeostasis and Disease: An Overview of Molecular Mechanisms and Modulators. *Cells*, 11(15), 2262. https://doi.org/10.3390/cells11152262.

Jang, Y. J., Kim, J. H., & Byun, S. (2019). Modulation of Autophagy for Controlling Immunity. *Cells*, 8(2), 138. https://doi.org/10.3390/cells8020138.

Johnstone, C., & Chaves-Pozo, E. (2022). Antigen Presentation and Autophagy in Teleost Adaptive Immunity. *International journal of molecular sciences*, 23(9), 4899. https://doi.org/10.3390/ijms23094899.

Ke P. Y. (2019). Diverse Functions of Autophagy in Liver Physiology and Liver Diseases. *International journal of molecular sciences*, 20(2), 300. https://doi.org/10.3390/ijms20020300.

Kroemer, G., Mariño, G., & Levine, B. (2010). Autophagy and the integrated stress response. *Molecular cell*, 40(2), 280–293. https://doi.org/10.1016/j.molcel.2010.09.023.

Kumar, G., Menanteau-Ledouble, S., Saleh, M., & El-Matbouli, M. (2015). *Yersinia ruckeri*, the causative agent of enteric redmouth disease in fish. *Veterinary research*, 46(1), 103. https://doi.org/10.1186/s13567-015-0238-4.

Kurhaluk, N., & Tkachenko, H. (2021). Antioxidants, lysosomes and elements status during the life cycle of sea trout *Salmo trutta* m. *trutta* L. *Scientific reports*, 11(1), 5545. https://doi.org/10.1038/s41598-021-85127-3.

Kurhaluk, N., Grudniewska, J., Pękala-Safińska, A., Pajdak-Czaus, J., Terech-Majewska, E., Platt-Samoraj, A., & Tkaczenko, H. (2024). Biomarkers of oxidative stress, biochemical changes, and the activity of lysosomal enzymes in the livers of rainbow trout (*Oncorhynchus mykiss* Walbaum) vaccinated against yersiniosis before a *Yersinia ruckeri* challenge. *Journal of veterinary research*, 68(3), 325–336. https://doi.org/10.2478/jvetres-2024-0050.

Menanteau-Ledouble, S., Nöbauer, K., Razzazi-Fazeli, E., & El-Matbouli, M. (2020). Effects of *Yersinia ruckeri* invasion on the proteome of the Chinook salmon cell line CHSE-214. *Scientific reports*, 10(1), 11840. https://doi.org/10.1038/s41598-020-68903-5.

Mokhtar, D. M., Zaccone, G., Alesci, A., Kuciel, M., Hussein, M. T., & Sayed, R. K. A. (2023). Main Components of Fish Immunity: An Overview of the Fish Immune System. *Fishes*, 8(2), 93. https://doi.org/10.3390/fishes8020093.

Mussap, M., Puddu, M., & Fanos, V. (2024). Metabolic Reprogramming of Immune Cells Following Vaccination: From Metabolites to Personalized Vaccinology. *Current medicinal chemistry*, 31(9), 1046–1068. https://doi.org/10.2174/0929867330666230509110108.

Osterloh, A. (2022). Vaccination against Bacterial Infections: Challenges, Progress, and New Approaches with a Focus on Intracellular Bacteria. *Vaccines*, 10(5), 751. https://doi.org/10.3390/vaccines10050751.

Pérez-Stuardo, D., Espinoza, A., Tapia, S., Morales-Reyes, J., Barrientos, C., Vallejos-Vidal, E., Sandino, A. M., Spencer, E., Toro-Ascuy, D., Rivas-Pardo, J. A., Reyes-López, F. E., & Reyes-Cerpa, S. (2020). Non-Specific Antibodies Induce Lysosomal Activation in Atlantic Salmon Macrophages Infected by *Piscirickettsia salmonis*. *Frontiers in immunology*, 11, 544718. https://doi.org/10.3389/fimmu.2020.544718.

Rahman, M. A., Sarker, A., Ayaz, M., Shatabdy, A. R., Haque, N., Jalouli, M., Rahman, M. H., Mou, T. J., Dey, S. K., Hoque Apu, E., Zafar, M. S., & Parvez, M. A. K. (2024). An Update on the Study of the Molecular Mechanisms Involved in Autophagy during Bacterial Pathogenesis. *Biomedicines*, 12(8), 1757. https://doi.org/10.3390/biomedicines12081757.

Tarasenko, T. N., & McGuire, P. J. (2017). The liver is a metabolic and immunologic organ: A reconsideration of metabolic decompensation due to infection in inborn errors of metabolism (IEM). *Molecular genetics and metabolism*, 121(4), 283–288. https://doi.org/10.1016/j.ymgme.2017.06.010.

Tkaczenko, H., Grudniewska, J., Pękala-Safińska, A., Terech-Majewska, E., & Kurhaluk, N. (2023). Time-dependent changes in oxidative stress biomarkers and activities of lysosomal and antioxidant enzymes in hepatic tissue of rainbow trout (*Oncorhynchus mykiss* Walbaum) following vaccination against *Yersinia ruckeri*. *Fisheries & Aquatic Life*, 31, 133-146. https://doi.org/10.2478/aopf-2023-0014.

van der Vaart, M., Spaink, H. P., & Meijer, A. H. (2012). Pathogen recognition and activation of the innate immune response in zebrafish. *Advances in hematology*, 2012, 159807. https://doi.org/10.1155/2012/159807.

Villumsen, K. R., Neumann, L., Ohtani, M., Strøm, H. K., & Raida, M. K. (2014). Oral and anal vaccination confers full protection against enteric redmouth disease (ERM) in rainbow trout. *PloS one*, 9(4), e93845. https://doi.org/10.1371/journal.pone.0093845.

Wangkahart, E., Secombes, C. J., & Wang, T. (2019). Dissecting the immune pathways stimulated following injection vaccination of rainbow trout (*Oncorhynchus mykiss*) against enteric redmouth disease (ERM). *Fish & shellfish immunology*, 85, 18–30. https://doi.org/10.1016/j.fsi.2017.07.056.

Wrobel, A., Leo, J. C., & Linke, D. (2019). Overcoming Fish Defences: The Virulence Factors of *Yersinia ruckeri*. *Genes*, 10(9), 700. https://doi.org/10.3390/genes10090700.

Zhou, Z., He, Y., Wang, S., Wang, Y., Shan, P., Li, P. (2022). Autophagy regulation in teleost fish: A double-edged sword. *Aquaculture*, 558, 738369. https://doi.org/10.1016/j.aquaculture.2022.738369.

Zwack, E. E., Snyder, A. G., Wynosky-Dolfi, M. A., Ruthel, G., Philip, N. H., Marketon, M. M., Francis, M. S., Bliska, J. B., & Brodsky, I. E. (2015). Inflammasome activation in response to the *Yersinia type* III secretion system requires hyperinjection of translocon proteins YopB and YopD. *mBio*, 6(1), e02095-14. https://doi.org/10.1128/mBio.02095-14.

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ПРАВИЛА ОФОРМЛЕННЯ ТА ПРИЙОМУ СТАТЕЙ ДО «НАУКОВО-ТЕХНІЧНОГО БЮЛЕТЕНЮ ІНСТИТУТУ ТВАРИННИЦТВА НААН»

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Структура статті	
Індекс УДК	виключка за лівим краєм без абзацного відступу, 12 pt
Назва статті	великими літерами, виключка по центру. 14 pt, напівжирним шрифтом
Авторські дані	Не більше 5-ти авторів. Ім’я та прізвище (повністю), науковий ступінь, звання, код ORCID, e-mail. (Якщо автор не зареєстрований в ORCID, потрібно створити обліковий запис за посиланням http://orcid.org.).
Афіліація	Напівжирним, виключка по центру, 12 pt. Організація (місце роботи), місто, країна.
Анотація	Курсивом, 12 pt, не менше 1800 знаків з пробілами.

	В анотації лаконічно та інформативно описуються дані щодо місця проведення дослідження, основні результати та їх практична цінність, НЕ вказується мета та методи роботи. Не можна в анотацію копіювати речення та абзаци з розділів статті.
Ключові слова	Курсивом, 12 pt, 5-7 слів або словосполучень.
Вступ	Виключка по ширині, 12 pt 1-2 стор. У розділі висвітлюється сучасний стан розглянутої проблеми, наукова новизна роботи, аналізуються дослідження та публікації за останні 5 років, обґрунтовується актуальність, напівжирним виділяється **мета дослідження.** В кожному посиланні коротко розкрити суть та результати досліджень автора (-ів), які цитуються. Посилання на літературу необхідно подавати у круглих дужках (…), наприклад: (Ivanov A. V., 2023; Zlobin S. E. et al., 2022a). Якщо потрібно зазначити посилання всередині рядка, варто вказувати тільки рік у (), наприклад: Тимошин М. (2023) у своїй роботі ....". В одному посиланні не варто цитувати більше 3-ох джерел.
Матеріали та методи досліджень	Зазначається об’єкт дослідження та експериментальна база. Викладена методологія повинна дати можливість повністю відтворити Ваші дослідження.
Результати досліджень	Розділ експериментальної статті повинен складати не менше 70 % від обсягу статті (не враховуючи анотацій та бібліографії) з повним обґрунтуванням отриманих наукових результатів. Таблиці, рисунки та формули повинні бути пронумеровані та подані після посилання на них у тексті. Слово “*Таблиця* __” писати справа курсивом з номером арабськими цифрами. **Заголовок таблиці** – напівжирним, виключка по центру. Посилання на таблицю у тексті вказують скорочено перед розміщенням таблиці. Примітки до таблиці – курсивом, шрифт 10 pt, без абзацного відступу. Формули – створювати у редакторі формул, виключка по центру. За наявності у тексті посилання на формулу, її нумерують арабською цифрою у круглих дужках з правого краю в межах форматування сторінки. Заголовок рисунка (Приклад: *Рис. 1.* **Назва**.) – напівжирним, виключка по ширині, без абзацного відступу. На кожен рисунок потрібно робити в тексті посилання – (рис. __). Умовні позначення та підписи на рисунку – шрифт 10 pt.

Обговорення	У цьому розділі необхідно зіставити результати власних досліджень з результатами робіт інших авторів, яких цитували у вступній частині, що найбільш близькі до теми дослідження.
Висновки	Рекомендується наводити пронумеровані висновки.
Подяки	Розділ не є обов'язковим, у ньому висловлюється подяка організаціям за посильну технічну допомогу; ідеї; фінансову підтримку, завдяки якому дослідження стало можливим.
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