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НАУЧНЫЙ ЖУРНАЛ
**СИБИРСКИЙ ВЕСТНИК
СЕЛЬСКОХОЗЯЙСТВЕННОЙ НАУКИ**
SIBIRSKII VESTNIK SEL'SKOKHOZYAISTVENNOI NAUKI

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СОДЕРЖАНИЕ

CONTENTS

*ЗЕМЛЕДЕЛИЕ
И ХИМИЗАЦИЯ*

*AGRICULTURE
AND CHEMICALIZATION*

Власенко Н.Г., Бурлакова С.В., Егорычева М.Т. Эффективность обработки семян яровой пшеницы Триходермином и Споробактерином

5 Vlasenko N.G., Burlakova S.V., Egorycheva M.T. Efficiency of spring wheat seeds treatment with Trichodermin and Sporobacterin

РАСТЕНИЕВОДСТВО И СЕЛЕКЦИЯ

PLANT GROWING AND BREEDING

Исачкова О.А., Логинова А.О., Коркина В.И. Содержание аминокислот в зерне голозерного овса при различных условиях возделывания

15 Isachkova O.A., Loginova A.O., Korkina V.I. Amino acid content in the grain of naked oats under various cultivation conditions

Марченко Л.А. Признаки качества плодов земляники садовой и селекция на их улучшение

24 Marchenko L.A. Quality attributes of garden strawberry fruits and breeding for their improvement

ЗАЩИТА РАСТЕНИЙ

PLANT PROTECTION

Мороховец Т.В., Мороховец В.Н., Маркова Е.С., Басай З.В., Вострикова С.С., Скорик Н.С. Фазовая чувствительность некоторых видов широколистных сорных растений к гербициду Флекс

32 Morokhovets T.V., Morokhovets V.N., Markova E.S., Basai Z.V., Vostrikova S.S., Skorik N.S. Phase sensitivity of some broad-leaved weed species to the herbicide Flex

Баскаков И.В., Орбинский В.И., Василенко В.В., Гиевский А.М. Озонная дезинсекция зерна от амбарного долгоносика и булавоусого хрущака

42 Baskakov I.V., Orobinsky V.I., Vasilenko V.V., Gievsky A.M. Ozone disinfection of grain from granary weevil and confused flour beetle

-
- Сурначева В.В., Казанцев М.П., Коробейников А.С., Ашмарина Л.Ф.** 49 **Surnacheva V.V., Kazancev M.P., Korobejnikov A.S., Ashmarina L.F.** Effect of temperature and humidity on the composition of wheat phytopathogens during grain storage
- Влияние температуры и влажности на состав фитопатогенов пшеницы при хранении зерна

*ЗООТЕХНИЯ
И ВЕТЕРИНАРИЯ*

*ZOOTECHNICS
AND VETERINARY MEDICINE*

- Позовникова М.В., Лейбова В.Б.** 56 **Pozovnikova M.V., Leibova V.B.** Exterior profile of Saanen goats with different genotypes of the *SPAG17* gene
- Экстерьерный профиль зааненских козлов с различными генотипами гена *SPAG17*
- Забелина М.В., Ледяев Т.Б., Корнилова В.А., Ловцова Л.Г., Преображенская Т.С.** 64 **Zabelina M.V., Ledyayev T.B., Kornilova V.A., Lovtsova L.G., Preobrazhenskaya T.S.** Evaluation of milk productivity and milk quality of goats of different genotypes depending on the number of lactations
- Оценка молочной продуктивности и качества молока коз разных генотипов в зависимости от числа лактаций
- Третьяков А.М.** 72 **Tretyakov A.M.** Parasite cenoses of the wild pig (*Sus scrofa*) on the Trans-Baikal Territory
- Паразитоценозы дикой свиньи (*Sus scrofa*) на территории Забайкальского края
- Лопсан Ч.О.** 79 **Lopsan Ch.O.** Dynamics and peculiarities of anthrax occurrence on the territory of the Republic of Tyva
- Динамика и особенности проявления сибирской язвы на территории Республики Тыва
- Ефремова Е.А., Марченко В.А., Смертина М.А.** 89 **Efremova E.A., Marchenko V.A., Smertina M.A.** Distribution of helminths of the gastrointestinal tract of horses in Central Altai
- Распространение гельминтов желудочно-кишечного тракта лошадей в Центральном Алтае
- Савельева Л.Н., Бондарчук М.Л.** 98 **Savelyeva L.N., Bondarchuk M.L.** The effect of phytobiotic preparations on morphochemical blood parameters of calves with dyspepsia
- Влияние фитобиотических препаратов на морфохимические показатели крови телят при диспепсии

МЕХАНИЗАЦИЯ, АВТОМАТИЗАЦИЯ,
МОДЕЛИРОВАНИЕ
И ИНФОРМАЦИОННОЕ ОБЕСПЕЧЕНИЕ

MECHANISATION, AUTOMATION,
MODELLING AND DATAWARE

- Алейников А.Ф.** Методы неинвазивной оценки полового диморфизма эмбрионов в яйце птицы **105** **Aleynikov A.F.** Methods for noninvasive assessment of sexual dimorphism of embryos in the poultry egg
- Чысыма Р.Б., Самбыла Ч.Н.** Проблемы и перспективы цифровизации сельского хозяйства в Республике Тыва **117** **Chysyma R.B., Sambyla Ch.N.** Problems and prospects of digitalization of agriculture in the Republic of Tyva

КРАТКИЕ СООБЩЕНИЯ

BRIEF REPORTS

- Федина Л.А., Маркова Т.О., Маслов М.В.** Новые местонахождения декоративной орхидеи *Spiranthes sinensis* (Orchidaceae) в Приморском крае **124** **Fedina L.A., Markova T.O., Maslov M.V.** New locations of the decorative orchid *Spiranthes sinensis* (Orchidaceae) in the Primorsky Territory

ИЗ ДИССЕРТАЦИОННЫХ РАБОТ

FROM DISSERTATIONS

- Павлов А.Г.** Амилолитическая активность изолятов бактерий *Bacillus subtilis*, выделенных из микробиоты диких животных **130** **Pavlov A.G.** Amylolytic activity of *Bacillus subtilis* isolates obtained from wildlife microbiota

НАУЧНЫЕ СВЯЗИ

SCIENTIFIC RELATIONS

- Ержебаева Р.С., Бабисекова Д.И., Дидоренко С.В.** Использование ДНК-маркеров в селекции сои для отбора фотопериодически нейтральных линий **136** **Yerzhebayeva R.S., Babissekova D.I., Didorenko S.V.** The use of DNA markers in soybean breeding to select photoperiod-neutral lines



ЭФФЕКТИВНОСТЬ ОБРАБОТКИ СЕМЯН ЯРОВОЙ ПШЕНИЦЫ ТРИХОДЕРМИНОМ И СПОРОБАКТЕРИНОМ

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Представлены результаты исследования по определению эффективности предпосевной обработки семян среднераннего сорта Новосибирская 31 биопрепаратами. Научный опыт проведен в условиях лесостепи Приобья в 2019–2021 гг. Изучено влияние биопрепаратов на развитие грибных заболеваний пшеницы и основные параметры посева: густоту стояния растений, их высоту и биомассу, площадь флагового листа, структуру колоса и урожайность зерна. При нарастании развития корневой гнили в течение вегетации от 1,3 до 3,4% к фазе кущения и до 10,0% к концу вегетации эффективность обработки семян биопрепаратами Триходермин, Споробактерин и протравителем Скарлет составила 32, 53 и 56% в начале вегетации и 21, 27 и 36% – в фазе молочно-восковой спелости зерна. Препараты Триходермин, Споробактерин проявляли среднюю эффективность в отношении септориоза – 40 и 34%, против мучнистой росы – 29 и 24%, протравитель Скарлет подавлял листовые инфекции на 51 и 43% при их развитии в контроле 9,3 и 9,0%. Длина ростков пшеницы в фазе 2-го листа в вариантах применения Триходермина, Споробактерина, Скарлет была больше, чем в контроле, на 7,6; 11,1 и 4,6%. Наибольший ростостимулирующий эффект наблюдали при обработке семян Споробактерином. В фазе молочно-восковой спелости зерна густота стояния растений пшеницы увеличилась в сравнении с контролем на 8,3; 21,7 и 15,2% соответственно, продуктивный стеблестой был выше при применении биопрепаратов на 15,2%, протравителя Скарлет – на 17,4%. Сбор зерна увеличился относительно контроля (2,31 т/га) в вариантах Триходермин и Споробактерин на 0,30 и 0,37 т/га, Скарлет – на 0,22 т/га. В результате обработки семян препаратом Скарлет получено зерно с содержанием белка 13,25%, при применении биофунгицидов Триходермин, Споробактерин оно повышалось на 0,14 и 0,28% относительно контроля (13,3%). В результате проведенных исследований показано, что биопрепараты Триходермин и Споробактерин при относительно невысоком развитии болезней способны сдерживать их развитие, немного уступая химическому препарату. Обладая ростостимулирующими свойствами, они могут обеспечить урожайность зерна даже выше, чем при применении протравителя.

Ключевые слова: яровая пшеница, биопрепараты, обработка семян, болезни, ростовые эффекты, урожайность

EFFICIENCY OF SPRING WHEAT SEEDS TREATMENT WITH TRICHODERMIN AND SPOROBACTERIN

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The results of the study to determine the effectiveness of pre-sowing treatment of seeds of medium-early variety Novosibirskaya 31 with biopreparations are presented. The scientific experiment was conducted in the forest-steppe conditions of the Priob'ye region in 2019–2021. The

effect of biological preparations on the development of fungal diseases of wheat and on the main parameters of sowing: the density of plants, their height and biomass, flag leaf area, ear structure and grain yield were studied. With the increase of root rot during the growing season from 1,3 to 3,4% to the phase of bushing and up to 10,0% by the end of the growing season the efficiency of seed treatment with Trichodermin, Sporobacterin and Scarlet bio-detergent was 32, 53 and 56% at the beginning of the growing season and 21, 27 and 36% - in the phase of milk-wax ripeness of grain. Trichodermin and Sporobacterin preparations were moderately effective against septoriossis (40 and 34%), against powdery mildew (29 and 24%), and the disinfectant Scarlet suppressed leaf infections by 51 and 43% against 9.3% and 9.0% of the control. The length of the wheat sprouts in the phase of the 2nd leaf in the variants of Trichodermin, Sporobacterin, Scarlet was greater than the control by 7.6; 11.1 and 4.6%. The greatest growth-stimulating effect was observed when the seeds were treated with Sporobacterin. In the phase of milk-wax ripeness the density of the wheat plants increased compared to the control by 8,3, 21,7 and 15,2% respectively, the productive stem structure was higher when using biopreparations by 15,2%, Scarlet dressing - by 17,4%. The grain harvest increased relative to the control (2.31 t/ha) in Trichodermin and Sporobacterin variants by 0.30 and 0.37 t/ha, Scarlet - by 0.22 t/ha. As a result of seed treatment with Scarlet the grain was obtained with protein content of 13,25%, while application of biofungicides Trichodermin, Sporobacterin increased it by 0,14 and 0,28% relative to the control (13,3%). As a result of the studies, it has been shown that the biopreparations Trichodermin and Sporobacterin are able to contain the development of diseases at a relatively low level, slightly inferior to the chemical preparation. With their growth-stimulating properties, they can provide grain yields even higher than when using a dressing.

Keywords: spring wheat, biological preparations, seed treatment, diseases, growth effects, yield

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Конфликт интересов

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Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

The orientation of crop production on resource-energy saving and environmental safety involves the reduction of pesticide load in agrocenoses [1]. The main task of modern agriculture is to obtain high crop yields using biotechnology, creating conditions for the preservation of ecologically pure fertile soils. In order to preserve and reproduce soil fertility, biological agro-technologies with a consistent replacement of energy-intensive agrochemicals and pesticides by new generation biopreparations are being developed [2, 3]. The main advantage of biological preparations is environmental friendliness, safety for humans and the environment. Biological preparations have an extended period of application, they can be used at almost any stage of

plant development, as they have a short safety interval after treatment [4]. Environmentally safe biopreparations when used competently (systemic treatments, timely application, etc.) not only ensure a high yield, but also increase its quality. Interest in biological preparations is also increasing due to the fact that many of them have an anti-stress effect, increasing the resistance of plants to abiotic environmental factors [5, 6]. Despite the significant advantages of the biological method of disease control, its widespread and effective use in crop cultivation is limited by a number of factors. Living organisms form the basis of most biopreparations, so in order to obtain the desired effect it is necessary to consider the shelf life of biopreparations, strictly follow the instructions for their storage, use biopesticides only

by results of phytosanitary monitoring, consider that under adverse weather conditions (drought, cold rainy weather) the effectiveness of biopreparations is reduced by 20-50% [7].

Pre-sowing treatment of cereal crops seeds is recognized as one of the effective ways to increase grain yield. It also contributes to seed disinfection from fungal and bacterial diseases, provides increased immunity and energy of plant germination [8]. Currently, to protect crops from diseases, preparations based on fungi of the genera *Trichoderma*, *Streptomyces*, different species and strains of bacteria of *Bacillus* and *Pseudomonas* and others are most widely used [9]. Unfortunately, biological preparations are often of low quality and low efficiency due to rapid loss of bioagent activity in the natural environment because of the negative effect of environmental factors or changes in the properties of microorganisms. In the case of biopreparations, accelerated growth of root system, secondary root formation, increased germination, germination energy and bushiness were noted. Some of them increase the drought and heat-resistance of plants and activate the activity of useful microbial community of the rhizosphere. They induce natural resistance of plants to diseases, which provides an increase in grain yield [10].

The purpose of the study is to evaluate the protective and growth-stimulating effect of treatment of spring wheat seeds with biological preparations based on *Trichoderma viride* and *Bacillus subtilis* when growing crops in the Priob'ye forest-steppe.

MATERIAL AND METHODS

The studies were conducted in 2019-2021 in the experimental field of the plant protection department of the Siberian Federal Scientific Center of Agro-BioTechnologies of the Rus-

sian Academy of Sciences (SFSCA RAS) under the conditions of the northern forest-steppe of the Priob'ye region. The growth-stimulating and protective effect of seed pre-sowing treatment was studied on the crops of mid-early wheat variety Novosibirskaya 31. Biopreparations Trichodermin, P (*Trichoderma viride*, titer more than 6 billion spores/g, 80 g/t) and Sporobacterin, WP (*Bacillus subtilis* + *Trichoderma viride*, strain 4097, 0.5 kg/t) were used for the seed treatment. Scarlet, ME (imazalil (100 g/l) + tebuconazole (60 g/l), 0.3 l/t) dressing was used as a reference. Seed treatment was carried out with wetting, the rate of working solution consumption was 10 l/t seed. In a single-factor field experiment spring wheat was placed on fallow preceding, plot area 44.1 m² (2.1 × 21), arrangement sequentially in one tier, fourfold repetition, the seeding rate of 6 million germinated grains/ha. Seeding was done by SZS-2.1 seeder with anchor coulters, and ammonium nitrate was applied at the rate of N₆₀ per 1 ha. Background spraying against cereal and dicotyledonous weeds was carried out in the phase of wheat tillering with a tank mixture of herbicides Axial, EC (1 l/ha) + Primadonna, SE (0.4 l/ha) + Hextar, WDG (10 g/ha). The working fluid consumption rate is 270 l/ha. The effect of biofungicides on the development and prevalence of common root rot during the phases of the third leaf, tillering, and milk-wax maturity of grain, as well as leaf infections - septoriose, powdery mildew, and brown leaf rust - during the phase of the beginning of milk maturity of grain were studied^{1,2}. The parameters of sowing were taken into account: plant density, total and productive bushiness, biomass, plant height, flag leaf area, ear structure, grain size^{3,4}. The yield was counted by direct harvesting with Sampo harvester, the grain was reduced to 14% moisture

¹Chulkina V.A., Toropova E.Yu., Stetsov G.Ya., Marmuleva E.Yu., Kirichenko A.A., Grishin V.M. Phytosanitary diagnostics of agroecosystems: tutorial. Ed. by E.Yu. Toropova. Novosibirsk. 2010. 127 p.

²Sanin S.S., Sokolova E.A., Cherkashin V.I. Diseases of grain crops (recommendations on phytosanitary monitoring). Moscow: Rosinformagrotech. 2010. 137 p.

³Toropova E.Yu., Kirichenko A.A. Phytosanitary ecological monitoring: method. instructions for laboratory and practical exercises and control work. Novosibirsk: NSAU, 2012. 38 p.

⁴Valeeva A.A., Sakhabiev I.A. Diary of training practice in agrogenology. Kazan: Kazan University, 2017. 26 p.

content and 100% purity, the amount of protein in the grain was determined. Statistical data processing was performed using Snedecor and Excel applications⁵.

Weather conditions varied significantly in the years of study. In general, according to meteorological indicators, 2019 was unfavorable for the formation of high plant productivity. A total of 189 mm of precipitation fell during the growing season, less than the norm by 43 mm, with 43 mm of atmospheric moisture coming in May, 26 mm in June, 98 mm in July, and 22 mm in August. Average daily temperatures were 10.9, 16.4, 19.2 and 18.3 °C, respectively. According to meteorological indicators, the vegetation period of 2020 was characterized by high heat supply and sufficient moisture content. The sum of precipitation for vegetative period from May till August amounted to 245 mm, their distribution on months was extremely irregular: in May it was 54,0 mm, in June - 24,0 mm, in July - 85,0 mm, in August - 82,0 mm. Average daily air temperatures were 16.5; 16.6; 19.6 and 18.6 °C, respectively. In 2021 air temperatures were close to average annual values: in May, June, July and August they were 14.3; 16.2; 19.6 and 18.1 °C. Precipitation was 45 mm less than the long-term

average, with a total of 187 mm during the growing season. Precipitation was 25.0 mm in May, 73.0 mm in June, 22.0 mm in July and 67.0 mm in August.

RESULTS AND DISCUSSION

Root rot development both at the beginning of vegetation and by the phase of milk-wax maturity of grain was not high and averaged for 3 years 1.3% in the phase of 3-4 leaves, 3.4% - in tillering and 10% - in the phase of milk-wax maturity of grain, varying by years from 1.1-1.5; 2.7-4.8 and 8.9-11.6%, respectively. Treatment of spring wheat seeds had an effect on plant infestation of root rot (see Fig. 1). Biological efficacy of Sporobacterin averaged 38.5 and 52.9% by year, Trichodermin 23.1 and 32.3% in the first two phases of recording, decreasing to 27.0 and 21.0% by the end of the crop vegetation, being inferior to Scarlet, 46.1; 55.9 and 36.0% respectively. Biological preparations did not suppress disease incidence, which was 98-100%, Scarlet chemical reference reduced this indicator only in the phase of the 4th leaf up to 88% and had almost no effect on it in subsequent phases.

The studied agents also suppressed leaf infections. In the phase of the beginning of milk

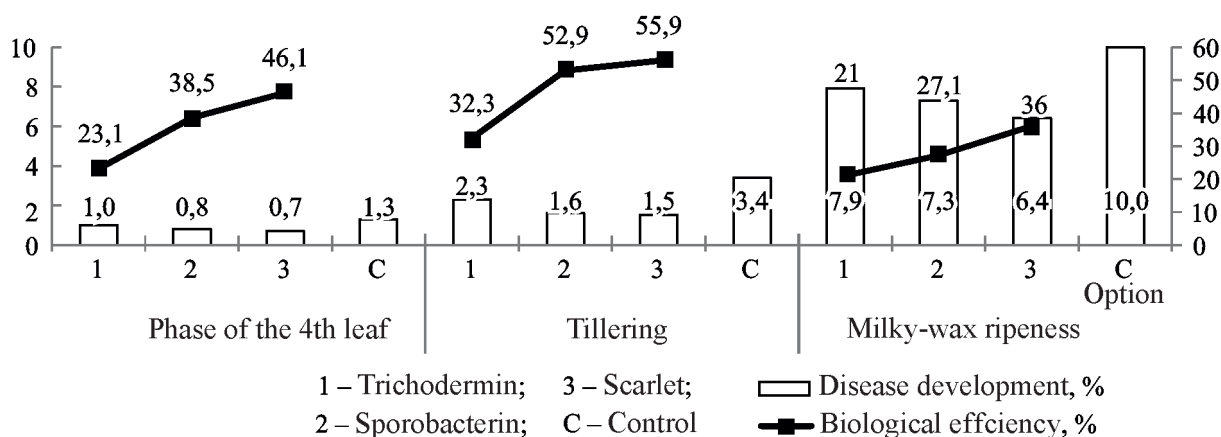


Рис. 1. Эффективность обработки семян биопрепаратами против корневой гнили яровой пшеницы (2019–2021 гг.), %

Fig.1. Effectiveness of seed treatment with biopreparations against spring wheat root rot (2019–2021), %

⁵Sorokin O.D. Applied statistics on computer; 2nd ed. Novosibirsk, 2012. 282 p.

ripening of grain with septoriose development in the control, the efficiency of Trichodermin and Sporobacterin biofungicides was 9,3% (14,2; 8,4 and 5,4% respectively, in 3 years of research); the efficiency of Scarlet dressing was 40 and 34%. At 97% septoriose prevalence in the control, it was reduced in the variants with bioformulations to 91 and 95%; the lowest was in the Scarlet variant - 88% (see Fig. 2).

The efficacy of the preparations Trichodermin, Sporobacterin and Scarlet against powdery mildew in the development of the disease in the control at an average of 9.0% for 3 years (10.8; 5.6 and 10.5% respectively years of research) was 29, 24 and 43%, the develop-

ment was reduced to 5.1-6.8%. The efficacy of seed treatment with Trichodermin and Scarlet was 22% and Sporobacterin 9% against leaf rust with a 2.3% control (6.1; 0.4 and 0.5%, respectively, in the years of the study). Prevalence of powdery mildew and leaf rust in the control were 75 and 34%; in the experiment it varied from 71 to 77% and from 38 to 47%, respectively.

The effect of the preparations on the formation of wheat sprouts was observed in the experiment (see Table 1). In the variants with treatment of seeds with Trichodermin and Sporobacterin, the leaf length significantly increased relative to the control by 9.3; 12.3%, while with Scarlet dressing application - by

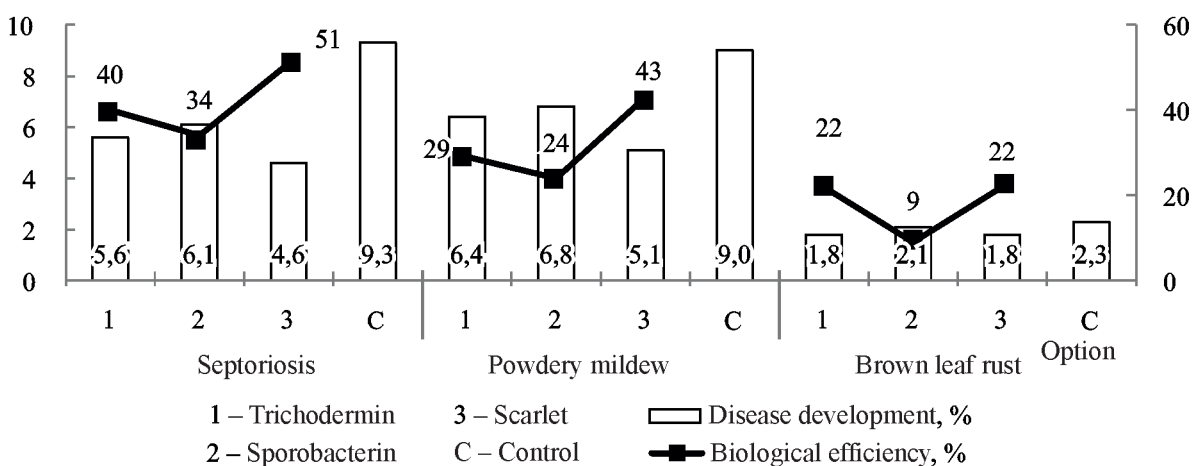


Рис. 2. Эффективность обработки семян биопрепаратами против листовых болезней яровой пшеницы в фазе начала молочной спелости зерна (2019–2021 гг.), %

Fig. 2. Efficiency of seed treatment with biopreparations against leaf diseases of spring wheat in the phase of the beginning of milk maturity of grain (2019–2021), %

Табл. 1. Морфологические показатели проростков пшеницы Новосибирская 31 в фазе 2-го листа при обработке семян биопрепаратами (2019–2021 гг.), см

Table 1. Morphological indices of Novosibirskaya 31 wheat seedlings in the phase of 2nd leaf when treated with biopreparations (2019–2021), cm

Option	Length			
	leave	stem	sprout	germinal roots
Control	11,41	4,63	16,04	5,31
Trichodermin	12,12	5,14	17,26	4,52
Sporobacterin	12,82	5,00	17,82	4,76
Scarlet	11,78	5,00	16,78	4,48
LSD ₀₅	0,32	0,18	0,50	1,30

3.2%. Stem length increased by 11.0; 7.9 and 7.9%, sprout length by 7.6; 11.1 and 4.6%. The greatest growth-stimulating effect was observed when the seeds were treated with Sporobacterin, the smallest - when a chemical dressing was used.

The inhibition of root system growth in the variants with biopreparations by 14.9 and 10.4%, and even greater - in the Scarlet variant by 15.6% relative to the control index was noted. The degree of inhibition of the root system of the seedlings at the level of 10-16% is an indicator characterizing the level of phytotoxicity of the soil, which in this case is low (inhibition is less than 21-30%). Seed treatment with biofungicides had a positive effect on the density of productive plant stand. On the average for the years of study, the variants Trichodermin, Sporobacterin and Scarlet increased the latter by 9,7%, 21,9% and 16,5% relatively to the control in the phase of the third leaf, and

by 8,3%, 21,7% and 15,2% respectively in the phase of milk-wax ripeness of grain. The number of surviving plants in these variants was 78-79% (see Table 2).

The number of plants per unit area for harvesting by 87-93% determined the density of the productive plant stand, which increased with the use of biopreparations by 15.2%, and Scarlet dressing - by 17.4%.

Of all seed treatments, the most active above-ground biomass of the crop by the end of the growing season was accumulated when treating the seeds with Scarlet fungicide - by 15.2% relative to the control, the root biomass - by 22.6% respectively (see Fig. 3, 4). No increase in the above-ground biomass was observed in Sporobacterin and Trichodermin treatments relative to the control until the flowering phase, the root mass increased only when Sporobacterin was used by 4.3%.

Seed treatment had little effect on such in-

Табл. 2. Влияние обработки семян биопрепаратами на густоту стеблестоя яровой пшеницы (2019–2021 гг.)

Table 2. Effect of seed treatment with biopreparations on the stem density of spring wheat (2019–2021)

Option	Standing density, plants/m ²		Plants viability, %	Productive stems, pcs/m ²
	3rd leaf phase	Milky-wax ripeness of grain		
Control	473	373	78,9	402
Trichodermin	519	404	77,9	463
Sporobacterin	577	454	79,2	463
Scarlet	551	430	78,1	472
LSD ₀₅	11	33	-	47

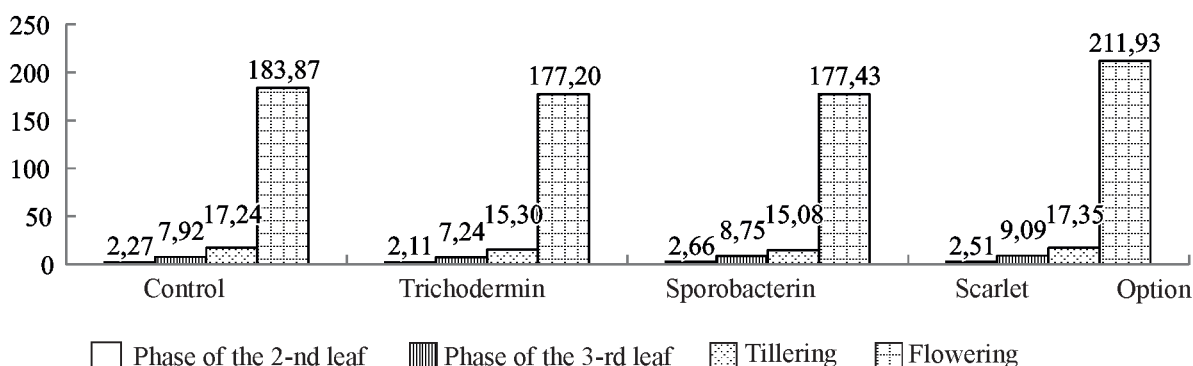


Рис. 3. Влияние обработки семян препаратами на надземную воздушно-сухую биомассу (2019–2021 гг.), г/100 растений (НСР₀₅ = 0,5; 2,4; 3,5; 4,7)

Fig.3. Effect of seed treatment on the aboveground air-dry biomass (2019–2021), g/100 plants (LSD₀₅ = 0.5; 2.4; 3.5; 4.7)

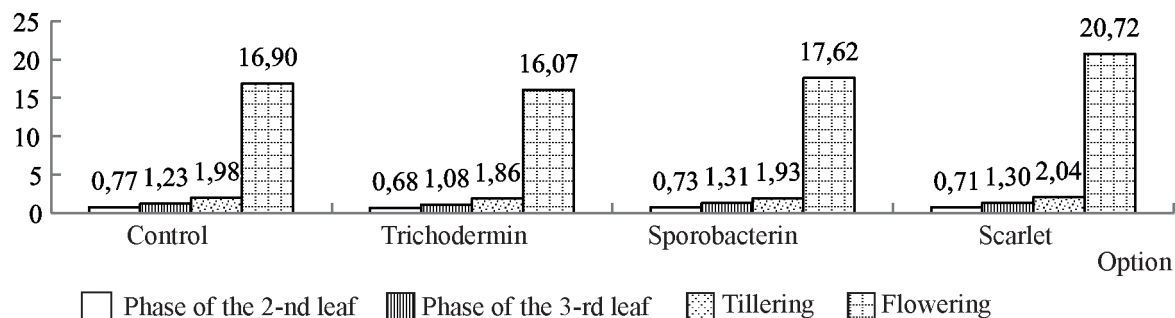


Рис. 4. Влияние обработки семян препаратами на корневую воздушно-сухую биомассу (2019–2021 гг.), г/100 растений ($HCP_{05} = 0,7; 1,3; 2,0; 2,1$)

Fig.4. Effect of seed treatment with preparations on the root air-dry biomass (2019–2021), g/100 plants ($LSD_{05} = 0.7; 1.3; 2.0; 2.1$)

dicator as plant height (see Table 3). In Trichodermin, Sporobacterin and Scarlet the plants height increased by 2,7; 2,2 and 3,6% relatively to the control. Thickening of the stems was noted in Trichodermin and Scarlet - by 4.4 and

2.2% relative to the control, in the variant with Sporobacterin thinning was noted by 4.4%, probably due to the higher density of standing plants in this variant.

Табл. 3. Влияние обработки семян препаратами на параметры посева яровой пшеницы в фазе цветения (2019–2021 гг.)

Table 3. Effect of seed treatment with preparations on sowing parameters of spring wheat in the flowering phase (2019–2021)

Option	Plant height, cm	Stem thickness, mm
Control	82,6	4,5
Trichodermin	84,8	4,7
Sporobacterin	84,4	4,3
Scarlet	85,6	4,6
LSD_{05}	2,9	0,2

Growth-stimulating effect of the preparations on the flag leaf area index was observed (see Fig. 5). In the experiment, the growth relative to the control was 9.8; 8.8 and 4.8% in the Trichodermin variant, according to the years of research, Scarlet - 11.4 and 15.2% in 2019 and 2020, the treatment of seeds with Sporobacterin increased the flag leaf area by 17.1% only in 2019.

Seed treatment had an effect on the ear formation, grain weight and wheat yield (see Table 4).

The largest increase in comparison with the control was in the variant with Trichodermin

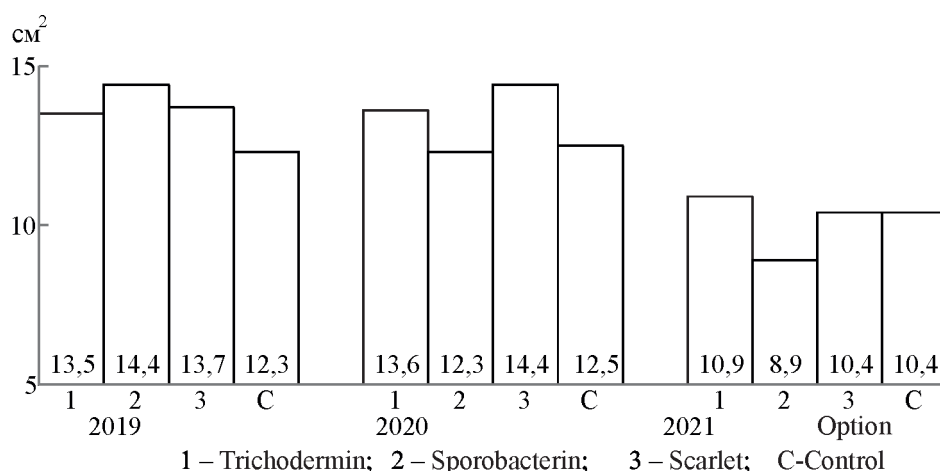


Рис. 5. Влияние обработок семян и посевов препаратами на площадь флагового листа (2019–2021 гг.), cm^2 ($HCP_{05} = 2,20; 1,62; 1,28$)

Fig. 5. Effect of seed and crop treatments on the flag leaf area (2019–2021), cm^2 ($LSD_{05} = 2.20; 1.62; 1.28$)

Табл. 4. Влияние обработки семян биофунгицидами на структуру урожая и урожайность яровой пшеницы (2019–2021 гг.)**Table 4.** Effect of seed treatment with biofungicides on the yield structure and spring wheat yield (2019–2021)

Option	Spike				TKW, g	Yield, t/ha	Protein content, %
	length, cm	number of spikelets	number of grains	grain mass, g			
Control	8,77	15,37	30,20	0,90	29,36	2,31	13,40
Trichodermin	9,69	16,41	35,92	1,12	30,27	2,61	13,54
Sporobacterin	8,92	15,77	30,66	0,94	30,52	2,68	13,68
Scarlet	9,21	15,69	33,29	1,01	30,40	2,53	13,25
LSD ₀₅	0,39	1,02	4,17	0,17	0,49	0,12	1,22

- by 10,5; 6,8; 18,9; 24,4%, respectively, the smallest - in the variant with Sporobacterin - by 1,7; 2,6; 1,5; 4,4%, in the variant with Scarlet - by 5,0; 2,1; 10,2; 12,2%. Seed treatment with the studied preparations contributed to the formation of larger grains. The highest thousand kernel weight was obtained when Sporobacterin was used - 1.16 g higher than in the control.

A significant increase in wheat yield was observed in all variants of the experiment. Seed treatment with biological preparations provided an average of 0.3-0.37 t/ha additional grain yield; with a chemical dressing - by 0.22 t/ha. The highest yield was obtained when the seeds were treated with Sporobacterin.

Grain protein content was higher relative to the control when using Trichodermin and Sporobacterin by 0.14 and 0.28%, in the Scarlet variant it decreased by 0.15%.

CONCLUSIONS

1. In cases of weak disease development, typical for the forest-steppe zone of Priob'ye region, chemical fungicides can be replaced by the biological ones. It has been established that the efficacy of the preparations Trichodermin, Sporobacterin, Scarlet in the phase of tillering reduces the development of root rot by 32, 53, 56% and by 21, 27, 36% respectively in the phase of milk-wax maturity of grain when treating the seeds of spring wheat of Novosibirskaya 31 variety placed on a fallow forecrop. In the earing phase of wheat Trichodermin and Sporobacterin reduce the develop-

ment of septoriose by 40 and 34%, powdery mildew by 29 and 24%, and Scarlet by 51 and 43%.

2. The growth stimulating effect on the development of the crop was manifested in the increase of plant stand density in Trichodermin, Sporobacterin and Scarlet variants by 9.7; 21.9 and 16.5% relative to the control. Seed treatment had a growth stimulating effect on the flag leaf of wheat. An increase in the leaf area index relative to the control was observed when seeds were treated with the above preparations by 7.9; 1.7 and 8.6%.

3. The protective effect of biological preparations and their growth-stimulating effect on wheat plants provided a significant increase in the yield when seeds were treated with Trichodermin and Sporobacterin by 0.30 and 0.37 t/ha; when using Scarlet dressing the yield increased by 0.22 t/ha.

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СОДЕРЖАНИЕ АМИНОКИСЛОТ В ЗЕРНЕ ГОЛОЗЕРНОГО ОВСА ПРИ РАЗЛИЧНЫХ УСЛОВИЯХ ВОЗДЕЛЫВАНИЯ

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Представлены результаты изучения формирования урожайности и содержания аминокислот в зерне сортов голозерного овса разных групп спелости под влиянием различных сроков посева. Исследования проведены в 2018–2020 гг. в полевом и лабораторном опытах на среднераннем сорте Гаврош и среднеспелом сорте Офеня в условиях Западной Сибири. Выявлено достоверное преимущество ранних сроков посева сортов голозерного овса: у сорта Гаврош в среднем на 25,9–29,6%, у сорта Офеня на 16,2–21,6% относительно более поздних сроков. При этом достоверно более высокая урожайность формировалась у среднеспелого сорта Офеня (на 0,1–0,4 т/га). Большее количество незаменимых аминокислот у сорта Гаврош отмечено при позднем сроке посева – 4,51% (при раннем и среднем сроках – 4,39 и 4,45% соответственно), заменимых аминокислот – при раннем сроке – 8,83% (при среднем и позднем – 7,80 и 8,46% соответственно). У сорта Офеня содержание незаменимых аминокислот составило 4,82–6,49%, заменимых – 7,28–9,49%. У данного сорта отмечена тенденция увеличения количественного состава аминокислот в зерне от раннего срока посева к позднему. Выявлены различия по влиянию условий влагообеспеченности на накопление аминокислот в сортах голозерного овса. Наиболее высокое содержание незаменимых и заменимых аминокислот у среднеспелого сорта Офеня формируется при повышенных значениях гидротермического коэффициента (ГТК) в период всходы – кущение ($r = 0,9467 \dots 0,9999$ при $R = 0,6660$), пониженных значениях в период выход в трубку – цветение ($r = -0,9338 \dots -0,9987$ при $R = 0,6660$), при повышении ГТК в период налив – созревание ($r = 0,4335 \dots 0,7888$ при $R = 0,6660$). У раннеспелого сорта Гаврош большее содержание незаменимых аминокислот отмечено при раннем сроке посева при низких значениях ГТК в период всходы – кущение, повышенных – в период выход в трубку – цветение и в засушливых условиях периода налив – созревание ($r = -0,8812, 0,8626, -0,6087$ при $R = 0,6660$ соответственно). Высокое содержание заменимых аминокислот у сорта Гаврош формировалось при поздних сроках в годы с пониженными значениями ГТК в период всходы – кущение, наличии осадков в период цветения и их отсутствии в период налив – созревание ($r = -0,8287, 0,8068, -0,6860$ при $R = 0,6660$ соответственно).

Ключевые слова: голозерный овес, урожайность, срок посева, аминокислоты

AMINO ACID CONTENT IN THE GRAIN OF NAKED OATS UNDER VARIOUS CULTIVATION CONDITIONS

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The results of studying the formation of yield and amino acid content in the grain of naked oats varieties of different ripeness groups under the influence of different sowing dates are presented. The research was carried out in 2018-2020 in field and laboratory experiments on the medium early Gavroche variety and the mid ripening Ofenya variety in Western Siberia. A significant advantage of early sowing dates of naked oats varieties has been revealed: the Gavroche variety has an average of 25.9-29.6%, the Ofenya variety has 16.2-21.6% relative to later dates. At the same time, a significantly higher yield was formed in the mid ripening variety Ofenya, by 0.1-0.4 t/ha. A greater number of essential amino acids in the Gavroche variety was noted at a late sowing period – 4.51% (at an early and average term of 4.39 and 4.45%, respectively), and interchangeable amino acids – at an early term – 8.83% (at an average and late – 7.80 and 8.46%, respectively). In the Ofenya variety, the content of essential amino acids was 4.82-6.49 g/kg, interchangeable 7.28-9.49%. At the same time, this variety has a tendency to increase the quantitative composition of amino acids in the grain from the early sowing period to the late one. Differences in the influence of moisture conditions on the accumulation of amino acids in the varieties of naked oats were revealed. The highest content of essential and interchangeable amino acids in the medium-ripened variety Ofenya is formed at elevated values of the hydrothermal coefficient (HTC) in the period of seedling – tillering ($r = 0.9467 \dots 0.9999$ at $R = 0.6660$), reduced values in the period of booting – flowering ($r = -0.9338 \dots -0.9987$ at $R = 0.6660$), with an increase in HTC during the filling – ripening period ($r = 0.4335 \dots 0.7888$ at $R = 0.6660$). In the early-maturing Gavroche variety, a higher content of essential amino acids was noted at an early sowing period under conditions of low values of HTC during the period of seedling – tillering, increased - during the period of booting-flowering and arid conditions of the period of filling–ripening - $r = -0.8812, 0.8626, -0.6087$ at $R = 0.6660$, respectively. The high content of interchangeable amino acids in the Gavroche variety was formed at late periods in the years with reduced values of HTC during the period of seedling -tillering, the presence of precipitation during flowering and their absence during the period of filling-ripening ($r = -0.8287, 0.8068, -0.6860$ at $R = 0.6660$, respectively).

Keywords: naked oats, yield, sowing period, amino acids

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Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

Naked oats are a unique crop with unique properties. Rich biochemical composition of the grain, which determines its dietary and therapeutic and preventive properties, determines the possibility of its use in the development of products for children's, functional and specialized purposes [1, 2]. Naked oats grains are rich in proteins (up to 18-20%), oil (up to 8-12%) and water-soluble beta-glucans (up to 6-8%). Oats are also rich in micro- and macroelements (potassium, magnesium, calcium, silicon, phosphorus, sodium, etc.). The content of vitamins A, B, E, K of naked oats grains is high [3-8].

Naked oats contain an almost complete balanced complex of essential and non-essential amino acids [9-11]. Amino acids are present in all plant tissues. They play an important role in metabolism, many of them are activators of enzymes and vitamins. The composition of amino acids affects the quality of food and feed. Their deficiency causes serious diseases in humans and animals. Amino acids are the end product of protein breakdown in the digestive tract. They are the structural material for the formation of proteins in the human and animal body. Eight amino acids - valine, isoleucine, leucine, lysine, methionine, threonine, tryptophan and phenylalanine - are essential and in the absence of at least one of them protein synthesis is impossible. These amino acids are not synthesized in the body and must be supplied in sufficient amounts with food. Lysine, methionine and tryptophan are basic or critical, since they limit the use of other amino acids for protein molecule synthesis [12]. Histidine and arginine are conditionally essential, they are formed in humans and animals, but in small quantities, and most of the need for these amino acids must be covered by food sources. Histidine is essential for the development and maintenance of healthy tissues; isoleucine contributes to normal blood clotting and muscle repair; leucine enhances the production of growth hormone and promotes muscle growth; lysine is involved in the production of collagen, which is crucial for

bone health; Methionine is a powerful antioxidant; phenylalanine helps produce stress and relaxation hormones; threonine is necessary for bone and cartilage formation; tryptophan is necessary for serotonin and melatonin production; valine prevents muscle breakdown and removes excess protein from the liver arginine is responsible for muscle repair, fast healing of wounds and injuries, removes toxins, strengthens the immune system; alanine is responsible for blood sugar levels; asparagine helps the immune system function; glutamine is "fuel" for the body during especially high loads, it strengthens memory, increases attention glycine - "raw material" for creating creatine, important for maintaining vitality; proline is necessary for connective tissue, fuels the body during exercise; serine is important for the nervous system, supplies cells with energy; tyrosine is able to adjust neurophysiological processes, such as attention, energy, mood, memory, vigilance [13]. The composition of amino acids in grain depends on the variety, farming technique, and growing conditions.

The purpose of the research is to study the composition of amino acids in the grain of naked-grain oats under different cultivation conditions in the northern forest-steppe of the Kemerovo region.

MATERIAL AND METHODS

The field studies were conducted in the experimental field of the Kemerovo Scientific-Research Institute of Agriculture - branch of the Siberian Federal Scientific Center of Agro-BioTechnologies of the Russian Academy of Sciences (Kemerovo NIISKh - branch of SF-SCA RAS) in 2018-2020. The research objects were varieties of naked oats: medium-early variety Gavrosh and medium-maturing variety Ofenya.

The forecrop was pure fallow. Sowing was conducted in three periods: the first (early) period - upon the appearance of soil physical ripeness (27.04-14.05), subsequent with an interval of 8-10 days: medium (09-21.05) and late (21-28.05). Sowing was carried out

using a CH-10 Ts seeder, the plot area 10 m² in fourfold repetition. Arrangement of the plots was randomized. Record-keeping, observations, and statistical processing of data were conducted in accordance with the approved guidelines¹⁻³.

Meteorological conditions in 2018 were characterized by low air temperatures and a large amount of precipitation during the seedling-ear formation period (HTC = 2.0). Excessive moisture and lack of sunshine during the tillering - flowering period contributed to significant development of leaf diseases. Grain ripening and maturing took place under optimal moisture supply and elevated air temperatures (HTC = 1.4-1.9). Weather conditions at the middle and late sowing dates were relatively favorable for the growth and development of naked oat plants. Grain filling and ripening at all terms of sowing took place at elevated air temperatures and lack of moisture (HTC = 0.3-0.7).

Weather conditions of the vegetation period of plants of the first sowing term 2019 were characterized by sufficient moisture supply. The vegetative phase of vegetation (seedling - ear formation) took place under sufficient moisture supply and elevated air temperatures (HTC = 1,1-1,3). Grain filling and ripening took place under favorable conditions (HTC = 1,1-1,2), which contributed to the formation of high yield. Water and temperature regimes at mid- and late sowing were characterized by lower air temperatures against the background of high precipitation (HTC = 1.7 and 1.8, respectively). Vegetative and generative subperiods were relatively favorable for growth and development of naked oat plants (HTC = 1.2-1.3).

The first half of vegetation of plants of the first sowing term 2020 took place under insufficient moisture supply and increased air temperatures (HTC = 0.7). Grain filling and ripening took place under drought conditions (HTC

= 0,2). Meteorological conditions during sowing - seedling and seedling - ear formation at the average sowing period were characterized by insufficient moisture supply (HTC = 1,1 and 0,9 accordingly), the ear formation - ripening period took place under excess moisture conditions (HTC = 1,8), which affected the reduction of the yield of variants. Water and temperature regimes of late period during sowing - seedling and seedling - ear formation were characterized by low air temperatures on the background of large amount of precipitation (HTC = 2,4 and 2,0 respectively), during the ear formation - ripening period increased air temperatures and the lack of moisture (HTC = 0,8) were noted.

Determination of amino acids in the grain of naked oats was carried out in the SFSCA RAS laboratory of biochemistry using capillary electrophoresis system "KAPEL" according to the M 04-87-2016 method. The method is based on sample decomposition by acid or alkaline hydrolysis with conversion of amino acids into free forms, obtaining FTC-derivatives and their further separation and quantification.

RESULTS AND DISCUSSION

In the formation of quantitative and qualitative indicators of the naked oat grain the conditions of plant growth which can be leveled by such element of cultivation technology as the sowing period are of paramount importance. The results of the study noted a higher yield of the studied varieties Gavrosh and Ofenya in 2019 - 2.89 and 3.07 t / ha at the optimum moisture supply (HTC = 1.2). The lowest yield is formed in 2020 (HTC = 1.0) - 2.37 and 2.74 t/ha, respectively.

The advantage of early sowing in both varieties in all years of the study was noted in grain productivity: in the Gavrosh variety on average by 25,9-29,6%, in the Ofenya variety by 16,2-21,6% relative to later dates (see Ta-

¹Methodology of state variety testing of agricultural crops. M., 1985. 270 p.

²Dospekhov B.L. Methodology of field experiment. Moscow: Agropromizdat, 1985. 352 p.

³Sorokin O.D. Applied statistics on computer. Krasnoobsk: SUE RPA SB RAAS, 2004. 162 p.

ble 1). At the same time, significantly higher yield was formed in the medium-ripening variety Ofenya (by 0.1-0.4 t/ha).

The value of naked oat genotypes is determined by the quality indicators of grain. Thus, according to the total content of amino acids in grain in the Gavrosh variety higher rates were found at the early sowing date - 13.22%, but differences in the essential and non-essential amino acids were observed. The greater amount of essential amino acids was observed at a late sowing date - 4,51% (at early and medium sowing dates 4,39 and 4,45% respectively), and the non-essential amino acids - at an early date - 8,83% (at a medium and late date - 7,80 and 8,46% respectively) (see Table 2).

Medium-maturing variety Ofenya differs from the medium-early variety Gavrosh by a higher content of both essential (4.82 - 6.49%) and non-essential (7.28-9.49%) amino acids. At the same time in this variety there was a tendency to increase the quantitative composition of amino acids in the grain from the early sowing date to the late sowing date. As for the sum of critical amino acids, large values were recorded for the late sowing period in both varieties - 0,69 g/kg in the Gavrosh variety, 0,95% in the Ofenya variety. From the group of essential amino acids in the variety Gavrosh high values were noted for arginine at the middle sowing date - 0.92%, valine at middle and late sowing dates - 0.53%, leucine + isoleucine at a late sowing date - 1.23% and phenyl-

alanine at an early sowing date - 0.68%. The variety Ofenya had high content of arginine, valine, leucine + isoleucine and phenylalanine at a late date - 1.05, 0.83, 1.89, 0.95% respectively.

In the group of nonessential amino acids, the best values in the variety Gavrosh were observed at an early sowing date (asparagine - 1.19%, glutamine - 4.21% and cysteine - 1.11%). Higher content of alanine, glycine, proline, serine and tyrosine was at the late sowing date and amounted to 0.46 to 0.59%. In the variety Ofenya the highest indicator is characteristic of glutamine - 3.50%. The content of other nonessential amino acids was 0,63 to 0,99%. At the same time, the maximum values of all amino acids were formed at a late sowing date.

The studies have established the influence of the formation of quantitative and qualitative indicators of the yield from the conditions of moisture supply during the growing season of plants. The tendency of formation of higher crop capacity at lower conditions of moisture supply during sprouting and flowering of plants ($r = -0,2647...-0,3258$ at $R = 0,5760$) and higher - during tillering and ripening ($r = 0,1250...0,1571$ at $R = 0,5760$) has been noted. At the same time, differences in the influence of moisture conditions on the accumulation of amino acids in the varieties of naked oats were revealed. Such dependencies were most clearly manifested in the mid-season variety

Табл. 1. Урожайность сортов голозерного овса, 2018–2020 гг., т/га

Table 1. Yield of naked oats varieties, 2018–2020, t/ha

Variety	Year of study	Sowing date			Average by the experiment
		early	medium	late	
Gavrosh	2018	3,11	2,51	2,45	2,69
	2019	4,19	2,52	1,97	2,89
	2020	2,43	2,16	2,43	2,37
	Average for the period	3,24	2,40	2,28	2,64
Ofenya	2018	3,70	2,81	2,33	2,97
	2019	3,53	3,13	2,54	3,07
	2020	2,78	2,47	2,98	2,74
	Average for the period	3,34	2,80	2,62	2,92

Note. LSD₀₅ factor A (year) = 0.09; factor B (variety) = 0.04; factor C (sowing date) = 0.07.

Табл. 2. Содержание аминокислот в зерне сортов голозерного овса, 2018–2020 гг., %
Table 2. Amino acid content in grain varieties of naked oats, 2018–2020, %

Indicator	Gavrosh			Ofenya			LSD ₀₅
	Sowing date						
	early	medium	late	early	medium	late	
Arginine	0,88	0,92	0,85	0,81	0,97	1,05	0,07
Valine	0,50	0,53	0,53	0,59	0,71	0,83	0,10
Histidine	0,22	0,21	0,22	0,22	0,26	0,29	0,02
Isoleucine + leucine	1,16	1,19	1,23	1,36	1,67	1,89	0,24
Lysine	0,38	0,38	0,42	0,44	0,55	0,60	0,07
Methionine	0,13	0,14	0,14	0,17	0,21	0,22	0,03
Threonine	0,29	0,31	0,32	0,37	0,45	0,53	0,08
Tryptophane	0,15	0,13	0,13	0,17	0,12	0,13	0,02
Phenylalanine	0,68	0,64	0,67	0,69	0,82	0,95	0,10
Alanine	0,48	0,48	0,52	0,55	0,71	0,84	0,12
Asparagine	1,19	1,06	1,06	0,80	0,98	0,99	0,10
Glycine	0,50	0,51	0,55	0,59	0,72	0,90	0,12
Glutamine	4,21	3,43	3,70	2,82	3,20	3,50	0,38
Proline	0,52	0,54	0,59	0,66	0,73	0,90	0,10
Serine	0,48	0,47	0,54	0,65	0,76	0,89	0,13
Tyrosine	0,34	0,41	0,46	0,45	0,53	0,63	0,08
Cysteine	1,11	0,90	1,04	0,76	0,88	0,84	0,10
Total sum of amino acids	13,22	12,25	12,97	12,10	14,27	15,98	1,17
Sum of critical amino acids	0,66	0,65	0,69	0,78	0,88	0,95	0,10
Sum of essential amino acids	4,39	4,45	4,51	4,82	5,76	6,49	0,69
Sum of non-essential amino acids	8,83	7,80	8,46	7,28	8,51	9,49	0,62

Note. LSD₀₅ factor A (variety) = 0,05; factor B (sowing date) = 0,06.

Ofenya in all the variants of the experiment. The higher content of both essential and non-essential amino acids in this variety is formed at higher values of hydrothermal coefficient in May (sprouting - tillering period) ($r = 0,9467 \dots 0,9999$ at $R = 0,6660$), lower values in June (booting - flowering) ($r = -0,9338 \dots -0,9987$ at $R = 0,6660$), and then with increasing HTC in August, when the variety Ofenya had its filling-ripening period ($r = 0,4335 \dots 0,7888$ at $R = 0,6660$) (See Table 3).

In contrast to the medium ripening variety Ofenya in the early maturing variety Gavrosh differences in the correlation dependence of the formation of the content of amino acids on the growing conditions were observed. Thus,

the highest content of essential amino acids was noted at an early sowing date with low HTC values in May (sprouting - tillering period) ($r = -0,8812$ at $R = 0,6660$), higher - in June, i.e. during booting - flowering period ($r = 0,8626$ at $R = 0,6660$), and droughty conditions of the filling - ripening period (July) ($r = -0,6087$ at $R = 0,6660$). Medium and late sowing dates were more influenced by flowering-filling period conditions (June) ($r = 0,7731 \dots 0,9495$ at $R = 0,6660$), and ripening (August) ($r = -0,8138 \dots -0,9683$ at $R = 0,6660$).

When analyzing the content of amino acids at early sowing dates in the variety Gavrosh the inverse relationship was noted: the increased values of amino acids were formed

Табл. 3. Зависимость накопления аминокислот в зерне голозерного овса от условий влагообеспеченности в различные периоды вегетации, 2018–2020 гг.

Table 3. Dependence of the accumulation of amino acids in the grain of naked oats on the conditions of moisture supply in different periods of vegetation, 2018–2020.

Sowing date	HTC of the vegetation period	Gavrosh		Ofenya	
		Sum of amino acids			
		essential	non-essential	essential	non-essential
Early	May	-0,8812	0,7460	0,9914	0,9619
	June	0,8626	-0,7201	-0,9956	-0,9508
	July	-0,6087	0,7775	0,0305	0,4251
	August	-0,2811	0,0486	0,7888	0,4797
Medium	May	-0,5013	-0,6827	0,9718	0,9700
	June	0,5339	0,6544	-0,9621	-0,9600
	July	0,7731	-0,8312	0,3895	0,3965
	August	-0,9683	0,0419	0,5135	0,5069
Late	May	-0,1565	-0,8287	0,9467	0,9999
	June	0,1940	0,8068	-0,9338	-0,9987
	July	0,9495	-0,6860	0,4705	0,1731
	August	-0,8138	-0,1823	0,4345	0,6928

Note: $R = 0,666$ when $n = 9$, significant at the 5% significance level.

in the years with high moisture supply in May, dry conditions in June and the presence of precipitation in July ($r = 0,7460, -0,7201$ and $0,7775$ with $R = 0,6660$ respectively). The high content of non-essential amino acids at middle and late terms was influenced by the lowered HTC values during the sprouting - tillering period (May) ($r = -0,6827...$ -the presence of precipitation during flowering (June) ($r = 0,6544 ... 0,8068$ with $R = 0,6660$) and its absence during the ripening period (July - August) ($r = -0,1823 ... - 0,8312$ with $R = 0,6660$).

CONCLUSION

The study of the naked oats varieties Gavrosh and Ofenya under different conditions of cultivation revealed differences in the formation of quantity and quality of yield. The higher values of yield were recorded at early sowing terms. High content of essential amino acids in the variety Gavrosh was recorded at a late sowing date - 4.51%, and the content of

non-essential amino acids - at an early sowing date - 8.83%. The variety Ofenya had a higher content of both essential and non-essential amino acids at a late sowing date (6.49 and 9.49%, respectively).

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ПРИЗНАКИ КАЧЕСТВА ПЛОДОВ ЗЕМЛЯНИКИ САДОВОЙ И СЕЛЕКЦИЯ НА ИХ УЛУЧШЕНИЕ

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На основе анализа отечественных и зарубежных литературных источников обобщены сведения об основных признаках качества плодов земляники садовой (крупноплодность, твердость или прочность плодов, биохимический состав) и селекционных возможностях их улучшения на современном этапе селекции. Качественные характеристики плодов земляники садовой различаются по группам: товарные, потребительские, биохимические, физико-механические, технологические. При выращивании земляники садовой для потребления в свежем виде выделены следующие крупноплодные сорта: Clery (Италия), Florence (Великобритания), Alba (Италия), Roxana (Италия), Vima Xima (Нидерланды), Vima Tarda (Нидерланды), Vima Kimberly (Нидерланды), Maya (Италия), San Andreas (США), Таира, Нелли, Кемия, Элегия, Альфа, Берегиня, Царица, Крымчанка 87, Аросса, Заря, Крымская ранняя, Юниол, Янтарная. Признак твердости плодов относится к технологическим характеристикам, однако от него зависит и внешний вид плодов при сборе и транспортировке, что обуславливает товарный вид. Высокой степенью твердости плодов обладают сорта: Царица, Сюрприз олимпиаде, Рубиновый кулон, Фейерверк, Акварель, Алина, Нелли, Induka (Нидерланды), Clery, Darselect (Франция), Tenira (Нидерланды), Selektа (Канада), Polka (Нидерланды), Irma (Италия), Alba, Asia (Италия), Syria (Италия), Onda (Италия), Vivaldi (Нидерланды). Плоды земляники характеризуются уникальным составом биологически активных соединений, определяющих пищевую ценность культуры как источника диетического и лечебно-профилактического питания. Исследования свидетельствуют о преимущественной роли генотипа в накоплении антиоксидантов в плодах земляники садовой, а также о влиянии условий выращивания на реализацию генетического потенциала сортов. В связи с развитием технологий возделывания и расширением знаний о нутрицевтической ценности земляники садовой одной из приоритетных задач селекционных программ во всем мире стало улучшение качества плодов. Для повышения уровня признаков качества плодов у земляники садовой наибольший эффект может быть достигнут при использовании в селекции исходных форм с подтвержденными донорскими свойствами по данным признакам.

Ключевые слова: земляника садовая, признак, крупноплодность, твердость плодов, биохимический состав, селекция

QUALITY ATTRIBUTES OF GARDEN STRAWBERRY FRUITS AND BREEDING FOR THEIR IMPROVEMENT

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Based on the analysis of domestic and foreign literary sources the information about the main quality attributes of garden strawberry fruits (large fruit size, hardness or firmness of fruits, biochemical composition) and breeding possibilities of their improvement at the present stage of breeding is summarized. Quality characteristics of garden strawberries are differentiated by groups: commercial, consumer, biochemical, physical and mechanical, technological. When growing garden strawberries for fresh consumption, the following large-fruited varieties were identified: Clery (Italy), Florence (UK), Alba (Italy), Roxana (Italy), Vima Xima (Netherlands), Vima Tarda (Netherlands), Vima Kimberly (Netherlands), Maya (Italy), San Andreas (USA), Taira, Nelly, Kemiya, Elegy, Alpha, Bereginya, Tsaritsa, Krymchanka 87, Arossa, Zarya, Krymskaya rannaya, Uniol, Jantarnaja. The trait of fruit hardness refers to the technological characteristics, but it also depends on the appear-

ance of fruits during harvesting and transportation, which determines the commercial appearance. The following varieties have a high degree of fruit hardness: Tsaritsa, Surprise olympics, Rubinov kulon, Feyerverk, Aquarelle, Alina, Nelli, Induka (Netherlands), Clery, Darselect (France), Tenira (Netherlands), Selekt (Canada), Polka (Netherlands), Irma (Italy), Alba, Asia (Italy), Syria (Italy), Onda (Italy), Vivaldi (Netherlands). Strawberry fruits are characterized by a unique composition of biologically active compounds that determine the nutritional value of the crop as a source of dietary and therapeutic and preventive nutrition. Studies indicate a predominant role of genotype in the accumulation of antioxidants in garden strawberry fruits, as well as the influence of growing conditions on the realization of the genetic potential of the varieties. Due to the development of cultivation technologies and the expansion of knowledge about the nutraceutical value of garden strawberries, improving the quality of fruits has become one of the priority objectives of breeding programs around the world. To increase the level of fruit quality traits in garden strawberry, the greatest effect can be achieved by using the original forms with proven donor properties on these traits in breeding.

Keywords: garden strawberry, trait, large fruit size, fruit hardness, biochemical composition, breeding

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INTRODUCTION

Production of strawberries in the world is constantly growing. According to FAOSTAT, 6.4 million tons of strawberries were produced in 2011 and 8.9 million tons in 2020. The increase in production is due to both the increase in acreage cultivated (from 324,085 ha in 2011 to 384,668 ha in 2020) and the increase in yields (from 19.68 t/ha in 2011 to 23.04 t/ha in 2020)¹.

The interest in garden strawberry among producers is due to high profitability, adaptability of the crop to different growing condi-

tions, responsiveness to the intensification of production, the degree of development of cultivation technology and the pace of creation of new varieties [1]. Strawberry is a dietary food, its fruits have high nutraceutical value, and are widely used both fresh and in various processed products [2].

Breeding programs on strawberries were initially focused on the creation of varieties that are highly adaptable to different soil and climatic conditions of growth, increasing their yield and resistance to diseases^{2,3} [1, 3].

¹FAO. FAOSTAT – Food and Agriculture Organization Corporate Statistical Database. URL: <http://www.fao.org/faostat/en/#data/QC> (reference date April 11, 2022).

²Zubov A.A. Genetic features and breeding of strawberry: method. instructions. Michurinsk: All Russian Research Institute for Genetics and Breeding of Fruit Plants named after I.V. Michurin, 1990. 81 p.

³Zubov A.A., Popova I.V. Strawberry breeding. Program and methods of breeding of fruit, berry and nut crops. Ed. by Sedov E.N. Orel: VNIISPK, 1995. pp. 387-416.

The purpose of the research is to summarize the current information published in domestic and foreign literature on the main quality characteristics of garden strawberry fruits (large fruit size, fruit hardness, vitamin C and anthocyanins content), their inheritance and sources for breeding.

MATERIAL AND METHODS

The method of analysis of modern literary sources published by Russian and foreign scientists on the issues of strawberry breeding for improvement of fruit quality, the peculiarities of inheritance of these features and identification of new sources for breeding was applied during the research.

RESULTS AND DISCUSSION

With the development of growing technologies, logistics systems, and expanding knowledge about the nutritional and medicinal value of the crop, great importance in breeding programs is now given to the creation of varieties with high quality fruits [4]. Quality characteristics of garden strawberries are differentiated by groups: commercial, consumer, biochemical, physical and mechanical, technological⁴. Requirements for fresh strawberry fruits include characteristics of appearance, taste and odor, color, maturity, size by the largest transverse diameter, content of toxic elements and pesticides⁵. Fruit size in strawberries refers to both consumer and commercial and technological characteristics. The feature of large-fruitedness closely and positively correlates with plant productivity [5], determines the variety of commercial products, and ultimately affects the profitability of production. When growing garden strawberries for fresh consumption, preference is given to large-fruited varieties [5]. In most berry-growing zones of

Russia, varieties with average berry weight of 9-12 g and more are considered to be large-fruited [6, 7]. For the southern region this criterion is within the range of 20 g and more [5]. It follows from the publications of foreign researchers that varieties with an average fruit weight of more than 20 g also belong to large-fruited [1, 8].

Involvement of large-fruited varieties in the breeding process is the most frequently used method of increasing productivity in strawberries. It was found that crossing of large-fruited varieties with each other is effective for increasing the level of the trait [5].

The following modern foreign strawberry varieties of commercial importance are characterized by high large-fruitedness: Clery (24.5-30.98 g), Florence (14.7-17.9) [9], NF 311 (Alba) (19.7), NF205 (Roxana) (18.2), Vima Xima (11.0-14.5), Vima Tarda (14.3), Vima Kimberly (15.9) [7, 9], Maya (15.7-18.1), San Andreas (21.1 g) [9].

Among domestic varieties, the high degree of manifestation of signs of large berry size was noted in the varieties Taira (16.0 g), Nelli (13,5-15,9), Kemia (15,7), Elegia (15,0) [10], Alpha (16-17), Bereginya (9-12), Tsaritsa (16-20)⁶ [10]; Krymchanka 87 (13.5), Arossa (10.6), Zarya (11.1), Krymskaya rannaya (11-15), Uniol (10.5-12.5), Yantarnaya (15.6 d) [11]. Inheritance of such trait as fruit size in garden strawberry occurs according to the type of quantitative traits with predominance of small-fruitedness [5]. However, taking into account that the first commercial cultivars of strawberry had average fruit size from 6 to 10 g, and modern cultivars - from 16 to 20 g⁷, it can be concluded that selection for a larger fruit size has positive dynamics.

The hardness of strawberry fruits is of great importance for preserving the marketable ap-

⁴Shokaeva D.B., Zubov A.A. Wild strawberry, garden strawberry, zemklunika. Program and methods of varietal study of fruit, berry and nut crops. Ed. by Sedov E.N., Ogoltsova T.P. Orel: VNIISPK, 1999. pp. 417-443.

⁵GOST 6828-89 Fresh strawberry. Requirements for procurement, supply and sale. URL: <http://www.gostedu.ru/11092.html>. (reference date April 12, 2022).

⁶Andronova N.V. Garden strawberry varieties for industrial cultivation. Agrarian Science to Agriculture: Proceedings of the XIII International Scientific and Practical Conference: in 2 vols. 2018. Book 1. pp. 214-216.

⁷URL: <https://reestr.gossortrf.ru/> (reference date April 12, 2022).

pearance of products and their transportation over long distances [1, 5].

"Hardness" is a term that characterizes structural and mechanical properties of fruits. Fruit hardness correlates with the concept of fruit strength and is expressed in units - kg/ cm². However, the existing difference in the ways of defining the above properties (hardness - the property of a body to prevent penetration of another, a harder body into it; mechanical strength - the property of a body to resist destruction (splitting into parts) as well as irreversible change of shape (plastic deformation) under the action of external loads) indicates the need for a clear choice of the term used. In most scientific publications of domestic researchers, the concept denoting hardness or strength of fruits is replaced by the term "density" [5, 9]. [5, 9]. The sign of fruit hardness refers to technological characteristics, but it also determines the appearance of fruits during harvesting and transportation, which ultimately determines the commercial appearance. Many factors, including genetic characteristics of the variety, growing conditions, and the degree of maturity, affect the hardness and firmness of strawberry fruits [12]. It is important to base the determination of biometric indicators on the most accurate data. In the case of determining the strength (by crushing force to the appearance of a drop of juice) it is difficult to maintain a fixed area of impact on the strawberry fruit. In this regard, the use of a penetrometer with a given area fitting seems to be the most optimal.

According to the existing criteria for evaluation of domestic garden strawberry varieties for industrial production in the Nonchernozem belt, fruit strength (crushing force) must be at least 10.0 H; berry pulp strength in the Central Black Earth Region and the southern regions - 380 g and above⁸ [6, 7].

Of the strawberry assortment, the following garden fruit varieties have hardness (9.8 H and more) in the conditions of the south of the

Nonchernozem belt: Tsaritsa, Surprise Olympiade, Induka, Ruby Pendant and Fireworks [13], Clery, Darselect, Tenira, Selekt, Polka, Irma, Aquarelle [14]. In our studies in the conditions of the Nonchernozem belt, the highest hardness of fruits was noted in the variety Nelli (10.758 H), the high level of the trait was shown by the varieties Alba, Asia, Syria (8.895-9.316 H). Varieties with high fruit pulp strength in the conditions of the Krasnodar Territory include: Clery, Syria, Onda, Vivaldi, Nelli, Alba, Alina (over 400 g) [15].

According to the literature data, non-additive genetic interactions have a predominant influence on the inheritance of berry pulp strength trait. At the same time, there are also facts of additive summative effect in crossing combinations, when individual varieties show their donor potential [16]. According to Andronova N.V. and Tumaeva T.A., the study of inheritance of fruit strength in garden strawberry hybrids revealed a wide amplitude of trait variation as well as its significant variability within combinations of crosses and populations from free pollination. Intermediate succession of the fruit hardness trait with deviation towards the good parent has been established [14]. Thus, to increase the level of fruit hardness in garden strawberry, the greatest effect can be achieved by using in breeding the original forms with proven donor properties on this trait.

Strawberry fruits are characterized by a unique composition of biologically active compounds that determine the nutritional value of the crop as a source of dietary and therapeutic and preventive nutrition [1-3]. Interest in healthy lifestyles and constantly growing base of knowledge about the nutritional value of strawberry fruits increases the importance of selection of culture for a rich biochemical composition. The results of domestic and foreign studies indicate the high antioxidant potential of strawberry, which is due to the increased accumulation of vitamins, anthocya-

⁸The program of the North Caucasus Center for the selection of fruit, berry, flower and ornamental crops and grapes for the period up to 2030. Krasnodar: SKZNIISiV, 2013. 202 p.

nins, ellagic, ascorbic, folic acids and other bioactive compounds in its fruits [1, 2].

Biochemical composition of strawberry fruits in the whole crop is sufficiently studied, but the features of accumulation of valuable nutrients in specific varieties depend on both their genotype and growing conditions. Varieties having high degree of manifestation of this trait are of significant interest for breeding as regards vitamin C content, anthocyanins and other groups of compounds. According to Russian researchers, the domestic varieties with the highest ascorbic acid content in fruits are Alpha (85.6 mg%), Kokinskaya Zarya (83.0), Tsaritsa (77.3), Solovushka (74.3)⁹, Pamyati Zubova (92.7), Privlekatelnaya (88.7) [7]; Assol (85.4), Bagryana (84.0), Krymchanka 87 (83.4), Zarina (79.9), Present (79.9), Aidarina (77.6), Atlantida (76.1), Yantarnaya (74.2 mg%) [17], as well as the varieties of foreign selection Vima Tarda (73.1 mg%) [17], Flora (65.0-68.0), Red Gaultlet (60.0-62.0 mg%) [7]. The Russian varieties Solovushka (80.0 mg%), Alpha (75.0), Kokinskaya Zarya (75.0), Tsaritsa (70.0), Fireworks (112, 2 - 119,8), Privlekatelnaya (87,6-115,2), Memory of Zubova (96,9-110,7), Kemia (80,0), Nelly (79,7), Hera (78,5 mg%), the variety of foreign selection Flora (67,0-78,4 mg%) [7] are characterised by a high content of anthocyanins. Foreign scientists distinguish Albion [18] and Romina [19] by the anthocyanin content in strawberry fruits.

Recent studies indicate the predominant role of genotype in the accumulation of antioxidants in garden strawberry fruits, as well as significant influence of the growing conditions (temperature and light conditions, crop load, dates of fruit picking) on the realization of the genetic potential of the varieties [20]. Studies on the type of inheritance of biochemical composition of fruits in garden strawberry are insufficient, which is caused by both the large spectrum of studied substances and the impos-

sibility at the present stage of breeding in this limited direction. At the same time, the study of nutraceutical value of strawberry fruits continues. It is believed that the inheritance of genes responsible for the production of valuable compounds is of the quantitative type. Currently, for breeding to improve the biochemical composition of fruits, the method of inclusion in crosses of varieties with increased content of the complex of valuable substances, i.e. sources and donors for the selected trait is used. Positive results to improve the biochemical composition of strawberry fruits were obtained by foreign scientists using three generations of inverse crosses (BC1, BC2, BC3) from F1 *Fragaria* × *ananassa* (Fxa) × *F. virginiana glauca* (FVG) [4].

CONCLUSIONS

1. Improving the quality of strawberry fruits is one of the priority objectives of the breeding programs around the world due to the development of cultivation technologies and increasing knowledge about the nutraceutical value of the crop.
2. The results achieved indicate the positive dynamics of the breeding to increase the level of traits of quality indicators of garden strawberry fruits.
3. To increase the level of fruit quality traits, the greatest effect can be achieved by using in the breeding of original forms with a high level of manifestation of these traits.

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ФАЗОВАЯ ЧУВСТВИТЕЛЬНОСТЬ НЕКОТОРЫХ ВИДОВ ШИРОКОЛИСТНЫХ СОРНЫХ РАСТЕНИЙ К ГЕРБИЦИДУ ФЛЕКС

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Изучено действие гербицида Флекс, ВР (действующее вещество фомесафен 250 г/л) на распространенные в посевах сои однолетние виды сорных растений. Исследования проведены в условиях вегетационного домика в 2019–2021 гг. Определена чувствительность амброзии полыннолистной *Ambrosia artemisiifolia* L., акалифы южной *Acalypha australis* L., мари белой *Chenopodium album* L., коммелины обыкновенной *Commelina communis* L., канатника Теофраста *Abutilon theophrasti* Medik., гибискуса тройчатого *Hibiscus trionum* L., сигезбекии пушистой *Sigesbeckia pubescens* Makino, щирицы запрокинутой *Amaranthus retroflexus* L., эльсгольции ложногребенчатой *Elsholtzia pseudocristata* Levl. et Vaniot и дурнишника сибирского *Xanthium sibiricum* Patr. ex Widd. Обработку сорняков препаратом Флекс в нормах расхода 0,75; 1,0; 1,25 и 1,5 л/га проводили трижды за сезон в разные фазы роста и развития растений. Об уровне чувствительности сорных видов к гербициду судили по снижению высоты и массы надземных органов опытных растений в сравнении с контролем. Установлено, что гербицид Флекс полностью уничтожает растения всех испытанных видов, находящихся на ранних стадиях роста и развития (1–4 настоящих листа). При обработке сорняков, сформировавших 3–10 листьев, высокую чувствительность к действию препарата (снижение надземной массы до 94–100%) сохраняют амброзия полыннолистная, щирица запрокинутая, акалифа южная, гибискус тройчатый и дурнишник сибирский. Применение Флекса по переросшим растениям приводит к значительному снижению его активности в отношении всех изученных сорных видов. При использовании в третий срок гербицид эффективно действует только на щирицу запрокинутую, подавляя массу растений на 76–86%.

Ключевые слова: гербицид, сорняк, вид, фаза роста и развития, чувствительность

PHASE SENSITIVITY OF SOME BROAD-LEAVED WEED SPECIES TO THE HERBICIDE FLEX

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The effect of herbicide Flex, AS (active ingredient fomesafen 250 g/l) on annual weed species common in soybean crops was studied. The studies were conducted under greenhouse conditions in 2019–2021. The sensitivity of ragweed *Ambrosia artemisiifolia* L., Asian copper leaf *Acalypha australis* L., common lamb's quarters *Chenopodium album* L., common dayflower *Commelina communis* L., China jute *Abutilon theophrasti* Medik., trailing hollyhock *Hibiscus trionum* L., St.-Paul's-wort *Sigesbeckia pubescens* Makino, green amaranth *Amaranthus retroflexus* L., elsholtzia *Elsholt-*

zia pseudocristata Levl. et Vaniot, and Siberian cocklebur *Xanthium sibiricum* Patr. ex Widd was determined. Weed treatment with Flex at rates of 0.75, 1.0, 1.25 and 1.5 l/ha was carried out three times a season at different stages of plant growth and development. The level of sensitivity of weed species to herbicide was judged by the decrease in height and weight of the aboveground organs of the experimental plants compared to the control. It was found that Flex herbicide completely destroys plants of all tested species in the early stages of growth and development (1-4 true leaves). When treating weeds that have formed 3-10 leaves, ragweed, green amaranth, Asian copper leaf, trailing hollyhock and Siberian cocklebur remain highly sensitive to the drug action (reduction of the aboveground weight up to 94-100%). Application of Flex on overgrown plants leads to a significant decrease in its activity against all studied weed species. When used in the third term, the herbicide is effective only on the green amaranth, suppressing the mass of the plants by 76-86%.

Keywords: herbicide, weed, species, growth and development phase, sensitivity

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Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

Soybean is one of the key crops of the world agriculture. The use of soy in modern Russian agriculture as an agricultural crop with high protein content makes it possible to solve the problem of its deficit for humans and animals and to provide industry and medicine with strategic raw materials¹. Soybean seeds contain 16-20% vegetable oil and 37-42% protein containing essential amino acids [1].

The Far Eastern Federal District (FEFD) is a traditional soybean growing area in Russia, which is due to the optimal agroclimatic conditions for the crop in the region [2]. According to Rosstat (Russian Federal State Statistics Service), the sown area of soybean in 2021 in Russia was 3.068 mln ha, in the FEFD - almost 1.2 mln ha; the average yield across Russia was 15.9 c/ha of the harvested area, in the FEFD - 14.8 c/ha, in the Primorsky Territory - 14.0 c/ha². Given the need to restore the rational and efficient use of arable land, it is planned

to increase gross soybean production in the Far East to 3 million tons by 2024 [3].

Effective measures to control diseases, plant pests and weeds are necessary to obtain high and stable yields of grown crops. Soybean yield losses caused by weeds vary from 25 to 80% depending on the varietal characteristics of the crop, species diversity and number of weeds, density and duration of crop infestation, as well as environmental conditions [4-6]. In the early stages of ontogenesis, soybean is characterized by weak competitive ability due to its relatively slow growth. Therefore, even low weediness leads to significant yield losses, and medium and high weediness reduces crop productivity by 3-5 times [7, 8].

Species composition of weeds, their number and distribution in agrocenoses are in constant dynamics determined by climatic changes, and directly depend on weather conditions of the growing season, as well as on the features of agricultural crops cultivation technologies. The

¹Innovative views on the modern technology of soybean cultivation in the Kursk region. Practical guide. Ministry of Science and Higher Education of the Russian Federation, FSBSI "Kursk Federal Agrarian Scientific Center". Kursk, 2019. 43 p.

²Bulletin "Gross yields and crop yields in the Russian Federation in 2021". (P. 2). URL: <https://rosstat.gov.ru/compendium/document/13277> (reference date 2022-03-28).

monitoring of soybean agrocenoses in the Primorsky Territory conducted by the staff of the Far Eastern Research Institute of Plant Protection (DVNIIZR) from 2016 to 2020 revealed 108 species of weeds belonging to 31 botanical families. Of these, dicotyledonous weeds greatly outnumbered monocotyledonous weeds - 89 (54 short-lived and 35 perennial) species versus 19 (10 short-lived and 9 perennial)³ [9]. Among the dicotyledonous short-lived plants, the maximum 10-year average frequency of occurrence in soybean crops (84-99%) was characteristic of Asian copper leaf *Acalypha australis* L., ragweed *Ambrosia artemisiifolia* L. and common lamb's quarters *Chenopodium album* L. China jute *Abutilon theophrasti* Medik., trailing hollyhock *Hibiscus trionum* L., St.-Paul'swort *Sigesbeckia pubescens* Makino were detected in 35-54% of the surveyed crops. This index was 12-25% for the green amaranth *Amaranthus retroflexus* L., elsholtzia *Elsholtzia pseudocristata* Levl. et Vaniot, and Siberian cocklebur *Xanthium sibiricum* Patr. ex Widd. The frequency of occurrence of the short-lived weed common dayflower *Commelina communis* L. was on average 56%.

Mean stand density in soybean crops in 2010-2020 was 71.8 plants/m² for Asian copper leaf, 20.1 for common lamb's quarters, 17.8 for common ragweed, 3.3 for green amaranth, 2.4 for common dayflower, 1.3 for trailing hollyhock and St.-Paul'swort, and 1.2 plants/m² for China jute. The average number of Siberian cocklebur and elsholtzia per 1 m² of soybean area during the last 10 years was less than 1 plant.

The biological threshold of harmfulness⁴ of annual dicotyledonous weeds in soybean crops is 2-6 pcs/m². According to our data, the average total weed infestation of soybean in the control variants on the experimental fields of

DVNIIZR in 2016-2020 ranged from 178 to 640 pcs/m², the crude aboveground weight of weeds was 2125-4797 g/m², soybean yield losses depending on the level of weeding reached 0.36-1.83 t/ha (29-96% of the yield in variants with manual weeding) [10]. In 2021 in the experimental soybean crops the average standing density of all weeds in the variants without the use of herbicides was 1132 pcs/m², their crude aboveground mass was 2656 g/m². The proportion of cereal annual species reached 73% of the total number of weeds, dicotyledonous annual and common dayflower - 22%, and perennial dicotyledonous species - 5%, which resulted in a decrease in soybean yield by 44% (0.68 t seeds/ha).

The most effective method of reducing the negative effects of pests on crops remains the use of chemical means of protection, provided that the basic agrotechnical measures are carried out. When implementing the chemical method, it is necessary to minimize environmental risks, exclude the toxic effects of pesticides on protected crops and non-target objects [11, 12]. The list of chemical and biological preparations is constantly updated, new plant protection agents enter the market, requiring a comprehensive study in the soil and climatic conditions of the region, for subsequent regulation of their effective and safe use in the agricultural production.

Currently, 227 herbicides based on 34 active substances (active substances)⁵ have been approved for use in soybeans in the Russian Federation. In February 2021, the selective post-emergent herbicide Flex, WS (v. fomesafen 250 g/l), designed to control a wide range of annual dicotyledonous weeds in soybean crops, was registered in the Russian Federation; its comprehensive study has begun in the Far East, Krasnodar Territory, Leningrad, Tambov and Astrakhan Regions⁶ [13-16]. The effectiveness

³Vostrikova S.S., Morokhovets V.N., Morokhovets T.V., Basai Z.V., Shterbolova T.V. Dynamics of weed component in soybean agrocenoses of the Primorsky Territory. Scientific support of soybean production: problems and prospects: Collection of scientific articles on materials of the International scientific conference devoted to the 50th anniversary of the All-Russian Soybean Research Institute. Blagoveshchensk April 18, 2018 / FSBSI VNII soybean. Blagoveshchensk: LLC "IPK "ODEON", 2018. pp. 131-140.

⁴Biological (economic) thresholds of harmfulness of pests, diseases and weeds in crops. Reference book. Priluki, 2018. 27 p.

⁵List of pesticides and agrochemicals permitted for use in the Russian Federation. 2022 M., 2022. 879 p.

⁶Morokhovets V.N., Morokhovets T.V., Shterbolova T.V., Basai Z.V., Baimukhanova A.A., Skorik N.S. Evaluation of the effectiveness of a tank mixture of new herbicide Flex with graminicide Fusilad Forte in soybean crops. Agrarian Science: Special issue for the International Scientific and Practical Conference "Plant Immunity to infectious diseases", dedicated to the 100th anniversary of the monograph by N.I. Vavilov. 2019. Vol. 2. pp. 150-155. DOI:10.32634/0869-8155-2019-326-2-150-155.

of selective herbicides largely depends on the species composition of the weed component of the protected agrocenosis and the stage of development of controlled weeds during treatment [17].

The purpose of the study is to investigate the species and phase sensitivity to Flex herbicide of annual broadleaf weeds common in soybean crops in the south of the Far East.

The objective of the study is to determine the level of herbicidal activity of the preparation Flex against annual weed species at different stages of growth and development.

MATERIAL AND METHODS

Experimental work was performed in 2019-2021 at the experimental base of DVNIIZR under the conditions of a greenhouse, according to the generally accepted methodology⁷. Latin and Russian names of the weed species are given according to the publication "Vascular Plants of the Soviet Far East. [18-20].

The weeds were grown in 500 cm³ growing vessels in a mixture of meadow-brown podzolized soil (according to its texture, average clay containing 3.8% organic matter (GOST 2621-91), 16 and 120 mg/kg of mobile phosphorus and exchangeable potassium (GOST 54650-2011), respectively, and pH_{salt} (GOST 26483-85) - 5.3) and rotted compost in the ratio of 1 : 1, sifted through a sieve with a hole size of 5 mm. The resulting substrate was placed in vessels, compacted, moistened, and evenly covered by the weed seeds on its surface in an amount sufficient to produce 10-15 plants, covered with soil and compost mixture with a layer of about 1 cm, tamped down and watered.

Before applying the drug to vegetating weeds in the vessels, excess plants were removed, leaving specimens close in height and phase of the development. Weed plants of the test species were treated in three periods with herbicide Flex, WS at the rates of 0.75; 1.0; 1.25 and 1.5 l/ha with the addition of surface acting agent (SAA) Trend 90, W (e.g. isodecyl

alcohol ethoxylate, 900 g/l) 0.2 l/ha using a stationary sprayer OP-5 designed by the All-Russian Research Institute of Phytopathology. The repetition of the experiment was 10-times. Data on the growth and development of weeds before the treatments are given in Table 1.

During the experiments by daily watering the soil humidity in vessels was maintained at an optimum level for plants (60-70% of full moisture capacity). Plant growth and development in control and experimental (with herbicide application) variants were regularly monitored, the following symptoms of herbicide toxic effect on the weed species under study were recorded: delayed growth and development of plants; changes in leaf coloration, deformation of leaf plates, appearance of burns, necroses; other changes and deviations from the control variant without treatment.

In 14-21 days after the treatment the weed plants were cut, their height was measured and the raw above-ground biomass was weighed. The level of herbicide toxicity for the studied weed species was judged by the decrease in height and weight of the above-ground organs of experimental plants compared to the control. For the evaluation of herbicidal activity our own adaptation of the efficiency levels gradation was used: reduction of heights and weights of experimental plants by 91-100% - very good effect, 76-90% - good, 51-75% - satisfactory and 0-50% - no effect/weak effect.

RESULTS AND DISCUSSION

Visually noticeable symptoms of the toxic effect of Flex on the studied weed species were observed one day after the treatment. When applying the herbicide in the first term, the loss of turgor, deformation and dieback of the upper point of growth, withering of leaves was noted in ragweed, Asian copper leaf, China jute and Siberian cocklebur. In trailing hollyhock, green amaranth and elsholtzia burns in the form of light and brown spots on the leaf plates, curling and wilting of leaves, and dieback of the

⁷Spiridonov Y.Y., Larina G.E., Shestakov V.G. Methodological guide for the study of herbicides used in crop production. Moscow: Pechatny gorod, 2009. 252 p.

Табл. 1. Фазы развития и высота сорных растений до обработки гербицидом Флекс, 2019–2021 гг.
Table 1. Phases of development and height of weeds before treatment with Flex herbicide, 2019–2021

Type of weed	First treatment period		Second treatment period		Third treatment period	
	Development phase	Height, cm min–max (variant average)	Development phase	Height, cm min–max (variant average)	Development phase	Height, cm min–max (variant average)
Ragweed	1-2 pairs of leaves	2,0–5,0 (3,3)	3–4 pairs of leaves	5,5–13,0 (8,7)	9–11 leaves, start of budding	11,0–25,5 (17,8)
Asian copper leaf	1–2 leaves	1,5–4,5 (3,1)	4–5 leaves, branching start	6,5–15,5 (10,2)	6–7 leaves, branching	12,0–23,5 (18,8)
Common lamb's quarters	1-2 pairs of leaves	2,0–7,0 (4,6)	4–5 pairs of leaves, branching start	8,0–21,0 (12,0)	10–14 leaves, branching, flowering	14,0–28,0 (20,0)
Common dayflower	1–2 leaves	1,5–4,5 (2,7)	3–5 leaves	6,0–12,5 (10,4)	6–7 leaves, branching start	12,0–25,5 (20,5)
China jute	1–2 leaves	3,5–7,5 (5,6)	3–4 leaves	9,0–17,5 (13,1)	6–7 leaves, start of budding	17,0–30,5 (23,6)
Trailing hollyhock	1–2 leaves	3,0–6,5 (4,8)	3–5 leaves	6,0–12,0 (9,2)	6–7 leaves, branching, flowering	13,5–29,5 (22,6)
St.-Paul's wort	1–2 pairs of leaves	1,0–3,5 (2,1)	2–3 pairs of leaves	3,5–10,0 (6,0)	3–4 pairs of leaves, branching start	12,5–18,0 (15,6)
Green amaranth	1–2 leaves	1,5–5,5 (3,8)	4–6 leaves	3,0–13,0 (8,1)	7–11 leaves, start of budding	11,0–21,0 (15,4)
Elsholtzia	1–2 pairs of leaves	2–3,5 (2,6)	3–4 pairs of leaves, branching start	7,0–11,5 (8,9)	4–5 pairs of leaves, branching	11,0–17,0 (14,0)
Siberian cocklebur	1 pair of leaves	3,5–8,0 (5,6)	2–3 pairs of leaves	6,0–17,5 (13,0)	3–4 pairs of leaves, start of budding	24,0–35,0 (27,8)

single upper growth points were noted. For common lamb's quarters, St.-Paul's wort and elsholtzia the suppressive herbicidal effect was expressed in extensive leaf chlorosis and leaf wilting, stem necrosis and dieback of the growth points. Local light spots of different sizes (burns) on leaves, damage (withering) of the upper growth point were recorded on common dayflower plants. It should be noted that the intensity of the weeds damage by herbicide was very high, up to 90-100%, and in the variants from the minimum to the maximum the rates of consumption differed slightly. One hundred percent death of all experimental plants of the studied species was recorded as early as 5 days after the treatment, which fully agrees with our earlier experimental data [10]. It was found that the most common and/or harmful dicotyledonous annual weeds typical of the Primorsky Territory agroecosystems in the early stages of growth

and development exhibit extremely high sensitivity to herbicide Flex.

After treatment in the second term, the herbicides were effective against ragweed, green amaranth, Asian copper leaf, trailing hollyhock, Siberian cocklebur, elsholtzia and China jute. The plants wilted, curling and desiccation of leaves, deformation and dieback of growth points, formation of numerous burns in the form of light and brown spots on the leaf plates were noted. For common lamb's quarters and St.-Paul's wort the toxic effect of the preparation was expressed in chlorosis and wilting of upper leaves, deformation of the growth point; for common dayflower it was expressed in formation of scattered spots on the leaf plates, drying and dying of the leaf buds in the upper growth points, in a lag in growth of experimental plants from the control specimens (without treatment).

After the application of herbicide Flex in the third term, the same signs of suppression were observed in the studied weed species as in the second term, but they were expressed much weaker. The plants of green amaranth and trailing hollyhock showed only curling and wilting of the upper and some lateral leaves, and slight damage (deformation) of the points of growth. Light and brown spots on the leaf plates, curling and withering of leaves at the upper point of growth were observed in ragweed and Asian copper leaf; curling and withering of upper leaves were observed in China jute, St.-Paul's wort, common lamb's quarters, elsholtzia, Siberian cocklebur and common dayflower - upper leaf curling and withering.

High herbicidal activity of Flex at the second treatment term in all tested rates was demonstrated against ragweed, green amaranth, Asian copper leaf and trailing hollyhock reducing plant biomass by 93-100% and plant height by 84-100% (see Tables 2, 3). Siberian cocklebur, elsholtzia and China jute were highly susceptible to the herbicide, reducing their above-ground mass by 77-94%. The preparation was less effective on common lamb's quarters, common dayflower and St.-Paul's wort (55-75% weight reduction).

Only green amaranth showed good sensitiv-

ity (suppression by weight by 76-86%) to Flex herbicide when treated in the third term. The preparation showed satisfactory effectiveness against ragweed, Asian copper leaf, trailing hollyhock and common dayflower suppressing their recruitment of the above-ground mass by 51-70%. Siberian cocklebur, China jute, common lamb's quarters and St.-Paul's wort lost 28-50% of the biomass and 18-44% of the height compared with the control, demonstrating relative resistance to herbicide treatment in the third term. The drug had a satisfactory effect on elsholtzia only in the application rates of 1.25 and 1.5 l/ha.

CONCLUSION

As a result of the studies conducted in the greenhouse conditions, it was found that the herbicide Flex at post-emergence application at a rate of 0.75-1.5 l / ha completely destroys plants of ragweed, Asian copper leaf, green amaranth, trailing hollyhock, Siberian cocklebur, elsholtzia, China jute, common lamb's quarters, St.-Paul's wort and common dayflower, which are in the early stages of growth and development.

When weeds are treated in the second term, in the later stages of growth (10-21 cm) and development (3-10 true leaves), ragweed, green

Табл. 2. Снижение надземной массы сорных растений при обработке гербицидом Флекс, % к контролю (среднее за 2019–2021 гг.)

Table 2. Reduction of the aboveground mass of weeds when treated with Flex herbicide, % to the control (average for 2019–2021)

Type of weed	Treatment time											
	first				second				third			
	Consumption rate of the preparation, l/ha											
	0,75	1,0	1,25	1,5	0,75	1,0	1,25	1,5	0,75	1,0	1,25	1,5
Green amaranth	100	100	100	100	99	100	100	100	76	78	79	86
Ragweed	100	100	100	100	99	100	100	100	57	61	64	66
Asian copper leaf	100	100	100	100	94	95	97	99	54	60	63	66
Trailing hollyhock	100	100	100	100	93	95	95	96	56	63	66	70
Siberian cocklebur	100	100	100	100	82	88	91	94	32	36	40	44
Elsholtzia	100	100	100	100	80	84	88	90	46	50	62	67
China jute	100	100	100	100	77	81	83	84	32	35	37	46
Common lamb's quarters	100	100	100	100	61	62	67	68	28	32	49	50
Common dayflower	100	100	100	100	55	66	69	75	51	54	58	58
St.-Paul's wort	100	100	100	100	55	55	62	66	36	37	43	46

Note. Here and in Table 3: 91-100% - very good action; 76-90% - good; 51-75% - satisfactory; 0-50% - no action/weak action.

Табл. 3. Снижение высоты сорных растений при обработке гербицидом Флекс, % к контролю (среднее за 2019–2021 гг.)**Table 3.** Reduction of weed height when treated with Flex herbicide, % to the control (average for 2019–2021)

Type of weed	Treatment time											
	first				second				third			
	Consumption rate of the preparation, l/ha											
	0,75	1,0	1,25	1,5	0,75	1,0	1,25	1,5	0,75	1,0	1,25	1,5
Ragweed	100	100	100	100	99	100	100	100	44	48	48	50
Green amaranth	100	100	100	100	98	98	99	100	54	60	62	69
Asian copper leaf	100	100	100	100	91	92	94	96	42	42	43	44
Trailing hollyhock	100	100	100	100	84	88	90	94	38	42	44	50
China jute	100	100	100	100	68	72	74	82	19	22	27	38
Elsholtzia	100	100	100	100	67	74	80	85	32	42	47	54
Siberian cocklebur	100	100	100	100	62	72	79	80	18	23	28	32
Common dayflower	100	100	100	100	61	71	74	80	56	58	62	62
Common lamb's quarters	100	100	100	100	55	58	60	60	24	30	42	44
St.-Paul's wort	100	100	100	100	45	48	56	60	28	29	32	36

amaranth, Asian copper leaf, trailing hollyhock and Siberian cocklebur remain highly sensitive to the action of the drug Flex. When used in the second term, the drug is also quite effective against elsholtzia and China jute.

When Flex is used in the third term, for overgrown weeds (plant height - up to 17-35 cm, the phase of development - from branching to flowering (formation of 6-14 true leaves)), herbicidal activity of the preparation as compared with the effectiveness of the treatment in the first term is reduced by 1.2-3.6 times.

Of the studied weed species, high sensitivity to the studied herbicide in all tested treatment rates and timing was revealed only in green amaranth. Regarding ragweed, Asian copper leaf, trailing hollyhock, Siberian cocklebur, elsholtzia and China jute good efficiency of Flex can be achieved only when using it before the formation of 4-8 true leaves by the weeds. For the destruction of common lamb's quarters, common dayflower and St.-Paul's wort the herbicide should be used only in the earliest stages of their growth (3.5-7.0 cm) and development (1-4 true leaves).

The experimental data are a convincing argument in favor of using Flex herbicide in the earliest phases of growth and development of weeds.

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ОЗОННАЯ ДЕЗИНСЕКЦИЯ ЗЕРНА ОТ АМБАРНОГО ДОЛГОНОСИКА И БУЛАВОУСОГО ХРУЩАКА

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Изучены способы борьбы с наиболее распространенными в центральных регионах Российской Федерации вредными насекомыми, которые наносят ущерб сельскохозяйственной продукции в период хранения. Эксперимент проведен в лабораторных условиях путем дезинсекции озонированием зернового вороха озимой пшеницы. Объект исследований – амбарные долгоносики (*Sitophilus granaries* L.) и булавоусые хрущак (*Tribolium confusum* L.). Отмечено, что рекомендуемые в настоящее время для дезинсекции концентрации озона могут быть опасны для здоровья обслуживающего персонала. Исследована возможность проведения озонной обработки при меньших значениях (до 5 мг/м³). Установлено, что для полного уничтожения амбарного долгоносика достаточна концентрация озона в диапазоне 3–5 мг/м³ при экспозиции 300 мин. Поскольку поддерживать постоянное содержание газа сложно, следует ориентироваться на дозу озонной обработки свыше 1315 мг·мин/м³. Для полного уничтожения булавоусого хрущака следует озонировать при заданных параметрах не менее 460 мин. Доза озонной обработки должна превышать 1935 мг·мин/м³. Озонирование следует проводить до момента гибели примерно половины насекомых. Оставшаяся часть вредителей погибает в течение следующих суток после обработки, так как озон, воздействуя на гемолимфу, практически исключает вероятность выживания жуков после их парализации. Применение исследованных показателей концентрации газа способствует повышению качества семян зерновых культур. Озонная дезинсекция с указанными параметрами может быть объединена с операцией предпосевной подготовки посевного материала. За счет высокой активности озона такие концентрации достаточно быстро распадаются на молекулярный кислород, что значительно снижает риск отравления человека.

Ключевые слова: озонирование, семена, зерно, озонная дезинсекция зерна, вредители зерна

OZONE DISINFECTION OF GRAIN FROM GRANARY WEEVIL AND CONFUSED FLOUR BEETLE

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The methods of combating the most common harmful insects in the central regions of the Russian Federation, which cause damage to agricultural products during storage, were studied. The experiment was carried out in laboratory conditions by ozonation disinsection of a grain heap of winter wheat. The object of research is granary weevils (*Sitophilus granaries* L.) and confused flour beetles (*Tribolium confusum* L.). It is noted that the currently recommended ozone concentrations for disinfection can be dangerous for the health of service personnel. The possibility of ozone treatment at lower values (up to 5 mg/m³) was investigated. It was found that ozone concentration in the range of 3-5 mg/m³ at an exposure of 300 min is sufficient for the complete destruction of the granary weevils. Since it is difficult to maintain a constant gas content, it is necessary to focus on the dose of ozone treatment over 1315 mg·min/m³. For the complete destruction of the confused flour beetles, at least 460 min should be ozonated at the specified parameters. The dose of ozone treatment should exceed 1935 mg·min/m³. Ozonation should be carried out until about half of the insects die. The remaining part of the pests dies within the next day after the treatment,

since ozone, acting on the hemolymph, practically eliminates the possibility of survival of beetles after their paralysis. The use of the studied gas concentration indicators contributes to improving the quality of grain seeds. Ozone disinfection with the specified parameters can be combined with the operation of pre-sowing preparation of the seed material. Due to the high activity of ozone, such concentrations quickly decompose into molecular oxygen, which significantly reduces the risk of human poisoning.

Keywords: ozonation, seeds, grain, ozone disinfection of grain, grain pests

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Конфликт интересов

Авторы заявляют об отсутствии конфликта интересов.

Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

During storage, insect pests may multiply in the grain, which destroy a significant part of the crop. In this regard, there is not only a decrease in the physical weight of the grain, but also an increase in the toxicity of the grain pile due to the increased content of products of pests in it. Contaminated grain cannot be used for fodder to avoid reduced animal productivity or mortality. Because of pests about 15% of the world's crops are spoiled every year [1-4]. In Russia, a significant part of grain is infested by weevils and flour beetles. According to the State Standard 13586.6-93 these pests have the coefficient of harmfulness from 0.4 to 1.5 depending on their type. Infestation of grain by granary weevil and confused flour beetle in Russia amounts to about 40% in total. However, even low levels of pests are dangerous because they develop quickly and the quality of grain and its products is greatly reduced. Currently, to combat grain pests mostly various expensive chemical preparations are used which are dangerous for both humans and the environment. Therefore, there is an active search for new, environmentally safe methods of disinsection [5- 10]. The most promising in this direction is considered the process of ozonation [11-15]. Ozone treatment does not require pre-production of gas because the

ozone-air mixture can be obtained from the air by using special equipment - ozonators. In this case after the disinsection ozone turns into oxygen, without polluting the atmosphere, but even enriching it. In this regard, research aimed at studying the process of ozonation to prevent the development of grain pests or their destruction is relevant.

The purpose of the research is to determine the regimes of ozone treatment of grain pests that provide effective destruction of granary weevil and confused flour beetle during disinsection and under laboratory conditions.

MATERIAL AND METHODS

Earlier studies allowed us to determine the effectiveness of ozone treatment against grain pests. However, ozone concentrations in ozone-air mixture used in these experiments are significant (from 70 to 2000 mg/m³). Such gas content is not only destructive for insects, but also very dangerous for humans. The use of ozone-air mixtures at these concentrations can be dangerous for the operating personnel. High gas content in the ozone-air mixture negatively affects the sowing qualities of the grain. Therefore, it is necessary to determine the effectiveness of ozone disinsection of grain pests under laboratory conditions at ozone concentrations up to 5 mg/m³. This gas

content contributes to improvement of quality indicators of seeds of cereal crops, such as germination energy, germination rate, growth force, etc. Consequently, if ozone desinsection with the above parameters is sufficiently effective, it can be combined with the operation of seed pre-sowing preparation. In addition, due to the high activity of ozone, such concentrations decompose quite quickly into molecular oxygen, which significantly reduces the risk of human poisoning.

To determine the efficiency of different ozone treatment modes, the insects were divided into five groups, each of which was placed in an individual air-permeable container. Four samples were subsequently ozonized in quadruplicate, and the fifth variant was left in natural conditions and served as a control. Beetles unable to move were considered dead, because no return of paralyzed individuals to life was recorded. The effectiveness of ozone treatment, expressed as a percentage, was determined by dividing the number of dead pests by their total number before the experiment.

Ozonation was carried out in a sealed glass container, which has supply and return lines of ozone-air mixture. Ozone concentration was determined at the output by means of gas analyzer "Sigma-03" equipped with two electrochemical sensors "Sigma-03. DE", one of which determined the gas level in the working area, the other - in the exhaust gas. Four containers with pests were placed inside the tank. The first sample was ozonized for 5 hours. Subsequently, the samples were removed from the container after 50-60 min. The ozone concentration in the exhausted ozone-air mixture varied from 3.3 to 5.0 mg/m³, and was determined every 10 min with the error of 0.02 mg/m³ (see Table 1).

The data in Table 1 show that it is difficult to maintain a given value of ozone concentration in the ozone-air mixture for a fairly long period of time. In addition, not all ozonators have the ability to adjust this parameter. That is why the "ozone treatment dose" is of great importance that can be obtained by multiplying the ozone concentration by the exposure

Табл. 1. Параметры озонной обработки вредителей зерна

Table 1. Parameters of ozone treatment of grain pests

Ozonation time, min	Ozone concentration, mg/m ³	Ozone dose, mg·min/m ³
0–10	4,82	48,2
11–20	5,0	50,0
21–30	5,0	50,0
31–40	3,34	33,4
41–50	4,0	40,0
51–60	4,23	42,3
61–70	5,0	50
71–80	3,96	39,6
81–90	4,02	40,2
91–100	5,0	50,0
101–110	3,48	34,8
111–120	5,0	50
121–130	4,62	46,2
131–140	4,0	40,0
141–150	4,3	43,0
151–160	5,0	50,0
161–170	4,2	42,0
171–180	3,98	39,8
181–190	4,52	45,2
191–200	4,34	43,4
201–210	4,65	46,5
211–220	4,58	45,8
221–230	4,72	47,2
231–240	4,43	44,3
241–250	4,21	42,1
251–260	4,14	41,4
261–270	4,28	42,8
271–280	4,42	44,2
281–290	3,96	39,6
291–300	4,37	43,7
1st batch extraction Average for 300 min	4,386	Totally 1316
301–310	3,88	38,8
311–320	3,94	39,4
321–330	4,21	42,1
331–340	4,1	41,0
341–350	4,17	41,7
2-nd batch extraction Average for 350 min	4,339	Totally 1519
351–360	3,52	35,2
361–370	3,78	37,8
371–380	2,98	29,8
381–390	5,0	50,0
391–400	5,0	50,0
3-rd batch extraction Average for 400 min	4,304	Totally 1722
401–410	2,76	27,6
411–420	3,51	35,1
421–430	3,84	38,4
431–440	3,68	36,8
441–450	3,75	37,5
451–460	3,89	38,9
4-th batch extraction Average for 460 min	4,208	Totally 1936

time. This parameter is easier to control during disinfestation. The same dose can be obtained at high ozone concentration, but a small time of the experiment, or at low ozone concentration, but a large exposure. All ozonators are individual and they cannot maintain the specified mode of gas content in the working mixture. Termination of the process when reaching a certain dose of ozone treatment is possible by simple calculations. MAC level of ozone in the working zone was continuously monitored, its exceedance in the course of the experiment was not observed. Consumption of the ozone-air mixture throughout the experiment was constant and amounted to 1 m³/h. Ambient air temperature was in the range of 22-24 °C, relative humidity of the working mixture varied from 55 to 60%.

RESULTS AND DISCUSSION

The experiment was carried out in the laboratory of the Voronezh State Agrarian University n.a. Emperor Peter the Great. The results of ozone disinfestation treatment of granary weevils and confused flour beetles developing in grain of winter wheat are presented in Tables 2 and 3.

Analysis of Tables 2, 3 shows that ozone treatment against the considered grain pests is quite effective. Moreover, granary weevil beetles were more susceptible to the effects of ozone-air mixture. Within 300 minutes of ozone treatment at the average ozone concentration of 4.386 mg/m³ and the dose of 1316 mg·min/m³ 60% of the insects were killed. In 5 h after the treatment, this rate increased by 10%; after 12 h, the mortality rate was 100%.

Табл. 2. Эффективность озонной обработки при озонировании амбарного долгоносика

Table 2. Efficiency of ozone treatment during ozonation of granary weevil

Indicator	Ozone dose, mg·min/m ³			
	1316	1519	1722	1936
Overall effectiveness of ozone disinsection, %:	100	100	100	100
Including dead beetles, number:				
during treatment	60	60	70	80
after treatment:				
after 5 h	70	80	80	90
after 12 h	100	100	100	100

Табл. 3. Эффективность озонной обработки при озонировании булавоусого хрущака

Table 3. Efficiency of ozone treatment during ozonation of confused flour beetle

Indicator	Ozone dose, mg·min/m ³			
	1316	1519	1722	1936
Overall effectiveness of ozone disinsection, %:	30	40	50	100
Including dead beetles, number:				
during treatment	20	30	30	30
after treatment:				
after 5 h	20	30	40	60
after 12 h	30	30	40	90
after 24 h	30	40	50	100

At the same time, 30% of the confused flour beetles died after 24 hours of observation.

Over the next 50 minutes of ozone treatment the dose of ozone treatment reached 1519 mg-min/m³. At the same time, the effect of ozone-air mixture on the granary weevil remained practically unchanged. Immediately after treatment, 60% of the beetles died. In 5 hours of observation, this indicator increased by 20%. The rest of the pests died in 12 hours after the treatment. At the same parameters, 30% of the confused flour beetles died during ozone treatment, and 10% died after one day of observations. Therefore, the effectiveness of ozone disinsection under these conditions against this pest was only 40%.

Further 50 min of ozonation allowed to reach the ozone treatment dose of 1722 mg/m³. Under these conditions the effectiveness of disinfestation against granary weevil was similar to the previous parameters, i.e., 70% of the beetles were killed immediately, and the remaining ones were killed after 12 h of observation. The same dynamics was observed in the experiment with the confused flour beetle. Thirty percent of individuals were paralyzed or dead right after ozonation. Another 10% of the beetles died during the next 5 hours of observation. One day after ozonation, the effectiveness of the process against the confused flour beetle reached 50%.

The next hour of ozone treatment made it possible to reach the ozone treatment dose of 1936 mg-min/m³. The effect of ozone-air mixture on granary weevil beetles increased. Immediately after ozone treatment 80% of the pests were killed, and the remaining part was killed within 12 h of observations. Under the same conditions, the effectiveness of the ozonation process for disinfestation of confused flour beetles also increased. Thirty percent of individuals were paralyzed right after ozone treatment and then died. This indicator increased twofold in 5 hours after ozone treatment and threefold in 12 hours. One day after the observations, the mortality rate of confused flour beetles reached 100%. The general effectiveness of ozone disinfestation in the

given regime one day after the treatment was 100% against both granary weevils and confused flour beetles.

CONCLUSION

To kill the granary weevil, the ozone concentration in the ozone-air mixture in the range of 3.3-5.0 mg/m³ requires 300 minutes of treatment and a dose of 1316 mg-min/m³. The death of all pests occurs after 12 h. To destroy the confused flour beetle at the ozone-air mixture ozone concentration in the range of 3.3-5.0 mg/m³ requires 460 minutes of disinfestation and a dose of 1936 mg-min/m³. Complete death of insects occurs 24 h after ozonation.

In case of high ozone capacity of ozonator it is necessary to calculate the disinsection time by dividing the recommended doses of ozone by current concentration in a particular experiment. It is not recommended to ozone until the complete extermination of grain pests, because ozone treatment has an after-effect, which contributes to the extinction of beetles in the next day after disinsection. To stop the process of disinsection of grain heap, you can focus on the paralysis of half of the number of beetles.

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ВЛИЯНИЕ ТЕМПЕРАТУРЫ И ВЛАЖНОСТИ НА СОСТАВ ФИТОПАТОГЕНОВ ПШЕНИЦЫ ПРИ ХРАНЕНИИ ЗЕРНА

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Высокая влажность, низкая температура и болезнетворные микроорганизмы способны значительно снизить качество семян зерновых культур при их хранении. Изучено влияние температуры и влажности на поражаемость семян пшеницы фитопатогенами при хранении. Выявлено снижение зараженности семенного материала всеми видами грибов в теплом складе при 14%-й влажности. При повышении влажности до 20% происходило увеличение общей зараженности. Показана зависимость поражаемости всходов зерновых культур от условий хранения семян. При теплом режиме хранения семян при нормальной влажности (14%) выявлен наиболее высокий индекс развития болезней – в 2,9 раза выше по сравнению с холодным режимом хранения, но с аналогичной влажностью. Аналогичные данные получены и при определении распространенности корневой гнили. Наибольший показатель распространенности болезни отмечен у растений, высеянных семенами, хранящимися при влажности 20% в теплом складе, что в 1,5 раза выше по сравнению с семенами, высеянными при стандартной влажности (14%). При холодном хранении семян распространенность корневой гнили при высокой влажности (20%) составила 63% – на 7% выше, чем при стандартной (14%). При повышенной влажности (20%) как в теплом, так и в холодном складе распространенность корневой гнили выше, чем при стандартной влажности. Отмечено, что при теплом режиме хранения семян при влажности 20% органотропная специализация характеризуется приуроченностью патогенов ко всем органам растения.

Ключевые слова: яровая пшеница, режим хранения, фитопатогены, зараженность, семена, всхожесть, развитие болезни

EFFECT OF TEMPERATURE AND HUMIDITY ON THE COMPOSITION OF WHEAT PHYTOPATHOGENS DURING GRAIN STORAGE

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High humidity, low temperature and pathogens can significantly reduce the quality of cereal crop seeds during storage. The effect of temperature and humidity on the infestation of wheat seeds by phytopathogens during storage was studied. A decrease in infestation of seed material with all types of fungi in a warm warehouse at 14% humidity was found. When humidity increased to 20%, there was an increase in total infestation. The dependence of seedling infestation on seed storage conditions is shown. Warm storage mode of seed storage at normal humidity (14%) revealed the highest index of disease development – 2.9 times higher compared to cold storage mode, but with similar humidity. Similar data were obtained when determining the prevalence of root rot. The highest incidence of the disease was noted in plants sown with seeds stored at 20% humidity in a warm warehouse, which is 1.5 times higher compared to the seeds sown at standard humidity (14%). When seeds were stored cold, the prevalence of root rot at high humidity (20%) was 63% – 7% higher than at standard humidity (14%). At higher humidity (20%) in both warm and cold storage, the prevalence of root rot is higher than at standard humidity. It was noted that under warm seed storage conditions at 20% moisture content, organotropic specialization is characterized by allocation of pathogens in all plant organs.

Keywords: spring wheat, storage mode, phytopathogens, infestation, seeds, germination, disease development

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Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

Currently, the seeds of agricultural crops in the conditions of Western Siberia are affected by a whole complex of pathogens [1-3]. This leads to a number of negative factors: reduction of sowing qualities, germination, germination energy, etc. [4-6]. The main indicators of grain freshness change under the influence of microorganisms: color, gloss, odor and taste [6-9]. Mycotoxins are products of molds that are extremely toxic to animals and humans. More than 200 toxic substances released by mold fungi have been found: aflatoxins, ochratoxins, patulin, zearalenone and others, among which aflatoxins released by *A. flavus* are the most dangerous due to their particular toxicity and carcinogenicity [10].

Treatment of seeds with various substances to protect them from diseases and pests is one of the most targeted, cost-effective and environmentally friendly measures to protect plants [11]. Chemical dressing agents, which disinfect seeds quite effectively, have a number of negative properties: the high cost of treatment and environmental risks associated with their use [12].

One effective way to improve the health of seed is to optimize storage conditions with the necessary levels of temperature and humidity in the storage rooms.

The purpose of the study is to examine the effect of temperature and humidity on the infestation of wheat seeds by phytopathogens during storage.

The research objectives are to:

- determine the effect of temperature and moisture factors on the total infestation of wheat seeds with a complex of phytopathogens;
- identify the degree of influence of temperature, humidity and infestation of wheat seeds on field germination;
- determine the degree of influence of grain storage conditions on the infestation of seedlings in the field conditions.

MATERIAL AND METHODS

The studies were carried out at the research and experimental base of the Siberian Research Institute of Fodder Crops, SFSCA, RAS. The influence of various factors (temperature, humidity) on the infestation of wheat seeds was evaluated in a laboratory experiment. The aim of the experiment was to identify the degree of influence of temperature, humidity and infestation of wheat seeds on the field germination.

Wheat seeds were stored under natural conditions, previously placed in storage containers (bags). Seeds were sampled to monitor their phytosanitary status and changes in the seed quality every 30 days of storage¹. Along with this, samples were taken to determine initial seed infestation. Mycological analysis of seeds was carried out according to the generally accepted method based on creation of optimal conditions that stimulate growth and development of pathogens in order to obtain spore formation². To study the composition of

¹GOST R 52325-2005. Seeds of agricultural plants. Variety and sowing qualities. Moscow: Standartinform, 2006.

²Naumov N.A. Analysis of seeds for fungal and bacterial infection. Moscow; L.: Selkhozgiz, 1951. 137 p.

mycoflora, seeds (10 pieces each) were placed on prepared sterile Chapek's agarized medium to study seed infestation by phytopathogens ($n = 50$). The observations and records were made in 7-14 days. For this purpose, each colony of fungi was viewed under a microscope and the percentage of seed microbial infestation was calculated. Species identification of fungi was performed according to the identifiers^{3, 4} [13]. For more detailed identification, the colonies were cast on agarized medium in test tubes for subsequent identification of species.

The effect of wheat seed storage conditions on plant infestation by root rot was evaluated under field conditions in a microfield experiment [10]. The repetition of the experiment was three times. Sampling for root rot infestation was carried out during the tillering phase of grain crops according to generally accepted methods. For this purpose, all plants were washed under running water and visual analysis was carried out. During the vegetation period the record of leaf-rolling infections in grain crops was carried out in dynamics. Mathematical processing was performed using Statistica software.

RESULTS AND DISCUSSION

Phyto-examination of spring wheat seeds for their initial infestation was carried out before starting the experiment (see the table).

At 20% moisture content in seeds, the tendency to higher infection by phytopathogenic

fungi of genera *Alternaria*, *Fusarium* and *Bipolaris* was noted, although it is statistically unreliable. Associated fungi, represented by species of genera *Aspergillus* and *Mucor*, had a low level of infestation (up to 6%).

Next, seeds with different moisture content were stored at different temperature regimes (warm and cold storage).

The studies conducted on the 136th day of the experiment showed that the highest infestation with phytopathogens was found when storing grain at 20% moisture content under both cold and warm storage regimes (see Fig. 1). The level of infection with one of the main root rot pathogens by *Bipolaris sorokiniana* (Sacc.) Shoemaker exceeded the threshold of harmfulness (TH 15%) and was 16 and 32%, respectively. Representation of species of the genus *Fusarium* was significant in these samples and reached, respectively, 52 and 26%, *Alternaria* species -76 and 68%.

The highest total infection with root rot pathogens was found in samples of wheat grain stored at high humidity of cold storage (194%) and warm storage (178%) (see Fig. 2). Common mycoflora, represented by *Penicillium* and *Aspergillus* species, was also higher at humidity of 20%.

Literature data indicate [5, 8, 9] that at higher humidity the number of phytopathogens increases, since more favorable conditions are created for their development. This agrees with the results we obtained. At high-

Исходная заселенность семян пшеницы грибами перед закладкой лабораторного опыта
Initial infestation of wheat seeds with fungi before the laboratory experiment

Average number of fungi colonies in Petri dishes *				
<i>Alternaria</i>	<i>Fusarium</i>	<i>Bipolaris</i>	<i>Aspergillus</i>	<i>Mucor</i>
Humidity 14%				
6,8 ± 1,1	3 ± 1,22	1,4 ± 0,89	0,4 ± 0,89	0,6 ± 0,89
Humidity 20%				
8,6 ± 1,14	5,2 ± 1,79	4,8 ± 1,3	0	0

* Mann-Whitney U-test difference ($p < 0,05$) in comparison with the control.

³Bilai V.I. Fusariums. Kiev: Naukova Dumka, 1977. 442 p.

⁴Pidoplychko N.M. Fungi parasites of cultivated plants: Identifier in 3 volumes. Kiev: Naukova Dumka, 1977. Vol. 1.

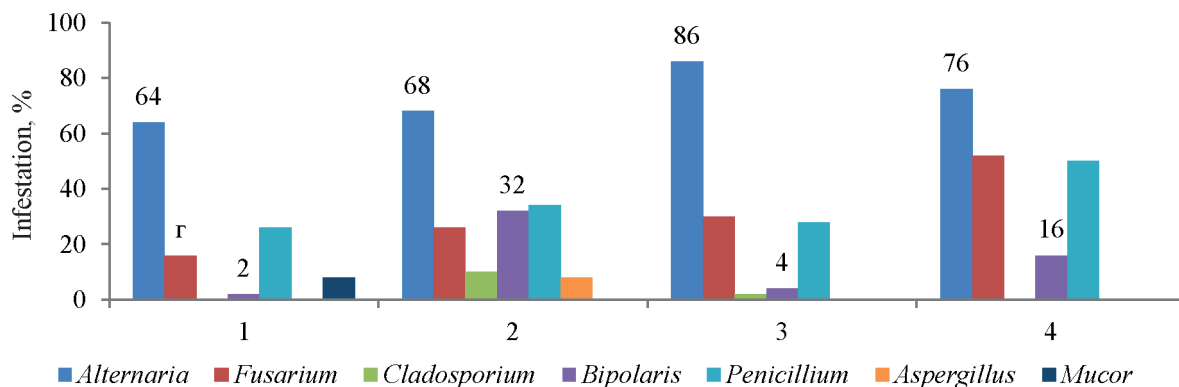


Рис. 1. Зараженность семян пшеницы:

г – значимость различия по тесту Данна ($p < 0,05$) по сравнению с соответствующим вариантом; здесь и на рис. 2–4: 1 – влажность 14%, теплый склад; 2 – влажность 20%, теплый склад; 3 – влажность 14%, холодный склад; 4 – влажность 20%, холодный склад

Fig. 1. Infestation of wheat seeds;

г - significance of the difference by Dunn's test ($p < 0.05$) compared to the corresponding variant; here and in Figs. 2-4: 1 – 14% moisture content, warm storage; 2 – 20% moisture content, warm storage; 3 – 14% moisture content, cold storage; 4 – 20% moisture content, cold storage

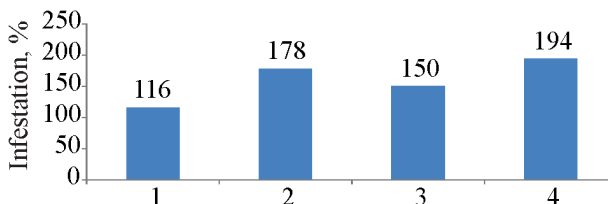


Рис. 2. Сумма зараженности пшеницы фитопатогенными грибами рода *Fusarium*, *Alternaria*, *Bipolaris*

Fig. 2. Sum of infestation of wheat by phytopathogenic fungi of *Fusarium*, *Alternaria*, *Bipolaris* genera

er humidity (20%), the level of infection of wheat seeds with representatives of genera *Alternaria*, *Fusarium*, *Bipolaris* was higher than at 14% humidity.

To find out the effect of seed storage regimes on the development of wheat diseases, a microfield experiment was carried out. The seeds under study were sown in the field. In the phase of full tillering, plants were analyzed for intensity of development and prevalence of root rot on various underground organs of spring wheat (see Fig. 3, 4).

It was found that the disease development

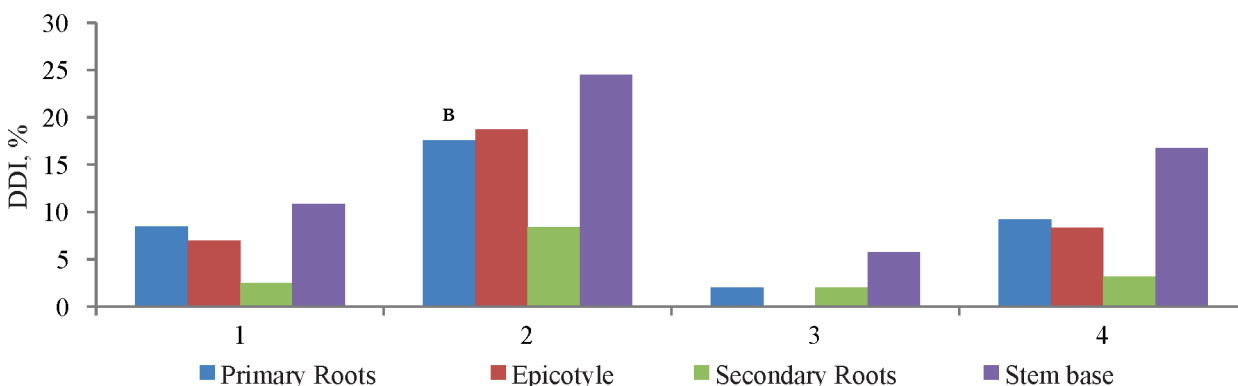


Рис. 3. Индексы развития корневой гнили пшеницы:

в – значимость различия по тесту Данна ($p < 0,1$) по сравнению с соответствующим вариантом

Fig. 3. Indices of wheat root rot development;

в – significance of Dunn's test difference ($p < 0.1$) compared to the corresponding variant

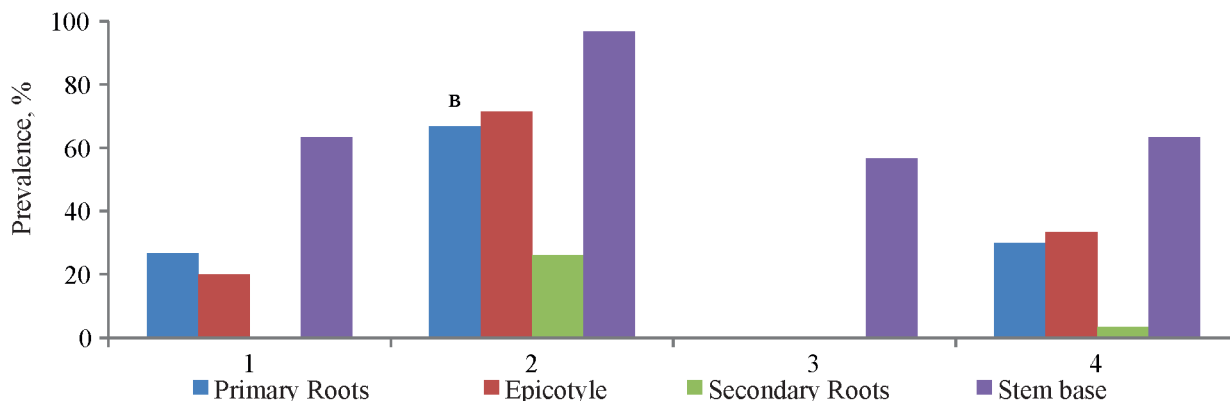


Рис. 4. Распространенность корневой гнили пшеницы;

в – значимость различия по тесту Данна ($p < 0,1$) по сравнению с соответствующим вариантом

Fig. 4. Prevalence of wheat root rot;

в - significance of Dunn's test difference ($p < 0,1$) compared to the corresponding variant

was greatest when sowing wheat seeds stored at high humidity (20%), compared with the variants where the seeds were sown with the standard humidity (14%). The level of root rot development was generally low; only in the variant with 20% humidity during warm storage it exceeded the threshold of harmfulness (TH 15%). It was 17,3% on the average per plant, which was 2,4 times higher compared to the variant with the humidity of 14% during the warm storage. Consequently, the moisture factor contributes to a more significant development of pathogens. This is confirmed by the fact that at the same humidity (20%) and cold storage the index of disease development was respectively 3.8 times higher compared to the variants with the same storage temperature regime, but at standard humidity (14%).

Warm storage mode of seed storage at normal humidity (14%) revealed the highest index of disease development - 2.9 times higher compared to the cold storage mode, but with similar humidity. This indicates that warm storage mode is more favorable for the development of pathogens, in particular root rot.

Similar data were obtained when determining the prevalence of root rot. The highest prevalence of the disease was noted in the plants sown with the seeds stored at 20% humidity in warm storage, which is 1.5 times higher compared with the plants from the seeds stored at standard humidity (14%). When seeds were

stored cold, the prevalence of root rot at high humidity (20%) was 63%, 7% higher than at standard humidity (14%). The main infestation was noted at the stem base. When the seeds were stored in cold storage at standard humidity (14%), the other plant organs were weakly affected by root rot.

The obtained data indicate that under increased humidity (20%) in both warm and cold storage the prevalence of root rot is higher than under standard humidity. It should be noted that organotropic specialization is characterized by confinement of pathogens to all plant organs in warm storage at 20% humidity.

CONCLUSIONS

1. Reduced infestation of seed material of cereal crops with all types of fungi in warm and cold storage at 14% humidity was detected. The highest values of total infestation were observed at humidity of 20% in a warm warehouse, indicating the importance of moisture and storage temperature factor for the development of mycoflora.

2. Dependence of wheat seedlings infestation on grain storage conditions was shown. The lowest values of wheat root rot prevalence and development were found in seedlings obtained from the seeds stored at normal humidity (14%) in a cold store. Thus, one of the significant factors for seedlings infestation is storage temperature and humidity of the seed material.

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ЭКСТЕРЬЕРНЫЙ ПРОФИЛЬ ЗААНЕНСКИХ КОЗЛОВ С РАЗЛИЧНЫМИ ГЕНОТИПАМИ ГЕНА *SPAG17*

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Экстерьер формируется в процессе роста и развития животного и является отражением внешнего вида и пропорций тела. Значимую роль в формировании экстерьера играет генетический фактор. Белок спермо-ассоциированный антиген 17, кодируемый геном *SPAG17*, выполняет ряд важных биологических функций в процессе роста и развития организма млекопитающих, в том числе влияет на рост и развитие костей. Целью нашей работы был анализ полиморфных вариантов (indel) гена *SPAG17* в связи с показателями промеров тела половозрелых козлов зааненской породы. В выборку включены 43 козла в возрасте 3–5 лет, принадлежащих одному из племхозов Ленинградской области. Для выделения образцов ДНК использовали метод фенольной экстракции. Генотипирование проводили по rs659761737 (indel 14 п.н., интрон 22) и по rs647063466 (indel 17 п.н., интрон 47) гена *SPAG17* методом АС-ПЦР. Перед началом случного сезона однократно проводили обмер половозрелых козлов зааненской породы с вычислением индексов телосложения. Анализ частоты генотипов и аллелей показал, что по rs659761737 (indel 14 п.н.) генотип DD определен на уровне 0,186, ID – 0,419 и II – 0,395. Для rs647063466 (indel 17 п.н.) гена *SPAG17* отмечена противоположная картина. Частота генотипов DD, ID и II составила 0,326; 0,512 и 0,163 соответственно. Животные с генотипом II по rs659761737 (indel 14 п.н.) гена *SPAG17* имели достоверно высокие значения обхвата пясти ($p < 0,01$), особи с генотипом ID по rs647063466 (indel 17 п.н.) отличались высокими значениями индекса высоконогости ($p < 0,05$) и грудного индекса ($p < 0,05$). Полученные данные позволяют предположить, что изучаемые SNP гена *SPAG17* вносят существенный вклад в формирование экстерьерного профиля козлов зааненской породы.

Ключевые слова: зааненская порода коз, indel-полиморфизм, индекс, промеры тела

EXTERIOR PROFILE OF SAANEN GOATS WITH DIFFERENT GENOTYPES OF THE *SPAG17* GENE

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The exterior is formed in the process of growth and development of the animal and is a reflection of the appearance and proportions of the body. Genetic factors play a significant role in the formation of the exterior. The protein sperm-associated antigen 17, encoded by the *SPAG17* gene, performs a number of important biological functions in the process of growth and development of the mammalian organism, as well as affecting the growth and development of bones. The aim of our

work was to analyze the polymorphic variants (indel) of the *SPAG17* gene in connection with the body measurements of mature Saanen goats. The sample included 43 goats aged 3-5 years from one of the breeding farms of the Leningrad region. Phenol extraction was used to isolate DNA samples. Genotyping was performed for rs659761737 (indel 14 bp, intron 22) and rs647063466 (indel 17 bp, intron 47) of the *SPAG17* gene by AS-PCR method. Before the start of the breeding season, sexually mature goats of the Saanen breed were once measured with the calculation of body built indices. The analysis of the genotypes and alleles frequency showed that, according to rs659761737 (indel 14 bp), the DD genotype was determined at the level of 0.186, ID - 0.419, and II - 0.395. For rs647063466 (indel 17 bp) of the *SPAG17* gene, the opposite pattern was observed. The frequency of DD, ID, and II genotypes was 0.326, 0.512, and 0.163, respectively. Animals with genotype II for rs659761737 (indel 14 bp) of the *SPAG17* gene had significantly high metacarpus girth values ($p < 0.01$), and the individuals with genotype ID for rs647063466 (indel 17 bp) were distinguished by high values of the index of leg height ($p < 0.05$) and chest index ($p < 0.05$). The obtained data suggest that the studied SNPs of the *SPAG17* gene make a significant contribution to the formation of the exterior profile of the Saanen goats.

Keywords: Saanen goats, indel-polymorphism, index, body measurements

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Конфликт интересов

Авторы заявляют об отсутствии конфликта интересов.

Conflict of interest

The authors declare no conflict of interest.

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INTRODUCTION

Dairy goat breeding in Russia is a successfully developing and promising branch of dairy cattle breeding. In 2020, the Saanen breed accounted for 82.8% of all goat breeds with a total herd of 17,901 heads¹. Exteriors are important for breeding animals and are formed in the process of growth and development of the animal and reflect the appearance and proportions of the body. The exterior is determined by the breed peculiarities and productive qualities of the animal, as well as a reflection of the biological adaptation of the organism to environmental conditions. A breeding animal must have a correct exterior shape and have a strong body composition².

Among many paratypic factors, genetic factor plays a key regulatory role in the formation of exteriors. Currently, research on the search and study of candidate genes associated with different phenotypes of animals is relevant, because the use of effective DNA markers in breeding can help breeders in the selection of animals with high genetic potential [1].

Sperm-associated antigen 17 protein, encoded by the *SPAG17* gene, performs many biological functions in the process of growth and development of the mammalian organism. A number of studies have shown that it is essential for male fertility, as homozygous mutations in its exons are reliably associated with severe asthenozoospermia [2]. It also plays a signifi-

¹Safina G.F., Chernov V.V., Khatataev S.A., Khmelevskaya G.N., Stepanova N.G., Pronin A.V., Baglay I.M. Yearbook on breeding work in sheep and goat breeding in the farms of the Russian Federation (2019). Lesnye Polyany: All-Russian Research Institute of Breeding, 2020. 344 p.

²Tsaregorodtseva E., Toshev V. Goat breeding. Textbook for Higher Education Institutions. Litres, 2021. 361 p.

cant role in the survival of newborn individuals and influences bone growth and development [3]. Some SNPs of *SPAG17* gene have been associated with idiopathic low growth in humans [4]. Recent studies confirm that some polymorphic variants of this gene are associated with the body measurements [5] and goat fertility [6]. Thus, the *SPAG17* gene can be considered as a promising candidate gene in the selection of farm animals.

The purpose of this work is to analyze the polymorphic variants (indel) of the *SPAG17* gene in relation to the body measurements of sexually mature Saanen breed goats.

Research objectives are:

- to measure sexually mature goats of the Saanen breed and calculate body indices for sexually mature goats;
- to do the genotyping of animals by indel polymorphic variants of the *SPAG17* gene, and estimate the frequency of genotypes and alleles;
- to study the relationship between different genotypes of the *SPAG17* gene and the parameters of body measurements and body mass indexes of animals.

MATERIAL AND METHODS

The material for the study was DNA isolated from the venous blood of sexually mature goats aged 3-5 years ($n = 43$) belonging to one of the breeding farms of the Leningrad Region. Phenolic extraction method was used to isolate the DNA samples. Genotyping of the *SPAG17* gene by rs659761737 (indel 14 bp) and rs647063466 (indel 17 bp) was performed by AS-PCR (see Table 1) [5]. PCR was performed on a Thermal Cycler T100 amplifier (Bio-Rad, USA).

Body measurements of goats were measured once before the start of the breeding season (July). The measurements were taken in centimeters: height at the withers, height at hips, chest depth, chest width, chest girth, cannon bone girth, and oblique body length. In addition, basic body mass indices were calculated as a percentage³. All animals were kept in the same housing and feeding conditions at the time of the study.

STATISTICA 13 from Dell Inc (2016, software.dell.com) was used to process the obtained numerical data. Significance of the difference between the compared values was assessed using the Kruskal-Wallis test.

RESULTS AND DISCUSSION

Information on animal genotypes was obtained from the analysis of electrophoregram data (see Fig. 1, 2).

In the analyzed group of animals, three genotypes were determined for the two SNPs studied. A 183-bp fragment was determined for the Ins variant (allele I) and a 169-bp fragment for the Del variant (allele D) according to rs659761737 (indel 14 bp). A 241-bp fragment corresponded to allele I according to rs647063466 (indel 17 bp), for allele D - 224 bp long. Analysis of the frequency of genotypes and alleles for the *SPAG17* gene showed that rs659761737 revealed a low frequency of allele D and only 18.6% of animals had the homozygous genotype DD (see Table 2). For rs647063466 of the *SPAG17* gene, the opposite picture was observed and only 16.3% of the individuals had homozygous genotype II.

Табл. 1. Характеристика праймеров, условия ПЦР

Table 1. Characterization of primers, PCR conditions

SNP	Primer	Position	Primer annealing temperature	Allele	PCR product size
rs659761737 (indel 14 bp)	F:GAGGGAATGTGAGCAGGAT	Intron 22	58–60 °C	I	183 p.o.
	R:TTTGATGACAAGGAAGGGA			D	169 p.o.
rs647063466 (indel 17 bp)	F:AAGTTCAGGGAGTGTTAAGGA	Intron 47		I	241 p.o.
	R:CTGTGCCAGACAGATGGTC			D	224 p.o.

³Kulikova N.I. Sheep and goat breeding: textbook. Krasnodar: KubSAU, 2017. 193 p.

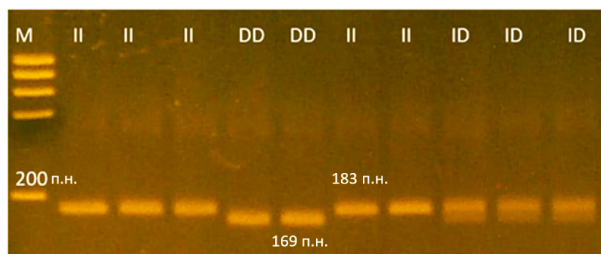


Рис. 1. Электрофореграмма фрагментов ДНК по rs659761737 гена *SPAG17* (indel 14 п.н.). М-маркер BIORON GmbH ДНК-маркер 100bp + (100bp plus DNA Ladder ready-to-use, Медиген)

Fig. 1. Electropherogram of DNA fragments at rs659761737 of the *SPAG17* gene (indel 14 bp). M-marker BIORON GmbH DNA marker 100bp+ (100bp plus DNA Ladder ready-to-use, Medigen)

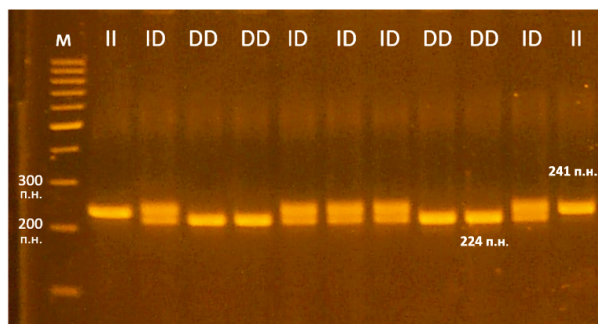


Рис. 2. Электрофореграмма фрагментов ДНК по rs647063466 гена *SPAG17* (indel 17 п.н.). М-маркер 50 + bp DNA Ladder (Евроген)

Fig. 2. Electropherogram of DNA fragments at rs647063466 of the *SPAG17* gene (indel 17 bp). M-marker 50 + bp DNA Ladder (Evrogen)

A comparative analysis of the mean values of the rs659761737 measures of the *SPAG17* gene showed that animals with genotype II were significantly superior to animals in the other groups in terms of metacarpal girth ($p < 0.01$, $p < 0.05$) (see Table 3).

Analysis of rs647063466 of the *SPAG17* gene showed that animals with the ID genotype were characterized by lower breast depth ($p < 0.01$) but were characterized by higher breast and high-legged indices ($p < 0.05$) compared with individuals with the DD genotype (see Table 4).

The results of our research showed that in the analyzed sample of goats of the Saanen breed there was a high frequency of allele I by rs659761737 (indel 14 bp) and allele D by rs647063466 (indel 17 bp) of the *SPAG17* gene. The available literature data indicate that the distribution of allele frequencies depends on

the breed and the direction of goat productivity, which may indicate a significant contribution of the analyzed polymorphic variants of the *SPAG17* gene in shaping the exterior of animals. In a study by S. Zhang et al (2019) two local cashmere goats of meat-wool type and Hainan black goats of meat type. The results showed that the white cashmere goat group had a high frequency of allele D and genotype DD in both indel 14 bp and indel 17 bp of the *SPAG17* gene (0.917 and 0.847; 0.787 and 0.610, respectively). However, the animals with genotype II by two indel polymorphisms reliably differed by a larger build and had high values of height at the withers, width at the chest, and body length. Hainan black goats were distinguished by uniform distribution of alleles at the analyzed indel-polymorphic loci. Animals with different genotypes did not differ significantly in the ana-

Табл. 2. Распределение частот генотипов и аллелей гена *SPAG17* в анализируемой группе животных
Table. 2. Frequency distribution of *SPAG17* genotypes and alleles in the analyzed group of animals

SNP	n	Genotype frequency		Allele frequency	
rs659761737 (indel14 bp)	8	DD	0,186	I	0,605
	18	ID	0,419	D	0,395
	17	II	0,395		
rs647063466 (indel17 bp)	14	DD	0,326	I	0,419
	22	ID	0,512	D	0,581
	7	II	0,163		

Табл. 3. Средние значения промеров тела козлов с различными генотипами гена *SPAG17* (rs659761737, indel 14 bp)

Table 3. Mean measurements of the goats body with different genotypes of the *SPAG17* gene (rs659761737, indel 14 bp)

Indicator (Mean ± Std.Err)	Genotype		
	DD	ID	II
	n = 8	n = 18	n = 17
Height, cm:			
at the withers	84,06 ± 2,00	89,10 ± 1,32	87,56 ± 1,87
at hips	84,06 ± 2,00	87,17 ± 0,81	88,11 ± 1,38
Oblique body length, cm	98,31 ± 2,60	100,53 ± 1,17	101,59 ± 1,56
Chest depth, cm	41,88 ± 1,59	42,50 ± 0,59	41,79 ± 0,49
Chest width, cm	26,81 ± 0,70	26,88 ± 0,60	27,47 ± 0,52
Girth, cm:			
chest	112,75 ± 2,60	115,0 ± 1,088	114,18 ± 1,24
cannon bone	11,50 ± 0,19 ^{a, c}	12,03 ± 0,15 ^d	12,39 ± 0,19 ^b
Index, %:			
stretchiness	117,12 ± 2,71	113,00 ± 1,07	117,05 ± 3,71
chest	64,31 ± 1,40	63,26 ± 1,06	65,90 ± 1,51
high-legged	49,96 ± 2,36	52,18 ± 0,80	51,85 ± 1,41
boniness	13,73 ± 0,38	13,56 ± 0,30	14,25 ± 0,36
compactness	114,97 ± 2,68	114,66 ± 1,37	112,65 ± 1,47
massiveness	134,56 ± 4,14	129,52 ± 1,81	131,31 ± 3,07

Note. a, b when $p < 0,01$; c, d when $p < 0,05$.

Табл. 4. Средние значения промеров козлов с различными генотипами гена *SPAG17* (rs647063466, indel 17 bp)

Table 4. Mean measurements of the goats body with different genotypes of the *SPAG17* gene (rs647063466, indel 17 bp)

Indicator (Mean ± Std.Err)	Genotype		
	DD	ID	II
	n = 14	n = 22	n = 7
Height, cm:			
at the withers	86,75 ± 2,22	87,97 ± 1,22	87,86 ± 2,43
at hips	87,11 ± 1,10	86,50 ± 1,12	88,11 ± 2,30
Oblique body length, cm	102,21 ± 1,49	100,09 ± 1,36	98,57 ± 2,09
Chest depth, cm	43,35 ± 0,83 ^b	41,02 ± 0,49 ^a	43,01 ± 0,89
Chest width, cm	26,90 ± 0,44	26,99 ± 0,43	27,86 ± 1,45
Girth, cm:			
chest	114,07 ± 1,62	114,45 ± 1,12	114,21 ± 1,76
cannon bone	11,93 ± 0,22	12,21 ± 0,16	11,93 ± 0,23
Index, %			
stretchiness	119,02 ± 4,23	113,96 ± 1,35	112,49 ± 2,61
chest	62,23 ± 1,13 ^c	65,90 ± 1,07 ^d	64,64 ± 2,46
high-legged	49,59 ± 1,69 ^c	53,20 ± 0,84 ^d	50,82 ± 1,65
boniness	13,89 ± 0,49	13,92 ± 0,21	13,65 ± 0,50
compactness	111,75 ± 1,61	114,61 ± 1,27	116,12 ± 2,60
massiveness	132,57 ± 3,84	130,46 ± 1,66	130,58 ± 4,03

Note. a, b when $p < 0,01$; c, d when $p < 0,05$.

lyzed indices. The data obtained in our study on the body measurements of mature Saanen goats indicate that the animals are properly developed and correspond to the dairy type of productivity. Based on the 14 bp of the *SPAG17* gene, the animals with genotype II were characterized by reliably high values of the metacarpal girth. This indicator is used to judge the overall strength of the animal's constitution, which to some extent can be associated with the fertility and viability of individuals. According to the data of the work [7], the metacarpal girth of bulls had a significant genetic correlation with the milk yield of the daughters in 305 days of lactation ($r = 0.460$), which, according to the author, indicates that animals with a strong constitution have a high adaptive capacity.

In our study, animals with genotype ID by indel 17 bp of the *SPAG17* gene were distinguished by high values of chest and high-legged indices, which indicates that the animals are large and have a well-developed chest. The chest measurements are primarily a reflection of the intensity of development of the animal's axial skeleton. For goats of dairy breeds a developed rib cage indicates the potential of animals for high milk productivity [8]. In studies [7], the chest girth indicator of bulls had a positive correlation with the milk yield of the daughters.

In goat breeding, exterior performance is a reflection of breed standards and an integral part of breeding in the development of selection and breeding criteria. According to the research [9], the main indicators of measurements have a reliably positive relationship with the milk yield of animals in Saanen goats. Similar data were obtained in other goat breeds. Linear body measurements were positively correlated with the milk productivity in Indonesian Etawa goats [10], native Turkish Kilis goats [11], and in Bedouin goats of the Sahara Desert [12]. In this regard, selecting large individuals for breeding will improve the milk productivity of herds.

CONCLUSION

Indel polymorphism variants of the *SPAG17* gene are associated with some indicators of the exterior of sexually mature goats of the Saanen breed. The animals with genotype II by

rs659761737 (indel 14 bp) of *SPAG17* gene had reliably high values of metacarpal girth, the individuals with genotype ID by rs647063466 (indel 17 bp) had high values of high-legged and chest indices. The data suggest that indel mutations in the *SPAG17* gene can be used as a DNA marker in marker-assisted selection in goat breeding to produce large animals with high milk production potential. However, these data are preliminary, as they were obtained on a small herd of goats and require further study.

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ОЦЕНКА МОЛОЧНОЙ ПРОДУКТИВНОСТИ И КАЧЕСТВА МОЛОКА КОЗ РАЗНЫХ ГЕНОТИПОВ В ЗАВИСИМОСТИ ОТ ЧИСЛА ЛАКТАЦИЙ

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Приведены результаты оценки динамики молочной продуктивности коз разных генотипов в зависимости от числа лактаций за 305 дней. Изучен уровень среднемесячных удоев. Для проведения опыта сформировали четыре группы коз зааненской и нубийской пород по 10 гол. в каждой. Начиная с 1-го месяца лактации во всех группах удой плавно увеличивался до 4-го месяца, затем к завершению лактации уменьшался. Максимальные среднемесячные удои приходились на 2–4-й месяцы лактации во всех четырех группах козоток. Наибольшие показатели по среднемесячным надоям были у коз всех генотипов и лактаций в 4-м месяце. Козы зааненской породы доминировали над козами нубийской по первой лактации на 14,43 кг, или 15,96%, по третьей – на 4,23 кг, или 4,48%. Исследованы показатели физико-химического и микробиологического состава молока коз зааненской и нубийской пород разного числа лактаций. Содержание основных питательных веществ молока: белка, жира и молочного сахара, а также минеральных веществ – выше у коз нубийской породы в обеих лактациях. Показатели плотности и кислотности козьего молока соответствовали ГОСТу. Точка замерзания образцов козьего молока коз обеих пород и лактаций колебалась в пределах 0,50–0,57 °С. Содержание соматических клеток в молоке коз обоих генотипов находилось в пределах от 128 до 1500 тыс./см³ и соответствовало нормам. Бактериальная обсемененность молока коз обеих пород и лактаций не превышала допустимых санитарных норм.

Ключевые слова: козы, зааненская порода, нубийская порода, молочная продуктивность, лактация, белок, молочный жир, лактоза

EVALUATION OF MILK PRODUCTIVITY AND MILK QUALITY OF GOATS OF DIFFERENT GENOTYPES DEPENDING ON THE NUMBER OF LACTATIONS

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The results of assessing the dynamics of milk productivity of goats of different genotypes depending on the number of lactations in 305 days are presented. The level of average monthly milk yields was studied. Four groups of Saanen and Nubian breed goats of 10 animals each were formed for the experiment. Starting from the 1st month of lactation in all groups, milk yield increased continuously until the 4th month, and then went down by the end of lactation. The maximum average monthly milk yields were in the 2nd-4th months of lactation in all four groups of female goats. Average monthly milk yields were highest for goats of all genotypes and lactations in the 4th month. Saanen goats dominated over Nubian goats in the first lactation by 14.43 kg, or 15.96%, in the third by 4.23 kg, or 4.48%. The parameters of physicochemical and microbiological composition of the milk of Saanen and Nubian goats of different lactation numbers were studied. The content of the main milk nutrients: protein, fat and milk sugar, as well as minerals is higher in the Nubian goat breed in both lactations. The density and acidity of goat milk were in accordance with the GOST (All Union State standard). The freezing point of goat milk samples of both breeds and lactations ranged from 0.50-0.57 °C. The somatic cell content in the milk of goats of both genotypes ranged from 128

to 1500 thousand/cm³ and conformed to the standards. Bacterial contamination of milk of the goats of both breeds and lactations did not exceed permissible sanitary standards.

Keywords: goats, Saanen breed, Nubian breed, milk productivity, lactation, protein, milk fat, lactose

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Конфликт интересов

Авторы заявляют об отсутствии конфликта интересов.

Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

Goat breeding is a traditional source of high-quality milk and a wide range of processed products. The most numerous and widespread dairy goat breed in the Russian Federation is the Saanen, and recently the Nubian breed as well. Goats of these breeds, genotypes and populations have unequal indicators of milk productivity and quality characteristics of milk, so there is a question of conducting a comprehensive assessment of these indicators in order to a more detailed and comprehensive application in the livestock industry [1-3].

Currently, goat milk is in high demand in the world, due to the high demand for ecologically pure and natural nutritional products. Numerous studies have established and proved the high biological value of goat milk, because in many of its properties it is the closest to women's milk, therefore, it is beneficially absorbed not only by adults, but also by children [4-8].

The purpose of the study is to study milk productivity, physical and chemical and microbiological parameters of goat milk of different genotypes according to the number of lactations.

MATERIAL AND METHODS

The work was performed from 2020 to 2022 at Saratov State Agrarian University named after N.I. Vavilov. The studies were carried out on the basis of LLC "ZooCenter Harmonia" (Saratov region). Four groups of 10 goats of Saanen

and Nubian breeds were formed. All animals were at the end of the 1st and at the beginning of the 2nd month of lactation. Feeding and housing conditions of the goats were identical. Feeding rations were made taking into account live weight and productivity of the goats according to the norms of the Stavropol Research Institute of Animal Husbandry and Forage Production. The physical and chemical composition of milk, as well as its properties, were studied according to generally accepted methods in the training research and testing laboratory for determining the quality of food and agricultural products at Saratov State Agrarian University named after N.I. Vavilov.

RESULTS AND DISCUSSION

Milk productivity, physical, chemical and microbiological composition, as well as the properties of milk are influenced by such factors as breed, age, lactation period, composition of diets, etc.

Data on milk productivity of goats of different genotypes given by L.N. Grigoryan and other authors [9] show that goats of the Saanen breed type had high milk yield per lactation - 663 kg, goats of the Nubian breed type were characterized by high fat content in milk - 5.68%. Goats of Alpine and La Mancha breeds had intermediate value.

S.A. Khatatayev et al. [10] point out that the milk productivity, composition and properties of goats' milk, in addition to the breed, are influenced by the lactation period, on which the

yield and quality of finished dairy products depend. In their studies, the analysis of the milk productivity of goats showed that the milk yield for 305 days of lactation in Saanen goats averaged 630 kg with an average daily milk yield of 2.15 kg, which is quite satisfactory for animals of this breed.

The results of the experiment on milk productivity of goats are given by I.V. Zasemchuk and M.V. Berdanova. In their work they note that according to the milk yield for 305 days of lactation the goats of Saanen breed exceeded their counterparts of Alpine and La Mancha breeds by 78.61 and 23.87 kg respectively [7].

In our studies, the dynamics of milk productivity of Saanen and Nubian goats of different lactations over 305 days showed different levels of average monthly milk yields (see Table 1).

Saanen goat females were the leaders in the amount of milk yielded in both first and third lactations over Nubian goats. The difference was 86.81 kg in the first lactation and 24.86 kg in the third ($p \geq 0.999$).

Starting from the 1st month of lactation to the 4th month of lactation, the milk yield gradually increased in all groups and gradually decreased towards the end of lactation. The maximum average monthly milk yields were in the 2nd-4th months of lactation in all four groups of female goats. The highest average monthly milk yields were observed for the goats of all genotypes and lactations in the 4th month. At the same time Saanen breed goats dominated over Nubian goats in the first lactation by 21.54 kg or 25.05%, in the third by 7.23 kg or 6.68% ($p \geq 0.999$).

Average monthly milk yields in 8-10 months of lactation were significantly lower and practically the same in all groups. The minimum value of average monthly milk yields was observed at the 10th month of lactation in goats of both genotypes, which is explained by the gestation period of the experimental animals, but the indicators of Saanen goats were better than those of the Nubian goat breed. Their difference

Табл. 1. Динамика среднемесячных удоев коз разных генотипов и лактаций, кг ($n = 10$)

Table 1. Dynamics of average monthly milk yields of goats of different genotypes and lactations, kg ($n = 10$)

Indicator	Breed			
	Saanen		Nubian	
	First lactation	Third lactation	First lactation	Third lactation
Lactation month:				
1-st	48,46 ± 0,46***	54,51 ± 0,50	44,23 ± 0,35	53,48 ± 0,49
2-nd	61,57 ± 0,65***	67,62 ± 0,62*	51,00 ± 0,60	65,25 ± 0,58
3-rd	95,42 ± 0,81***	100,31 ± 0,99 **	76,83 ± 0,76	96,15 ± 0,78
4-th	107,53 ± 1,50***	115,47 ± 0,67 ***	85,99 ± 0,91	108,24 ± 0,61
5-th	63,14 ± 0,66***	70,23 ± 0,63*	55,86 ± 0,65	67,94 ± 0,59
6-th	56,12 ± 0,60***	62,17 ± 0,51*	47,42 ± 0,57	60,67 ± 0,48
7-th	47,64 ± 0,63***	53,67 ± 0,40	45,25 ± 0,56	52,54 ± 0,41
8-th	43,34 ± 0,58***	49,41 ± 0,34 *	38,00 ± 0,46	48,26 ± 0,36
9-th	30,27 ± 0,59***	36,15 ± 0,30***	26,16 ± 0,45	34,41 ± 0,32
10-th	22,12 ± 0,42***	26,57 ± 0,31 ***	18,06 ± 0,41	24,31 ± 0,29
Over the lactation period	575,61 ± 2,84***	636,11 ± 2,59 ***	488,80 ± 2,50	611,25 ± 2,44

* $p \geq 0,95$.

** $p \geq 0,99$.

*** $p \geq 0,999$.

was 4.06 kg or 22.48% in the first lactation and 2.26 kg or 4.85% in the third ($p \geq 0.999$).

Saanen goats of both lactations exceeded Nubian goats in average monthly milk yield in the 1st month of lactation by 9.56 and 1.93%, in the 2nd - by 20, 73 and 3.63%, in the 3rd by 24.20 and 4.33%, in the 5th by 13.03 and 1.87%, in the 6th by 18.35 and 2.47%, in the 7th by 5.28 and 2.15%, in the 8th by 14.05 and 2.38%, in the 9th by 15.71 and 5.06%, in the 10th by 22.48 and 9.30% respectively.

During the whole 9 months of the first lactation the reliable superiority ($p \geq 0,999$) of Saanen breed goats over their Nubian counterparts was noted which was 86,81 kg according to the results of the first lactation ($p \geq 0,999$). A similar pattern was observed in the third lactation, but at the 7th month of lactation no reliable difference between the milk yields of goats of both breeds was observed. But at the end of the third lactation the Saanen goats had an advantage over their Nubian counterparts by almost 25 kg ($p \geq 0.999$). It should be noted that Nubian goats in this farm are well enough matured by the third lactation, then their productivity is less inferior to Saanen goats.

A number of authors: I.V. Zasemchuk and M.V. Berdanova [7], A.M. Karpenya et al. [1], A. Orazov et al. [11], A. B. Ospanov et al. [12], E. L. Revyakin et al. [13], A.V. Tkachev [3], N.G. Chamurliiev et al. [14], A.P. Nikitina et al. [15], N.G. Chamurliiev et al. [16] believe that when studying the milk productivity of goats, it is extremely important to take into account the indicators of the physical, chemical, microbiological composition and properties of their milk, since the nutritional value and quality of dairy products prepared from it depend on them.

E.M. Shchetinina and Z.R. Khodyreva [17] cite studies on goat milk quality and note that Nubian goats have the highest protein content in milk as compared to Czech, Saanen and Toggenburg breeds.

V.V. Brunchugin and A.S. Shuvarikov. [18] note that the highest content of milk solids in their experiment was in the goats of Nubian breed. They explain this by the fact that the animals of this breed compared to Saanen breed

have higher level of nonfat milk solids, fat and lactose mass fraction in the milk.

When determining the nutritional value and technological properties of goat milk, the highest content of the mass fraction of fat, protein, casein, dry matter, milk sugar, minerals and nonfat milk solids was found in the milk of goats of the Nubian breed (see Table 2).

The milk of the Nubian goat breed contains more dry matter than its Saanen counterparts in both lactations studied (see Table 2). Accordingly, the content of the main nutrients in milk: protein, fat and milk sugar, as well as minerals, is higher in Nubian goats. However, these differences are not significant. Nevertheless, they are consistent with the studies of other scientists confirming the higher fat content and protein content in milk of the Nubian goat breed.

The nutrient content of the milk of the Saanen and Nubian goats is shown in the figure.

The milk density of the studied goat breeds was from 1027.5 to 1029.0 kg/m³, which meets the requirements of the GOST 32940-2014 "Goat milk. Technical conditions", with the Nubian breed having higher data on this indicator than the Saanen goats.

The highest indicator of titratable acidity was also found in Nubian goats, which is explained by the high content of proteins and minerals in their milk.

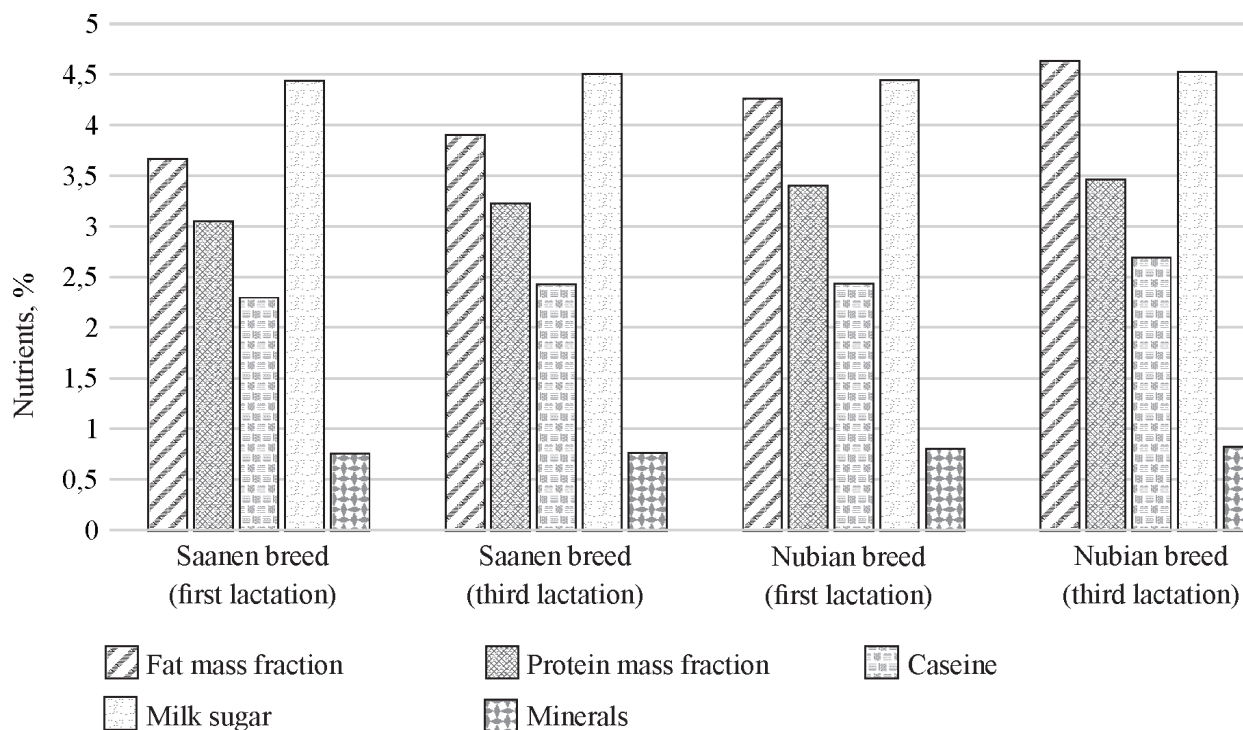
The freezing point is an important indicator in determining the quality of milk. The standard for cattle milk is a freezing point of -0.52 to -0.55 °C. When the freezing point is within 0 °C, there is reason to believe that it is diluted with water and can be classified as adulterated. When determining the freezing point of goat milk samples of both breeds and lactations it ranged from 0,50-0,57 °C. It should be noted that the milk of Nubian goats had a slightly lower freezing point than that of Saanen goats.

Numerous studies have established [1, 19] that goat milk contains an increased number of somatic cells in contrast to cow milk. It is known that this peculiarity is related to the peculiar excretion of milk from goats' udders. When milk is excreted, parts of cell membranes are excreted together with it, so if the test for cow's milk is applied, it will reveal them additionally

Табл. 2. Физико-химические и санитарно-гигиенические показатели молока подопытных коз ($n = 10$)
Table 2. Physico-chemical and sanitary-hygienic parameters of milk of experimental goats ($n = 10$)

Indicator	Breed			
	Saanen		Nubian	
	First lactation	Third lactation	First lactation	Third lactation
Dry matter, %	11,85 ± 0,17	12,34 ± 0,14	12,86 ± 0,19	13,39 ± 0,20
MSNF, %	8,20 ± 0,11	8,45 ± 0,09	8,61 ± 0,12	8,77 ± 0,10
Fat mass fraction, %	3,65 ± 0,08	3,89 ± 0,04	4,25 ± 0,05	4,62 ± 0,07
Protein mass fraction, %	3,04 ± 0,08	3,21 ± 0,09	3,39 ± 0,07	3,45 ± 0,06
Caseine, %	2,28 ± 0,07	2,41 ± 0,09	2,42 ± 0,06	2,68 ± 0,07
Milk sugar, %	4,42 ± 0,17	4,49 ± 0,15	4,43 ± 0,15	4,51 ± 0,18
Minerals, %	0,74 ± 0,06	0,75 ± 0,04	0,79 ± 0,05	0,81 ± 0,03
Calories, kcal/100 g	64,53	67,75	68,93	74,23
Milk density, kg/m ³	1027,5 ± 0,11	1028,3 ± 0,13	1028,6 ± 0,12	1029,0 ± 0,13
Milk acidity, ° T	18,02 ± 0,36	18,34 ± 0,41	17,79 ± 0,38	18,36 ± 0,42
Freezing temperature, ° C	-0,50	-0,52	-0,55	-0,57
Somatic cells, thous. /cm ³	438,4 ± 10,2***	442,6 ± 10,6***	515,3 ± 13,6***	524,6 ± 14,2***
Bacterial infestation, thous. /cm ³	Up to 300	Up to 300	Up to 300	Up to 300

*** $p \geq 0,999$.



Содержание питательных веществ в молоке коз разных генотипов, %
 The ratio of nutrients in the milk of goats of different genotypes, %

as somatic cells. Our research showed that the content of somatic cells in the milk of goats of both genotypes was 128 - 1500 thousands/cm³ ($p \geq 0,999$) and met the sanitary norms and requirements of the Technical Specifications 9837- 001 for the highest grade of goat milk.

All world standards for assessing the quality of milk regulate the level of the index of infestation with various forms of microorganisms, which is informative enough to show in what sanitary and hygienic conditions milk is obtained [20].

The norm for bacterial insemination is from 100 to 500 thousand/cm³. Our studies have shown that bacterization of milk of goats of both breeds and lactations does not exceed the acceptable sanitary standards.

CONCLUSIONS

1. Dairy productivity of Saanen goats in the first and third lactations in 305 days is higher than that of the Nubian goat breed by 15.08 and 3.91%, respectively.

2. The milk of Nubian goats, depending on the number of lactations, prevails over the milk of Saanen goats in terms of physical and chemical composition: the content of the mass fraction of fat, protein, casein, dry matter, milk sugar and minerals. The indicators of density and acidity of goat milk corresponded to the GOST. The freezing temperature of milk in goats of both breeds and lactations was within the acceptable values for milk of the highest category. Somatic cells and bacterial contamination were within the sanitary norm limits.

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ПАЗАРИТОЦЕНОЗЫ ДИКОЙ СВИНЬИ (*SUS SCROFA*) НА ТЕРРИТОРИИ ЗАБАЙКАЛЬСКОГО КРАЯ

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Проведено комплексное ветеринарное исследование популяции дикой свиньи на территории Забайкальского края на гельминтозы и микробоносительство. Изучена гельминтофауна диких свиней, установлена циркуляция возбудителей бактериальных болезней в популяции этих животных. Объектом исследований стали материалы, полученные в полевых исследованиях в районах Забайкальского края. Изучение пораженности эндопаразитами проведено у 37 диких свиней в возрасте от 6 мес до 3 лет в течение 2019–2021 гг. На территории Забайкальского края зарегистрирована зараженность диких свиней восемью видами различных гельминтов. Максимальная экстенсивность инвазии (32,4%) зарегистрирована у *Setaria labiato-papillosa*. Дикie свиньи заражены как имагинальными гельминтами *Setaria labiato-papillosa*, *Metastrongylus elongatus*, *Ascaris suum*, *Trichocephalus suis*, *Oesophagostomum dentatum*, так и личинками гельминтов *Cysticercus tenuicollis* и *Cysticercus cellulosae*. Изучение морфологии половозрелых возбудителей свидетельствует о паразитировании у свиней гельминтов рода сетария, вида *Setaria labiato-papillosa*. Из 37 исследованных диких свиней у 12 животных зафиксирована ассоциативная инвазия сетариоз + аскаридоз, у трех свиней одновременно паразитировали аскариды + трихоцефалы, у двух животных зарегистрирована ассоциация сетариоз + метастронгилез. У пораженных ассоциативной инвазией свиней при органолептическом исследовании туши отмечено уменьшение в 2 раза и более толщины хребтового и бокового шипика в сравнении с агельминтными животными. У свиней, зараженных аскаридозом и метастронгилезом, из паренхиматозных органов выделены микробные культуры *S. typhimurium* и *E. rhusiopathiae*. В организме диких свиней зафиксированы паразитоценозы, сочленами которых являются разные виды гельминтов и гельминты + высокопатогенные бактерии.

Ключевые слова: дикая свинья, гельминтозы, патогенные бактерии, паразитоценоз

PARASITE CENOSES OF THE WILD PIG (*SUS SCROFA*) ON THE TRANS-BAIKAL TERRITORY

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A comprehensive veterinary study of the wild pig population on the territory of the Trans-Baikal Territory for helminthiasis and microbial transmission was carried out. The helminth fauna of wild pigs was studied and the circulation of bacterial pathogens in the population of these animals was established. The object of the research were materials obtained in the field studies on the Trans-Baikal Territory districts. The study of endoparasite infestation was conducted in 37 wild pigs aged 6 months to 3 years during 2019-2021. Infestation of wild pigs with eight different types of helminths was registered on the Trans-Baikal Territory. The maximum extent of infestation (32.4%) was recorded in *Setaria labiato-papillosa*. Wild pigs are infected with both imaginal helminths *Setaria labiato-papillosa*, *Metastrongylus elongatus*, *Ascaris suum*, *Trichocephalus suis*, *Oesophagostomum dentatum*, as well as with helminth larvae *Cysticercus tenuicollis* and *Cysticercus cellulosae*. The study of the morphology of sexually mature pathogens indicates parasitization of helminths of the genus *Setaria labiato-papillosa* in pigs. Out of 37 wild pigs studied, the association infestation of setariosis + ascaridosis was recorded in 12 animals, three pigs were simultaneously parasitized by ascaridosis + trichocephalus, and two animals had the association of setariosis + metastrongylosis.

Organoleptic examination of carcasses of pigs affected by associative infestation showed a 2-fold or more reduction in the thickness of the backbone and side fat in comparison with helminth-free animals. In pigs infected with ascaridosis and metastrongylosis, microbial cultures of *S. typhimurium* and *E. rhusiopathiae* were isolated from paraenchymatous organs. Parasite ecosystems with different types of helminths and helminths + highly pathogenic bacteria have been recorded in the body of wild pigs.

Keywords: wild pig, helminthiasis, pathogenic bacteria, parasitocenosis

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Conflict of interest

The author declares no conflict of interest.

INTRODUCTION

The wild pig (*Sus scrofa*) is widespread in the Trans-Baikal Territory and is a common species of its fauna. Wild pigs are distinguished from other game animals inhabiting the territory of the region by their pantophagy, high fecundity, and wide ecological plasticity [1-3]. Wild pigs are the main commercial species for many hunting farms in the Trans-Baikal Territory.

Many scientific works in different natural and climatic zones of the Russian Federation and beyond are devoted to the study of the helminth fauna of wild pigs as one of the valuable game animals¹ [1, 4-8]. In Kazakhstan, 15 helminth species have been registered in wild boars: three species of trematodes, two of cestodes, nine of nematodes, and one of scrapie species [9]. T.G. Melnikova established that in Kyrgyzstan, wild boar are hosts of 21 helminth species. V.A. Strelchik et al. observed 11 species of parasitic worms in wild boar in the Primorsky Territory. Studies on helminth infestations of wild boar were also carried out in the middle belt of Russia. V.A. Romashov analyzed the helminth fauna of wild boar in the Voronezh Nature Reserve. In the Usmanskiy pine forest, he found 12 helminth species in wild boar. Two species of trematodes, five

species of nematodes and three species of protozoa were found in wild boars in the territory of the National Park "Losiny Ostrov" with an extensiveness of infestation of 59% [10].

A number of authors [2, 11] point out² that 72 helminth species parasitize in wild pigs and 98 in domestic pigs. A.I. Mozgovykh's monograph (see footnote 1) states that the world fauna includes 139 species of swine helminths. Besides, 78 parasitic worm species were registered in the territory of the former USSR in domestic and wild pigs: 53 species in domestic pigs and 33 in wild boar by 1967.

At the same time, it should be noted that there is almost no information in the scientific literature on the taxonomic association (viruses, bacteria, protozoa, helminthes) of parasites in wild pigs. The departure from the monistic point of view on infectious and invasive diseases and transition to the concept of associative diseases allows to significantly accelerate further development and widespread implementation of various means and methods of combating infectious and invasive diseases (see footnote 1) [12]. All this requires a comprehensive approach and the development of fundamentally new methods to ensure rapid and accurate diagnosis of diseases caused by the participation of different species of organisms at different levels of the hierarchical ladder.

¹Mozgovoy A.I. Helminths of domestic and wild pigs and diseases caused by them. Moscow: Nauka, 1967. pp. 129-164.

²Parasitocenosis and associative diseases: collection of articles. Moscow: Kolos, 1984. 302 p.

The above indicates that the problem of parasitocenosis in the pathology of wild animals is very relevant, since the data obtained as a result of a comprehensive study of associative members can be used for new approaches to the deciphering of pathogenesis, timely diagnosis, specific prevention and treatment of associative diseases of animals.

The purpose of the study is to give a comprehensive helminthological and microbiological assessment of the infestation of wild pig populations in the Trans-Baikal Territory.

The objectives of the study are to study the helminth fauna of wild pigs; to establish the circulation of bacterial pathogens in the population of wild pigs.

MATERIAL AND METHODS

The work was carried out at the Research Institute of Veterinary Sciences of Eastern Siberia, branch of the Siberian Branch of the Russian Academy of Sciences. The work is based on the materials obtained in the field studies in the Trans-Baikal Territory. The study of endoparasite infestation was carried out in 37 wild pigs aged from 6 months to 3 years during 2019-2021.

Helminthological studies were performed using generally accepted parasitological methods (Darling, Fulleborn, PGV by K.I. Skryabin, Berman helmintholarkoskopy method)³.

Microbial cultures were cultured in a thermostat (dry-air electric thermostat cabinet 2C-405M) at 37 °C with daily viewing for the first 5 days. After culture isolation, the morphological properties of the colonies were described: shape, type of edges, colony profile, and size. The mobility of the colonies was examined by the crushed drop method, stained again with Gram stain, and microscopically examined. The isolated cultures were identified by their cultural and biochemical, tinctorial, and serological properties.

Microscopic examination was carried out on smears, imprints of internal organ samples, which were obtained after 3-4 times touch-

ing the surface of an organ slice with a slide. Smears were prepared directly from the native material and stained according to Gram, Romanowsky-Giemsa, Kozlowsky, Peshkov and Trujillo.

Virulence and pathogenicity of cultures were studied by infecting laboratory animals (white mice): 1 cm³ of freshly prepared suspension was injected intraperitoneally.

When isolating a pure culture, the identification of the pathogen by motility, enzymatic properties, catalase test, agglutination reaction with sera, sensitivity to bacteriophages, and seeding on elective media was performed.

Mobility was determined by the hanging and crushed drop method, as well as by inoculation in semi-liquid meat-and-peptone agar. For this purpose, an 18-h broth culture grown at room temperature was used.

To determine the enzymatic properties, the pure broth culture was transferred to a mottled row (11 sugars). The cultures were incubated in an incubator at 37 °C.

The isolated culture was identified on the basis of its cultural, morphological, biochemical, serological, and biological properties.

RESULTS AND DISCUSSION

To determine the circulation of infestation agents, 37 wild pigs aged from 6 months to 3 years old that were hunted in the Trans-Baikal Territory were subjected to complete helminthological autopsy (CHA). Among them 17 animals were infected with different helminths species (AI 46%). Since in most cases only pieces of diaphragm were used for trichinellosis tests, the number of examined carcasses was much greater (272) than in full autopsy. A total of 272 carcasses were examined for trichinellosis, of which 2 (0.7%) were found to be infected with larvae.

As can be seen from the table, *Setaria labiato-papillosa* has the highest distribution of helminths in the wild pig population in the Trans-Baikal Territory, the extensiveness of which is 32.4%. In our opinion, the main

³Kotelnikov G.A. Helminthological studies of animals and the environment. Moscow: Kolos, 1983. 208 p.

Систематический состав гельминтов дикой свиньи на территории Забайкальского края
Systematic composition of wild pig helminths on the territory of the Trans-Baikal Territory

Type of helminth	Location	The number of examined/affected animals, Prevalence, %
<i>Trichinella native</i>	Thoracic diaphragm	272/2 (0,7)
<i>Metastrongylus elongatus</i>	Bronchi	37/4 (11,4)
<i>Setaria labiato-papillosa</i>	Abdomen	37/12 (32,4)
<i>Ascaris suum</i>	Small intestine	37/4 (10,8)
<i>Trichocephalus suis</i>	Large intestine	37/3 (8,1)
<i>Cysticercus tenuicollis</i>	Veil fat, pleura	37/2 (5,4)
<i>Cysticercus cellulosae</i>	Muscles	37/1 (2,7)
<i>Oesophagostomum dentatum</i>	Large intestine	37/2 (5,4)

source of infection of wild pigs with setariosis is agricultural animals, in particular cattle, in the population of which there is a persistent vulnerability to setariosis in the territory of the region. Although in the available literature, we found no information on the disease of wild pigs setariosis, but our research, namely the study of morphology of sexually mature pathogens, indicates parasitization of pig helminths of *Setaria labiato-papillosa* species.

Nematodes *Metastrongylus elongatus* were found in bronchi of more than 10% of wild pigs examined. *Metastrongylus elongatus* was detected in animals under one year of age, which coincides with the data of other researchers on the age dynamics of metastrongylosis.

Ascarids were found in the small intestine in 10.8% of the examined animals. The age of animals affected by ascariasis ranged from 4 to 8 months, no ascarids were found in older pigs. It should be noted that investigations of piglets younger than 4 months of age were not carried out due to lack of material. In our opinion, a certain role in the spread of ascariasis is played by feeding grounds where animals are concentrated. Besides, helminth eggs were repeatedly found in soil from these sites. Given the ability of eggs to remain viable for a long time (years), hunting farms should deworm these areas and change their locations.

Trichocephalus suis was detected in the large intestine of three pigs older than one

year, the intensity of trichocephalus infestation was 8.1%.

Infestation of wild pigs with *Cysticercus tenuicollis* amounted to 5.4%, the same number of animals was infected with intestinal strongylates *Oesophagostomum dentatum* (see table).

The larvae of the swine chain *Cysticercus cellulosae* were found in the heart muscle and skeletal muscle of one pig. It can be concluded that there are people infected with taeniasis.

The infestation of wild pigs with trichinellosis in the region is rather low and does not exceed 0.7%. Out of 272 examined carcasses, trichinella larvae were found in two animals. However, the risk of human infection with trichinellosis remains even in this case, taking into account the fact that several hundred wild pigs are harvested in the region annually.

Out of 37 wild pigs studied, the association infestation of cetariasis + ascariasis was recorded in 12 animals, three pigs were simultaneously parasitized by ascariasis + trichocephalus, two animals had association of cetariasis + metastrongylosis. Organoleptic examination of carcasses of pigs affected by associative infestation showed a 2-fold or more reduction in the thickness of the backbone and side fat compared to those of helminthic animals.

Along with the study of helminth infestation, the microbial infestation of wild pigs was studied. As a result, a number of bacteria were isolated, including pathogenic agents.

A culture of *Erysipelothrix rhusiopathiae* was isolated from the lung tissue and mediastinal lymph nodes of a pig (age 2 years) infected with metastrogilus.

Morphological properties. Gr+ polymorphic, thin, slightly curved rods, immobile. Arranged singly and in short chains.

Cultural properties. On MIB it caused slight turbidity and grayish sediment. On MPA it formed small dewy colonies.

Biochemical properties. Lactose, glucose, maltose, sucrose, rhamnose fermented with formation of acid without gas. The catalase reaction was negative; reaction with methylroth, neutralroth was negative. It formed hydrogen sulfide.

Biological test. The death of white mice was observed on day 3.

A culture of *E. coli* (serotype 08) was isolated from the contents of the gastrointestinal tract of a pig (6 months old) infected with ascarids.

Morphological properties. Microscopy of smears revealed polymorphic Gr+ bacilli, solitary, mobile.

Cultural properties. Large, white, grayish-white colonies with smooth edges on MPA, uniform turbidity on MIB, and crimson-red colonies with metallic luster on Endo medium.

Biochemical properties. Fermented with the formation of acid and gas glucose, lactose, mannitol, sucrose, did not decompose urea. It formed indole, didn't secrete hydrogen sulfide, the test on Voges-Proskauer was negative.

Biological test. It did not cause the death of mice.

Serodiagnosis. Reaction with polyvalent group O coli serum I positive.

A culture of *S. typhimurium* was isolated from parenchymatous (lungs, spleen) organs of a boar (8 months old) infected with ascariasis.

Morphological properties. Gr- rods, with rounded ends, were arranged individually, less often in pairs.

Cultural properties. When sown on MPA, it formed tender, smooth, juicy colonies, in MIB it caused uniform turbidity, on Endo medium - transparent bluish colonies.

Biochemical properties. Fermented glucose, maltose, sucrose, did not ferment lactose and sucrose. Emitted hydrogen sulfide. It didn't form indole, didn't curdle milk, didn't decompose gelatin.

Serodiagnosis. Reaction with salmonellosis serum is positive (see footnote 1) [12].

Biological test. The death of laboratory mice was noted on the 3rd day.

Thus, associations of helminths and bacteria, including pathogenic ones, have been registered in wild pigs inhabiting the territory of the Trans-Baikal Territory. The pathogens of salmonellosis, escherichiosis, swine eruption, and potentially dangerous pathogens have been isolated.

The causative agents of swine eruption were isolated from the pulmonary tissue, mediastinal lymph nodes of pigs infected with metastrongylus, and from the causative agents of metastrongylosis. According to Y. F. Petrov [12], swine eruption pathogens can persist in metastrongylosis for a long time, without changing the morphological structure, cultural properties. However, as the defenses of the animal organism decrease under the action of nematodes, the virulence of *Erysipelothrix rhusiopathiae* and their number can increase sharply to the level of pathogenicity. In pigs infected with ascariasis, a microbial culture of *S. typhimurium* was isolated from the lungs and spleen.

In our opinion, the entry of bacteria into the lungs, lymph nodes and other parenchymatous organs occurs on the surface of helminth larvae bodies, in our case - ascarids or metastrongylus from the intestines. A parasitocenosis is formed in the animal organism, whose co-members are helminths and highly pathogenic bacteria. Their synergistic action leads to focal or diffuse purulent-catarrhal bronchopneumonia with high mortality of animals.

CONCLUSIONS

1. In the Trans-Baikal Territory, infestation of wild pigs with eight species of various helminths has been registered. The maximum extent of infestation (32.4%) was recorded for

Setaria labiato-papillosa. Wild pigs were infected with both imaginal helminths (*Setaria labiato-papillosa*, *Metastrongylus elongatus*, *Ascaris suum*, *Trichocephalus suis*, *Oesophagostomum dentatum*) and helminth larvae of *Cysticercus tenuicollis* and *Cysticercus cellulosae*.

2. The study of the morphology of sexually mature pathogens of infestations indicates parasitization of helminths of *Setaria labiato-papillosa* genus in pigs. In the available literature no information on the disease of wild pigs setariarhiosis was found.

3. Of 37 wild pigs studied, the association infestation of cetariasis + ascariasis was recorded in 12 animals, three pigs were simultaneously parasitized by ascariasis + trichocephalus, two animals had association of cetariasis + meta-strongylosis. Organoleptic examination of carcasses of pigs affected by associative infestation showed a 2-fold or more reduction in the thickness of the backbone and side fat compared to those of helminthic animals.

4. In pigs infected with ascariasis and metastrongylosis, microbial cultures of *S. typhimurium* and *E. rhusiopathiae* were isolated from parenchymatous organs.

In the body of wild pigs, parasitocenoses have been recorded, whose co-members are different types of helminths and helminths + highly pathogenic bacteria.

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ИНФОРМАЦИЯ ОБ АВТОРЕ

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ДИНАМИКА И ОСОБЕННОСТИ ПРОЯВЛЕНИЯ СИБИРСКОЙ ЯЗВЫ НА ТЕРРИТОРИИ РЕСПУБЛИКИ ТЫВА

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Представлены результаты изучения эпизоотического благополучия в условиях резко континентального климата Восточной Сибири. Для проведения исследований использованы материалы статистических отчетностей управлений ветеринарии районов и Службы по ветеринарному надзору Республики Тыва, информация Госкомстата. Отмечено, что в течение изученного периода (1933–2022 гг.) регион официально считается стационарно неблагоприятным, сибирская язва зарегистрирована в 199 неблагоприятных пунктах на территории 13 административных районов и г. Кызыл. Причиной распространения эпизоотий является наличие большого количества почвенных очагов инфекции в регионе. В результате ретроспективного анализа динамики и особенностей проявления сибирской язвы по республике за десятилетние периоды установлено, что эпизоотии максимального уровня напряженности происходили в 1933–1982 гг. В последующие два десятилетия (1983–2002 гг.) зарегистрированы эпизоотии среднего уровня напряженности. Минимальный характер проявления эпизоотической ситуации по сибирской язве отмечен в последние два десятилетия (с 2003 по 2022 г.), что демонстрирует выраженную положительную динамику к тенденции снижения напряженности эпизоотической ситуации. Спорадические вспышки сибирской язвы происходят в настоящее время из-за активации почвенных очагов инфекции, что указывает на стационарное неблагоприятие региона. При районировании территории Республики Тыва по эпизоотической активности сибирской язвы за 1933–2022 гг. к первой группе районов (максимальной эпизоотической активности) относятся Барун-Хемчикский, Дзун-Хемчикский, Улуг-Хемский, Тес-Хемский, Тандинский, Эрзинский, Чаа-Холский, Овурский районы и территория г. Кызыл. Ко второй группе (со средним уровнем эпизоотической активности) – Бай-Тайгинский, Каа-Хемский, Пии-Хемский, Кызылский, Тоджинский районы. К третьей группе (с минимальным риском эпизоотической активности) относятся свободные от сибирской язвы Монгун-Тайгинский, Сут-Холский, Чеди-Холский и Тере-Холский районы.

Ключевые слова: сибирская язва, неблагоприятные пункты, степень благополучия, индекс эпизоотичности, эпизоотия, спорадические случаи

DYNAMICS AND PECULIARITIES OF ANTHRAX OCCURRENCE ON THE TERRITORY OF THE REPUBLIC OF TYVA

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The results of the study of epizootic welfare in the sharply continental climate of Eastern Siberia are presented. Materials of statistical reports of the District Veterinary Departments and the Veterinary Supervision Service of the Republic of Tyva, and the information from the Goskomstat (State Statistics Committee) were used for the research. It was noted that during the period under study (1933-2022) the region was officially considered stationary unfavorable, anthrax was registered in 199 unfavorable points on the territory of 13 administrative districts and Kyzyl. The reason for the spread of epizootics was the presence of a large number of soil foci of infection in the region. As a result of a retrospective analysis of the dynamics and peculiarities of anthrax occurrence in the republic over the ten-year periods, it was established that the epizootics of maximum intensity occurred in 1933-1982. In the next two decades, 1983-2002, epizootics of medium intensity were recorded. Minimum character of anthrax epizootic situation is observed in the last two decades

from 2003 to 2022, which demonstrates a pronounced positive tendency to reduce the tension of the epizootic situation. Sporadic outbreaks of anthrax are currently occurring due to the activation of soil foci of infection, which indicates a stationary problem of the region. When zoning the territory of Tyva Republic according to anthrax epizootic activity for 1933-2022 the first group of districts (maximum epizootic activity) includes Barun-Khemchiksky, Dzun-Khemchiksky, Ulug-Khemsy, Tes-Khemsy, Tandinsky, Erzinsky, Chaa-Kholsky, Ovursky districts and the territory of Kyzyl. The second group (with an average level of epizootic activity) includes Bai-Taiginsky, Kaa-Khemsy, Pii-Khemsy, Kyzylsky and Todzhinsky districts. The third group (with minimal risk of epizootic activity) includes anthrax-free Mongun-Taiginsky, Sut-Holsky, Chedi-Holsky and Tere-Holsky districts.

Keywords: anthrax, permanently disadvantaged points, degree of distress, epizootic index, epizootic, sporadic cases

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Автор заявляет об отсутствии конфликта интересов.

Conflict of interest

The author declares no conflict of interest.

INTRODUCTION

The current state of anthrax in the world remains unstable, anthrax is observed in many countries of Asia, South America and Central Africa [1-3]. In Russia in 2013-2022 this disease was registered in the North Caucasian, Southern, Volga, Ural and Siberian federal districts (SFD). In SFD the anthrax was registered in the southern part. The Siberian region can be conventionally divided into three groups of epizootic unfavorable conditions according to the registered cases of the disease. Altai and Trans-Baikal Territories, the republics of Buryatia and Tyva, Omsk and Novosibirsk Regions, the Republic of Khakassia and Krasnoyarsk Territory, Irkutsk and Tyumen Regions, and the Republic of Altai and Tomsk Region are considered to be relatively free of disease [4, 5].

Anthrax is a dangerous infectious disease common to animals and humans [6]. The causative agent of *Bacillus anthracis* exists in two main forms: bacillary and spore forms. The source of the infectious agent is a sick animal. Bacilli with biomaterial of sick animals in the external environment in the presence of oxygen turn into spores, are preserved in the soil, being soil foci and factors of animal infection, which makes anthrax a soil-focal stationary infection

[7]. The main route of infection of animals is nutritional, through feed and water. Humans become infected through direct or indirect contact with sick animals during maintenance, slaughtering, carcass cutting, and various professional activities [8, 9].

The purpose of the research is to study the dynamics and characteristics of anthrax in the territory of the Tyva Republic for 1933- 2022.

The research objectives are to conduct a retrospective analysis of the anthrax epizootic situation with the identification of the degree of vulnerability and the epizootic index.

MATERIAL AND METHODS

The object of the research is anthrax in the territory of the Tyva Republic. Materials of statistical reports of the district veterinary departments and the Veterinary Surveillance Service of the Tyva Republic, information from the State Statistics Committee were used for the studies. For a comprehensive study of the dynamics and peculiarities of anthrax manifestation, the data on the number of populated and active stationary contaminated sites (SCS), administrative districts where epizootics took place were used. The data on the number of unfavorable years and epizootic recurrences (from 1933 to 1972

in the census of the populated areas 1970¹, from 1973 - in the census of 2010 which remained unchanged since 2001) were analyzed by chronological reflection method, using which the indicators of unfavorability were revealed.

The proportion of active SCSs (UNF - unfavorability)² characterizes the spread of the disease as a percentage, expressed as a ratio of the number of all registered SNPs to the total number of available settlements in the administrative territory, is calculated by the formula

$$\text{UNF} = \frac{\text{The number of active SCSs}}{\text{Number of settlements}} \times 100.$$

Index of epizooticity (IE)³ characterizes the severity of the epizootic situation over time, expressed as a ratio of the number of years during which the disease was observed to the number of years of observation and is calculated by the formula

$$\text{IE} = \frac{N}{T},$$

where N is the number of years during which the disease was registered; T is the number of years of observation.

The work was conducted at the Tuvan Research Institute of Agriculture in 2022.

RESULTS AND DISCUSSION

The climate of Tyva is sharply continental. The main natural and climatic zones are tundra plateaus with permafrost, taiga massifs, connecting with steppe and desert. The soils of Tyva are mountain-taiga-derived permafrost, mountain-forest-taiga, brown mountain-forest, peat-marsh, chernozem-steppe, chernozem forest-steppe, etc. Chernozems are most favorable for preservation of anthrax activity in soil. Anthrax outbreaks are often noted in dark chestnut, chestnut and loam soils. The persistence of anthrax spores in soil depends on its composition and properties; a moist alkaline

and calcium-rich soil environment is the most favorable [10].

According to the administrative-territorial structure, the Republic of Tyva, with Kyzyl as its capital, includes 18 administrative units with 147 settlements. Anthrax in the analyzed 90 years (1933-2022) was registered on the territory of 13 administrative districts and the city of Kyzyl, which makes up 77.8% of the administrative territory of the Tyva Republic. On the scale of the republic, active manifestations of anthrax were recorded in 56 unfavorable years (a total of 126 in districts) in 199 active unfavorable sites. The degree of unfavorability (UNF) was 135.4% (see Table 1), index of epizooticity (IE) 0.622 for the republic, 1.4 for the districts in total (see Table 2). 2), whose indexes divided the ten-year periods into three levels: maximum severity, where there are 8 to 11 unfortunate areas, 20 to 60 unfavorable sites, with epizootic recurrences of 8 to 10 unfavorable years for the region, 18 to 26 for the districts, the UNF degree of 13.6 to 40.8%, the IE of 0.8 to 1.0 for the region, 0.2 to 0.288 for the districts-the first five ten-year periods from 1933 to 1982.

The peculiarity of anthrax epizootics during these periods is that the dynamics of epizootic indicators was uneven. In the same period the number of active SCSs and the degree of the UNF decreased, gaining positive dynamics, and the number of unfavorable years and the IE asymmetrically this had negative dynamics, rising to marginal negative indicators (11 districts, the IE - 1,0 for the region, 0,288 - for districts) or vice versa. This led to their association and characterization of epizootic situation as the maximum degree of tension. The next two periods (from 1983 to 2002) are characterized as a medium degree of epizootic tension with positive dynamics, with the number of: 3 to 6 unfavorable districts, 3 to 4 unfavorable years per region, 6 to 8 per districts, 8 unfavorable sites; with equal degree of the UNF (5.4%), the IE - 0.3 to 0.4 per region, 0.066 to 0.088 per

¹Register of populated areas of the RSFSR, unfavorable for anthrax. Methodical recommendations. M., 1976. Vol. 4. pp. 194-200.

²Dzhupina S.I., Kolosov A.A. Methods of epizootological studies. Methodological recommendations. Russian Academy of Agricultural Sciences. Siberian Branch, Novosibirsk, 1991. 61 p.

³Sidorchuk A.A., Voronin E.S., Glushkov A.A. General epizootology. Moscow: KoloS, 2005. 176 p.

Табл. 1. Степень неблагоприятности (НБ) по сибирской язве на территории районов и всего по Республике Тыва за 1933–2022 гг.
Table 1. The degree of problems with anthrax (DP) on the territory of the districts and the Republic Tyva for 1933-2022

District	Number of settlements	The number of active SCSs and the degree of unfavorability over the ten-year periods														Total unfavorable sites	UNF 1933–2022, % by districts				
		1933–1942	UNF, %	1943–1952	UNF, %	1953–1962	UNF, %	1963–1972	UNF, %	1973–1982	UNF, %	1983–1992	UNF, %	1993–2002	UNF, %			2003–2012	UNF, %	2013–2022	UNF, %
Mongun-Taiginsky	3																			0	
Bay-Taiginsky	8		37,5	3																4	50
Barun-Khemchik	9	15	166,7	6	66,6	1	11,1	1	11,1	5	55,6	1	11,1				3	33,3	32	355,5	
Sut-Holsky	7																			0	0
Dzun-Khemchik	13	11	84,6	11	84,6		38,5	5	38,5	1	7,7								28	215,4	
Chaa-Holsky	4			1	25,0		125,0	5	125,0										11	275	
Ulug-Khemsky	10	13	130,0	7	70,0	2	20,0	2	20,0	2	20,0	1	10,0						27	270	
Tandinsky	13	11	84,6			3	23,0	1	7,7	7	53,8								22	169,2	
Chedi-Holsky	6																		0	0	
Kyzyl	1									1	100,0								2	200	
Kyzylsky	12	1	8,3	1	8,3					1	8,3								4	33,3	
Kaa-Khemsky	17			4	23,5	2	11,8			1	5,9								7	41,2	
Pii-Khemsky	14	1	7,1	5	35,7														7	50	
Tes-Khemsky	7	2	28,6	1	142,8	13	185,7	3	42,8	2	28,6								25	357,1	
Erzinsky	6	4	66,7	1	16,7	5	83,3	5	83,3										18	300,0	
Ovursky	6			10	166,7														10	166,7	
Todzhinsky	7	2	28,6																2	28,6	
Tere-Holsky	4																			0	
Tyva Republic	147	60	40,8	50	34,0	26	17,7	23	15,6	20	13,6	8	5,4	8	5,4	1	0,7	3	2,0	199	135,4

District	The number of anthrax-unfavorable years and Epizootic index (EI) over the ten-year observation periods														EI	The total number of unfavorable years for 1933–2022	EI			
	1933–1942	1943–1952	EI	1953–1962	EI	1963–1972	EI	1973–1982	EI	1983–1992	EI	1993–2002	EI	2003–2012				EI	2013–2022	
Mongun-Taiginsky																				
Bay-Taiginsky		2	0,2																3	0,033
Barun-Khemchik	5	6	0,6	1	0,1	1	0,1	5	0,5	1	0,1								21	0,233
Sut-Holsky																				
Dzun-Khemchik	7	4	0,4			4	0,4	1	0,1										16	0,177
Chaa-Holsky		1	0,1			5	0,5			3	0,3	2	0,2						11	0,122
Ulug-Khemsy	4	3	0,3	1	0,1	2	0,1	1	0,1	1	0,1								12	0,133
Tandinsky	4			3	0,3	1	0,1	6	0,6										14	0,155
Chedi-Holsky																				
Kyzyl								1	0,1	1	0,1								2	0,022
Kyzylsky	1	1	0,1			1	0,1	1	0,1										4	0,044
Kaa-Khemsy		2	0,2	2	0,2			1	0,1										5	0,055
Pii-Khemsy	1	3	0,3							1	0,1								5	0,055
Tes-Khemsy	2	1	0,1	6	0,6	3	0,3	2	0,2			2	0,2	1	0,01				17	0,188
Erzinsky	1	1	0,1	5	0,5	4	0,4					2	0,2						13	0,144
Ovursky		2	0,2																2	0,022
Todzhinsky	1																		1	0,011
Tere-Holsky																				
Total by districts	26	26	0,29	18	0,2	21	0,23	18	0,2	8	0,09	6	0,07	1	0,01	2	0,02	126	1,4	
Total districts by Republic of Tyva	9	9	0,9	8	0,8	10	1	10	1	4	0,4	3	0,3	1	0,1	2	0,2	56	0,622	
Всего районов по Республике Тыва	9	9	0,9	8	0,8	10	1	10	1	4	0,4	3	0,3	1	0,1	2	0,2	56	0,622	

districts. The last two periods (2003-2022) are characterized as below average, when 1 to 2 unfortunate sites with recurrent outbreaks of 1 to 2 unfortunate years, with the UNF degree of 0.7 to 2.0%, the IE of 0.1 to 0.2 per region, 0.011 to 0.022 per region, were registered in one district. There were no favorable periods for anthrax in the territory of the Republic of Tyva for 1933-2022; the situation remains unfavorable for anthrax.

The retrospective analysis of the epizootic situation in the Republic of Tyva during each 10-year period showed that during the first period (from 1933 to 1942) anthrax epizootics were registered in 9 districts (50% of the administrative territory of the region), 9 unfavorable years on a regional scale, 26 unfavorable years in total by districts, with the maximum number of unfavorable sites (60), the UNF - 40.8% and high IE (for the region - 0.9, for districts - 0.288). In 1943-1952 negative dynamics was outlined with the expansion of the administrative territories covered by anthrax to 11 districts, 11% more than in the previous period, accounting for 61.1% of the administrative units of the region. However, the number of unfavorable years covered by the epizootic did not change (9 for the region, 26 for the districts), but the number of unfavorable sites decreased by 16.7% (to 50 units), in connection with this the degree of UNF decreased by 6.8% (to 34.0%), but the IE remained as high (0.9 for the region and 0.288 for the districts). In 1953-1962 positive dynamics towards narrowing the territories of unfavorable districts to 6, which was 1.8 times fewer than in the previous period, covering 33.3% of administrative units of the region, with epizootic recurrence on a regional scale of 10; at the regional level there were 18 epizootics with a reduction of the number of the unfavorable sites 1.9 times to 26, the UNF - 1.8 times (to 17.7%) and inconsiderable decrease of the IE (0.8 for the region, 0.2 for the districts).

In the next 10-year period (1963 - 1972) negative epizootic dynamics was recorded with expansion of territory coverage by 11,1% (up to 8 districts), 44.4% of administrative units of the region with an increase in the number of dis-

eased years to 10 for the region, 21 in total for districts, but with a slight decrease (2,1%) in the number of unfavorable sites to 23 units and a decrease in the UNF by 2,1% (to 15,6%). The increase in the number of unfavorable years in the region affected the tension of the epizootic, respectively, the IE rose to 0.1 in the region, to 0.233 in the districts. Compared to the previous period (1973-1982) the dynamics of the number (8), the percentage of districts (44.4%) covered by the epizootic, and the number of years affected (10) remained unchanged in the region. However, the number of years by districts was insignificantly decreased (to 18 units), unfavorable sites to 20, the degree of the UNF by 2% (to 13.6%) correspondingly insignificantly decreased, so the IE remained at 1.0, and by the districts decreased to 0.2.

In 1983-1992 positive dynamics to the decline of epizootic boundaries by 11,1% (to 33,3%) was observed, administrative units - to 6 districts, the number of unfavorable years decreased almost 2,5 times (to 4 on the region, 8 - to districts), the number of unfavorable sites decreased 2,5 times (to 8), respectively the UNF - to 5,4%, IE - 0,4 for the region, 0,088 - for districts, showing a sharp decrease of epizootic tension on the anthrax. Between 1993 and 2002, the number of districts covered by the anthrax epizootic was halved (to 3 districts), covering 16.6% of administrative units in the region. Epizootic outbreaks were observed 3 years in the region, 6 years in the districts, but the number of active SCSs remained at the same level (8), resulting in a 1.3-fold increase in tension in the 3 districts affected. Overall, the UNF rate remained at 5.4%, while the IE decreased slightly (to 0.3 for the region, 0.066 for the districts).

In 2003-2012 there was a positive trend with a 3-fold decrease in the number of unfavorable areas (up to one Tes-Khemsy district), which equals 5.5% of the administrative units of the region, 1 unfavorable site was registered, 1 unfavorable year, the UNF decreased by 7.7 times, or 0.7%, the IE by region decreased 3 times, to 0.1 in the region, the IE by districts - 6 times, to 0.011. In 2013-2022 anthrax was registered in one Barun-Khemchiksky district, but the num-

ber of active SCSs increased 3 times (to 3), respectively, the degree of the UNF - 2.9 times to 2.0%, the IE - 2 times (to 0.2 and 0.022).

The dynamics of epizootic situation compared to previous periods gradually and steadily acquired a positive trend with some transition to the negative side in some years. However, the tension of epizootic situation decreased with favorable periods of 7-8-9 years. This was influenced by the transfer of cattle breeding to an industrial basis with the construction of dairy-commodity farms and industrial complexes. Enforced immunization of animals against anthrax by Cenkovsky's vaccine was carried out in Tyva until 1955. Since 1955, annual scheduled preventive immunization with STI and GNKI (All-Russian State Research Institute of Control, Standardization, and Certification of Veterinary Preparations) vaccine, and since 1986, with more effective and harmless vaccines, such as live dry vaccine from the 55-VNIIViM strain, etc. [11]. There was a positive impact on the improvement of the situation, the ban on burial of the corpses of fallen animals introduced in 1953, construction of burial grounds and their arrangement with a sanitary protection zone.

However, despite the positive dynamics of the epizootic situation, sporadic outbreaks of anthrax are still observed. This confirms the high degree of stationary ill-health and the spread of soil anthrax foci in the burial sites of anthrax-dead animals, cattle graves without sanitary and protective solid fences, identification signs, which retain epizootic potential for tens, and even hundreds of years [12]. Soil types are of great importance, where the spores of the infectious agent can not only persist but also accumulate, increasing the epizootic danger of the soil focus, which, in turn, affects the confinement of anthrax to a particular territory. The probability of animals becoming infected increases with free and uncontrolled grazing of animals in private farms and subsistence farms with large herds under traditional nomadic animal husbandry in Tyva, most of the time being on grazing and watering from reservoirs. It is known that the peculiarities of epizootic activity of anthrax infection in the stationary

contaminated sites are to a greater extent associated with the influence of different natural and geographical environmental conditions, in particular climatic, soil and landscape and others, typical for the Tyva Republic. Morbidity in animals is noted from early spring to late autumn in lowlands and swampy areas, during snow melting, mudflows, river floods, abundant rainfall, alternating floods and drought, when pathogen spores are washed from the soil, as well as low poor coarse dry grass, while eating which animals injure the mucous membrane of the mouth.

Epizootic zoning for anthrax showed that there are three groups of districts in the territory of the Republic of Tyva according to the degree of risk of epizootic activity: the first group of districts with the maximum risk level - from 11 to 32 registered unfavorable sites, the degree of UNF - from 169.2 to 357.1%, unfavorable years - from 11 to 21, the IE - from 0.122 to 0.233 - includes 9 administrative units: Barun-Khemchik, Dzun-Khemchik, Ulug-Khemsy, Tes-Khemsy, Tandinsky, Erzinsky, Chaa-Holsky, Ovursky districts and the city of Kyzyl.

The second group with an average risk of epizootic activity - from 1 to 10 unfavorable sites, the degree of UNF - from 28.6 to 50.0, the IE - from 0.022 to 0.055 - includes 5 districts: Kaa-Khemsy, Pii-Khemsy, Bay-Taiginsky, Kyzylsky, Todzhinsky districts.

The third group with minimal risk of epizootic activity includes 4 districts free of anthrax: highland Mongun-Taiginsky, Tere-Holsky, Sut-Holsky and Chedi-Holsky districts. However, epizootological zoning shows that the districts with a real risk of anthrax activation of the epizootic situation with stationary soil foci of infection on the territory of the Tyva Republic prevail and make up 77.8% of the available 18 administrative districts, which can affect the situation in districts with a minimum risk of anthrax manifestation. This situation is conditionally safe, since during the reorganization of the administrative structure of the region at different times the districts were either merged into one district or separated into several districts. The settlements were transferred to one and another district, so it is inexpedient to categori-

cally assess these 4 districts as safe without the risk of anthrax outbreak episodes.

CONCLUSION

The territory of the Tyva Republic is stationary anthrax-unfavorable to a high degree. The risk of anthrax outbreaks remains high, especially in the unfavorable areas with abundant stationary soil foci of infection, unrecorded, abandoned, without established sanitary protection zones of cattle burial grounds [13]. The risk increases with the traditional transhumance grazing, and free-range farming of large herds of cattle with uncontrolled grazing on grazing forage.

Epizootological zoning of the territory of the region allows differentiated implementation of managerial measures to supervise livestock burial grounds, burial places of animals dead of anthrax, input of lands for livestock stables, premises, residential buildings, to perform veterinary and sanitary control of cattle slaughtering, sale of animal products, grazing and cattle driving to seasonal stables and pastures. It is necessary to pay special attention to the main tool of anthrax prevention and control - annual qualitative total immunization of animals against anthrax [14].

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РАСПРОСТРАНЕНИЕ ГЕЛЬМИНТОВ ЖЕЛУДОЧНО-КИШЕЧНОГО ТРАКТА ЛОШАДЕЙ В ЦЕНТРАЛЬНОМ АЛТАЕ

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Представлены результаты исследований (2021, 2022 гг.) распространения в коневодческих хозяйствах возбудителей гельминтозов. В формировании гельминтокомплекса животных Центрального Алтая участвуют паразитические черви двух классов – Nematoda и Cestoda. Зарегистрированы нематоды двух подотрядов Strongylata и Ascaridata (*Parascaris equorum*). У цестод, представленных ленточными червями подотряда Anoplocephalata, выделено два вида: *Anoplocephala perfoliata*, *Paranoplocephala mamillana*. Инвазированность однокопытных стронгилятами желудочно-кишечного тракта как в провинции (87,8%), так и в отдельных административных районах существенно превышает зараженность животных гельминтами подотряда Ascaridata (14,1%) и цестодами подотряда Anoplocephalata (10,9%). В формировании нозологического профиля кишечных гельминтозов основную роль играют нематоды подотряда Strongylata. Стронгиляты составляют ядро гельминтокомплекса пищеварительной системы, а инвазированность однокопытных ими и их доля в структуре гельминтокомплекса как в провинции, так и в отдельных административных районах наиболее высока. Показатели ЭИ (экстенсивность инвазии) и значения ИП (индекс паразитокомплекса) стронгилят варьируют в разрезе административных районов соответственно от 53,3 и 69,1% до 95,2 и 80,8% и в среднем по Центральному Алтаю составляют 87,8 и 77,8%. Установлено, что в большинстве районов зараженность однокопытных гельминтами пищеварительной системы в целом и нематодами подотряда Strongylata отличается незначительно. Инвазированность однокопытных *Parascaris equorum* и цестодами подотряда Anoplocephalata в регионе зарегистрирована соответственно на уровне от 14,1 и 10,9% с ИП 12,5 и 9,7, что в 6–8 раз меньше, чем аналогичные показатели при стронгилятозах животных. Установлена достоверная прямая зависимость выделения яиц стронгилят во внешнюю среду от показателей зараженности животных. С повышением значений зараженности животных стронгилятами увеличивается количество пропативных форм, выделяющихся в окружающую среду.

Ключевые слова: гельминты пищеварительной системы, лошади, структура гельминтокомплекса, динамика выделения яиц, региональные особенности эпизоотического процесса

DISTRIBUTION OF HELMINTHS OF THE GASTROINTESTINAL TRACT OF HORSES IN CENTRAL ALTAI

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The results of the studies (2021, 2022) on the spread of helminth infectious agents in horse breeding farms are presented. Parasitic worms of two classes Nematoda and Cestoda participate in the formation of the helminthic complex of Central Altai animals. Nematodes of two suborders Strongylata and Ascaridata (*Parascaris equorum*) were recorded. Two species *Anoplocephalaperfoliata* and *Paranoplocephalamamillana* were identified in cestodes represented by the tapeworms of suborder Anoplocephalata. In the province (87.8%) and in some administrative districts, infestation of whole-hoofed animals with gastrointestinal strongylates is much higher than infection with helmin-

thes of suborder Ascaridata (14.1%) and cestodes of suborder Anoplocephalata (10.9%). Nematodes of suborder Strongylata play the main role in forming the nosological profile of intestinal helminth infections. Strongylates form the core of the helminthocomplex of the digestive system, and the infestation of whole-hoofed animals with them and their share in the structure of the helminthocomplex both in the province and in individual administrative districts is the highest. The values of IP (invasion prevalence) and PCI (parasite complex index) of strongyloides vary by administrative regions from 53,3 and 69,1% to 95,2 and 80,8%, respectively, and amount to 87,8 and 77,8% on average in the Central Altai. It was found that in most areas, the infestation of whole-hoofed animals with helminths of the digestive system in general and nematodes of suborder Strongylata differs insignificantly. Infestation of whole-hoofed animals by *Parascaris equorum* and cestodes of suborder Anoplocephalata is registered in the region at 14,1 and 10,9% with PCI 12,5 and 9,7 respectively, which is 6-8 times lower than analogous indices for strongylatosis of animals. Reliable direct correlation between the release of strongylate eggs into the external environment and the index of animal infestation was established. The number of propagative forms released into the environment increases with increasing values of animal infestation with strongylates.

Keywords: helminths of the digestive system, horses, structure of the helminthocomplex, dynamics of egg release, regional features of the epizootic process

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Conflict of interest

The authors declare no conflict of interest.

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INTRODUCTION

Horse breeding, being an important part of cattle breeding in the Altai Republic, plays a significant role in meeting the region's needs for specific types of raw materials and foodstuffs. Currently, there is a tendency to increase the number of horses in the farm segment [1]. The restraining factor in increasing the number of animals and their productivity is diseases, including those of parasitic etiology.

According to the results of studies of domestic and foreign scientists, helminth infections of the gastrointestinal tract of horses are widespread and in most cases are characterized

by a chronic course without significantly pronounced clinical manifestations [2-8]. However, helminth infestants affecting all parts of the digestive system, with a high degree of infestation are the cause of mass diseases not only in young animals, but also in adult animals. These diseases are manifested by colic, diarrhea, nervous manifestations, decreased performance and all kinds of productivity [9-11].

The results of studies demonstrate that the species composition of the parasitic worm community depends on the conditions of horse keeping and the use of anthelmintics from the group of macrocyclic lactones and benzimid-

azoles in horse breeding, on the natural and climatic characteristics of the area and other biotic and abiotic factors [6, 7, 12].

The information on the species diversity of multicellular parasites, their spatial distribution in the context of the administrative division and with regard to natural and climatic zones, the peculiarities of the epizootic process in the Siberian region is limited and requires clarification¹⁻⁴ [13-17], given the considerable study of helminths of the European part of the Russian Federation.

The purpose of the research is to characterize the structure of the helminth complex and the epizootic situation of gastrointestinal helminthiasis of horses in the Central Altai.

MATERIAL AND METHODS

The researches were carried out in 2021, 2022 in 13 farms of 5 administrative regions of Central Altai (Shebalinsky, Chemalsky, Ongudaysky, Ust-Kansky, Ust-Koksinsky). To study the infestation of animals with helminths, fecal samples obtained from spontaneously infested horses were examined by the helminth coproscopic flotation method according to Füllerborn with subsequent counting of eggs per gram of feces [18].

Taking into account that the dam of anoplocephalids is of a closed type, and cestodes excrete segments into the external environment, their dispersal forms in fecal samples were not counted. Taxonomic differentiation of nematodes was performed taking into account the morphometric features of the eggs; cestodes were identified by the size and shape of the pear-shaped apparatus [19]. A total of 719 samples of biomaterial were examined.

According to the results of the oviscopy the indices of the intensity of infestation were calculated: IE (infestation extensity) is the propor-

tion of infected animals in percentage, IEam is the arithmetic mean of the IE indices in samples (surveys) in percentage, IEgm is the geometric mean of the IE indices in samples. The indicators of the infestation intensity: II is the arithmetic mean of the number of eggs per one infected animal in a gram of feces in specimens, IIam is the arithmetic mean of the II indices in samples in percentage, IIgm is the geometric mean of the II indices in samples. Significance of differences in infestation indices was established according to the geometric mean of IEgm and IIgm with calculation of Student's t test ($p \leq 0.05$, $df = n_1 + n_2 - 2$).

Since helminth infections have an associative form of progression, it is necessary to consider peculiarities of the whole parasitic complex formed in a particular territory, taking into account its ecological characteristics, when forming the systems of treatment and prophylactic measures. In this regard, the determination of peculiarities of the helminth complex structure of the gastrointestinal tract of horses and the significance of individual taxons in it is an important component in the development of programs to control the number of animal parasites. For this purpose, for a formalized description at the cenotic level of the parasite community, an indicator such as the parasite complex index (PCI), which reflects the value of a species, genus, or other taxon in its structure, was additionally used [20].

RESULTS AND DISCUSSION

In helminth oviscopy examinations of horses, nematode eggs of two suborders Strongylata and Ascaridata (*Parascaris equorum*) were detected in fecal samples. Cestodes represented by tapeworms of suborder Anoplocephalata include two helminth species - *Anoplocephala perfoliata* and *Paranoplocephala mamillana*.

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The widespread distribution of helminth infestants in Central Altai horse breeding farms has been revealed. In the province as a whole (87.8%) and in some administrative districts the invasiveness of whole-hoofed animals is much higher than infestation with helminths of the suborder Ascaridata (14.1%) and with cestodes of the suborder Anoplocephalata (10.9%) (see Table 1). Minimum and maximum values of horse infestation by strongylates were registered in Ust-Kansky and Chemalsky regions. They are 53.3 and 95.2%, respectively, with II

at 174.0 and 467.5 eggs/g (eggs per gram of feces) (see Tables 1, 2).

According to the data of ovoscopy, the rates of infestation of horses with digestive system helminths by districts are differentiated into two groups: Ongudaysky and Ust-Kansky regions (IE = 79,3 and 56,3%), other regions (IE = 93,3 -95,8%) (see table 1). Indicators of intensity of infection are similarly distributed: Ongudaysky and Ust-Kansky districts (II = 326.9 and 186.7 e/g), other districts (II = 455.8-464.7 e/g) (see Table 2).

Табл. 1. Инвазированность (ЭИ) лошадей Центрального Алтая гельминтами желудочно-кишечного тракта (овоскопия)

Table 1. Infestation (IP) of Central Altai horses by gastrointestinal helminths (ovoscopy)

Administrative District	Number of samples	Number of tests	IP, %			IP total, %	$\frac{IP_{am}}{IP_{ga}}$
			ST	PAR	ANOPL		
Shebalinsky	20	417	93,0 ± 1,2	13,4 ± 0,4	9,8 ± 1,4	93,3 ± 1,2	$\frac{95,3 \pm 1,9}{1,9 \pm 0,01}$
Chemalsky	4	48	95,2 ± 8,1	0	23,8 ± 9,3	95,8 ± 8,3	$\frac{96,3 \pm 2,1}{1,9 \pm 0,01}$
Ongudaysky	4	72	79,4 ± 6,9	14,7 ± 6,1	8,8 ± 4,8	79,2 ± 4,8	$\frac{79,1 \pm 3,9}{1,9 \pm 0,02}$
Ust-Kansky	6	78	53,3 ± 5,7	10,2 ± 3,4	12,8 ± 3,8	56,3 ± 5,6	$\frac{64,6 \pm 9,9}{1,8 \pm 0,01}$
Ust-Koksinsky	4	104	93,5 ± 2,3	22,1 ± 4,1	11,5 ± 3,1	94,2 ± 2,3	$\frac{93,3 \pm 2,5}{1,9 \pm 0,01}$
Total	38	719	87,8 ± 1,2	14,1 ± 1,3	10,9 ± 1,2	89,2 ± 1,2	—

Note. ST – helminths of the suborder Strongylata; PAR – nematodes *Parascaris equorum*; ANOPL – cestodes of the suborder Anoplocephalata.

Табл. 2. Интенсивность заражения (ИИ) лошадей Центрального Алтая нематодами желудочно-кишечного тракта (овоскопия)

Table 2. Intensity of infestation (II) of Central Altai horses with gastrointestinal nematodes (ovoscopy)

Administrative District	Number of samples	Number of tests	II, egg/g		II total, egg/g	$\frac{II_{am}}{II_{ga}}$
			ST	PAR		
Shebalinsky	20	417	430,6 ± 36,0	5,9 ± 1,5	464,7 ± 37,8	$\frac{436,2 \pm 36,1}{2,35 \pm 0,06}$
Chemalsky	4	48	467,5 ± 37,1	0	458,3 ± 37,8	$\frac{467,5 \pm 37,1}{2,59 \pm 0,05}$
Ongudaysky	4	72	402,8 ± 71,1	30,7 ± 17,7	326,9 ± 42,3	$\frac{433,6 \pm 67,9}{2,04 \pm 0,19}$
Ust-Kansky	6	78	174,0 ± 49,1	12,7 ± 5,5	186,7 ± 51,5	$\frac{186,7 \pm 51,6}{1,64 \pm 0,26}$
Ust-Koksinsky	4	104	432,9 ± 102,1	0	455,8 ± 41,3	$\frac{432,9 \pm 102,1}{2,51 \pm 0,03}$
Total	38	719	405,9 ± 28,1	7,5 ± 1,6	431,5 ± 31,2	$\frac{413,3 \pm 28,2}{—}$

Note: ST– helminths of the suborder Strongylata; PAR – nematodes *Parascaris equorum*.

In most cases, differences in infestation rates between groups of districts are statistically reliable and are caused mainly by climatic conditions of the territories of Ongudaysky and Ust-Kansky districts, unfavorable for the development of propagative forms of parasites in the environment (low temperatures, levels of precipitation, etc.) (see Table 3).

Infestation of animals by *Parascaris equorum* of the suborder Ascaridata varies respectively from 10.2 (CI=12.7 e/g) in Ust-Kansky District to 22.1% in Ust-Koksinsky District and averages 14.1%. In the fecal samples of Chemalsky District horses the representatives of ascaridata were not isolated, which is associated with the insufficient volume of material taken for the study.

Cestodes of the suborder Anoplocephalata are represented by two species - *Paranoplocephala-mamilana* and *Anoplocephalaperfoliata*. It should be noted that *A. perfoliata* is registered in all administrative areas and is the dominant species among tapeworms. This is consistent with the results of the studies of foreign and domestic researchers indicating its ubiquitous distribution and prevalence among other cestode species [11, 21].

Paranoplocephala mamilana is rare and isolated in horses of Shebalinsky district (Diktiek village). Infestation of horses by anoplocephalids was registered as minimal in Shebalinsky (9,8%) and Ongudaysky (8,8%) districts,

the greatest one was registered in Chemalsky (23,8%) with the average value for the Central Altai of 10,9%.

There is a significant direct correlation between the number of strongylate eggs excreted with feces in the external environment, and the rate of extensiveness of infection ($r = 0,97$).

The number of propagative forms released into the environment increases with increasing values of Strongylates infestation in animals (see Tables 1, 2, and the Fig.).

In the Altai Mountains, helminth infections occur in the form of mixed infestations with diverse variations in both helminth complex components and their quantitative characteristics (see Table 4).

It was found that in Central Altai, the parasites of two classes Nematoda and Cestoda form the helminth complex of the gastrointestinal tract of ungulates, with a clear dominance of nematodes (PCI = 90,3, II = 431,5 y/y). The ratio of nematode and cestode representatives in helminthological complex was expressed as 9,3: 1,0. In administrative areas this tendency is preserved. The group of helminths of the Strongylata suborder is the most numerous in the Nematoda class. The PCI of Strongylata ranges from 69.1 to 80.8 and averages 77.8 (II = 405.9 y/y). *Parascaris* are the subdominant element of the helminth complex in both the province and the districts. The PCI of *P. equorum* varies from 11.5 to 17.3, which is 4-7 times lower than that

Табл. 3. Критерии достоверности различий показателей зараженности лошадей гельминтами по среднегеометрическим значениям овоскопии ($t/p < 0.05$)

Table 3. Criteria of reliability of differences in indicators of helminth infestation in horses by geometric mean values of ovoscopy ($t/p < 0.05$)

Administrative District	Shebalinsky <i>n</i> = 20	Chemalsky <i>n</i> = 4	Ongudaysky <i>n</i> = 4	Ust-Kansky <i>n</i> = 6	Ust-Koksinsky <i>n</i> = 4
Shebalinsky	0	0	4,5* < 0,01	2,18**	2,14**
Chemalsky	3,07*	0	4,09*	2,1	1,43
Ongudaysky	1,51	4,09*	0	1,27	3,18**
Ust-Kansky	2,73**	2,39*	1,25	0	1,91
Ust-Koksinsky	2,39**	1,38	2,47**	3,34*	0

Note. Data above zero series – geometric mean values of the IP, below zero series - geometric mean values of the II.

* $p < 0,01$;

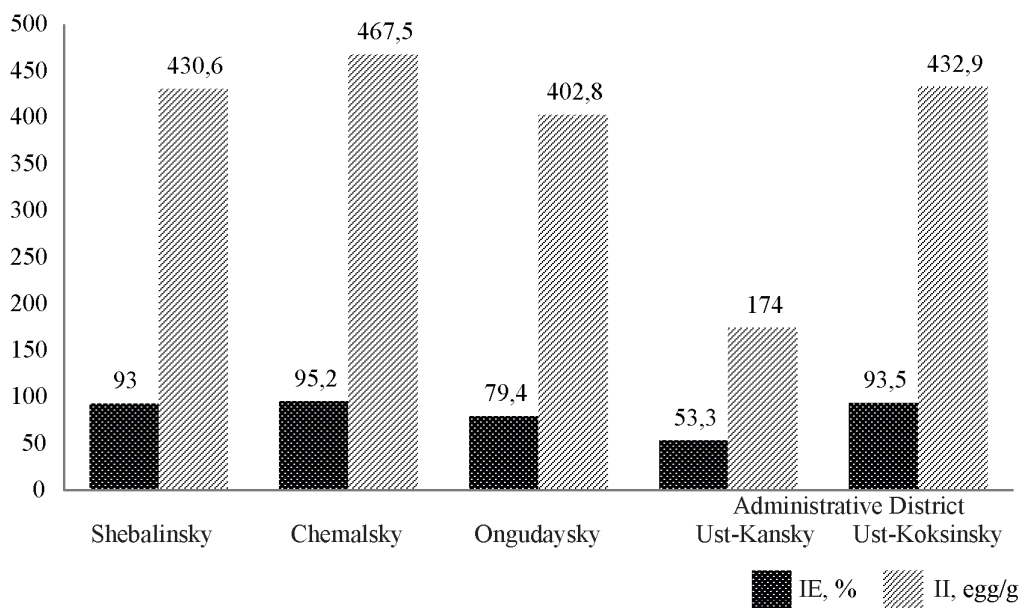
** $p < 0,05$;

n – number pf samples.

of suborder Strongylata nematodes and averages 12.5.

Variation of the PCI values of flatworms of the suborder Anoplocephalata (Cestoda, Cyclophyllidea) within regions is more pronounced (from 8.6 to 19.2 with average value 9.7), which is probably caused by intrazonal diversity of natural and climatic conditions and orographic features of mountain territories determining population density of intermediate host oribatid mites.

In general, the ratio of Strongylidae, Parascaris and Anoplocephalata suborder cestodes in the helminthic complex of the digestive system of ungulates in Central Altai is expressed as 8 : 6 : 1. The ubiquitous distribution of nematodes of Strongylata suborder, their maximal indexes of IE, II and PCI, both in the province and in administrative regions are conditioned by the expressed species diversity, their development in the environment without an intermediate host and the high adaptability of propagative forms to unfavourable environmental factors.



Связь показателей экстенсивности заражения лошадей кишечными стронгилятами с интенсивностью выделения яиц нематод во внешнюю среду

Correlation of extensibility indicators of equine infection with intestinal strongylates and intensity of nematode eggs release into the external environment

Табл. 4. Структура гельминтокомплекса желудочно-кишечного тракта лошадей Центрального Алтая (по результатам овоскопии)

Table 4. Structure of the helminth complex of the gastrointestinal tract of horses in the Central Altai (based on the results of ovoscopy)

Administrative District	IP		
	ST	PAR	Anoplocephalata
Shebalinsky	80,1	11,5	8,4
Chemalsky	80,8	0	19,2
Ongudaysky	77,1	14,3	8,6
Ust-Kansky	69,1	13,7	17,2
Ust-Koksinsky	73,7	17,3	9
Central Altai	77,8	12,5	9,7

Note. ST – helminths of the suborder Strongylata; PAR – nematodes *Parascaris equorum*; ANOPL – cestodes of the suborder Anoplocephalata.

CONCLUSION

The results of the studies testify to the ubiquitous distribution of helminthiasis of the gastrointestinal tract of horses in Central Altai. Infestation of unimpaired horses by helminths of different taxonomic groups forming the helminth complex of the gastrointestinal tract, including strongylates, parascaris and anoplocephalids, both in the province and within administrative districts, differs significantly. The analysis of the infestation indices of IE and PCI indicates that everywhere the nematodes of Strongylata suborder were dominant with PCI of 77.8 (IE = 87.8%, II = 405.9 e/g), parascaris being the subdominants (PCI = 12.5; IE and II respectively 14.1% and 7.5 e/g). The infestation of horses with cestodes is 10.9% with PCI of 9.7. There was a direct reliable dependence of the number of propagative forms of strongylates isolated in the external environment on the level of infection of animals.

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ВЛИЯНИЕ ФИТОБИОТИЧЕСКИХ ПРЕПАРАТОВ НА МОРФОХИМИЧЕСКИЕ ПОКАЗАТЕЛИ КРОВИ ТЕЛЯТ ПРИ ДИСПЕПСИИ

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Представлены результаты влияния комплексных фитопрепаратов на показатели крови при лечении диспепсии молодняка крупного рогатого скота. Материалом исследований служили три группы новорожденных телят герефордской породы с признаками диспепсии. Первой опытной группе задавали препарат № 1 (плоды черемухи, корневище элеутерококка, цветы ромашки, пробиотик) 2 раза в сутки с интервалом 12 ч по 2 мл/кг живой массы; 2-й – выпаивали разработанный препарат № 2 (корневище бадана, лист фенхеля, цветы календулы, пробиотик) 2 раза в сутки с интервалом 12 ч по 2 мл/кг живой массы. Животным контрольной группы не задавали изучаемые препараты, лечили по схеме, принятой в хозяйстве (выпаивали кипяченую воду с NaCl 9 г/л кипяченой воды, панкреатин, тетрациклин). До начала эксперимента у заболевших телят отмечали все клинические признаки диспепсии. После применения фитопрепаратов в опытных группах через 7 дней наблюдали нормализацию лейкоцитов на 60,4 и 48,7% соответственно. Количество эритроцитов во всех исследуемых группах находилось в пределах нормы, уровень гемоглобина в контрольной группе зарегистрирован ниже на 10% в сравнении с нормой и на 20,5% по сравнению с показателем 2-й группы животных. Гематокрит в опытных группах соответствовал норме (35,2 и 38,7% соответственно), в контрольной – превышал норму на 26% ($p < 0,01$). Содержание натрия и калия в сыворотке крови в контрольной группе было снижено на 44 и 17,1% соответственно, в опытных группах находилось в пределах референсных значений. В опытных группах отмечена положительная динамика нормализации кальциево-фосфорного соотношения и железа. Уровень глюкозы достоверно ($p < 0,05$) в 1-й опытной группе увеличился на 2,7%, во 2-й – на 3,9%. Полученные данные свидетельствуют о высокой эффективности препаратов (90 и 100% соответственно) и положительном влиянии на физиологические показатели телят, а также на потребительские качества продукции.

Ключевые слова: телята, диспепсия, лейкоциты, эритроциты, гематокрит, биохимия сыворотки крови

THE EFFECT OF PHYTOBIOTIC PREPARATIONS ON MORPHOCHEMICAL BLOOD PARAMETERS OF CALVES WITH DYSPEPSIA

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The results of the effect of complex phytopreparations on blood parameters in the treatment of dyspepsia in young cattle are presented. Three groups of newborn Hereford calves with signs of dyspepsia were used as the research material. The first experimental group was given the developed preparation № 1 (bird cherry fruit, eleutherococcus rhizome, camomile flowers, probiotic) 2 times a day at 12-hour intervals at the rate of 2 ml/kg of live weight; the 2nd - was given the developed preparation № 2 (bergenia rhizome, fennel leaf, calendula flowers, probiotic) 2 times a day at 12-hour intervals at the rate of 2 ml/kg of live weight. Animals of the control group were not given the studied preparations and were treated according to the scheme adopted at the farm (they were given boiled water with NaCl 9 g/boiled water, pancreatin, tetracycline). Prior to the experiment, all clinical signs of dyspepsia were noted in sick calves. After the application of herbal preparations in the experimental groups in 7 days normalization of leukocytes by 60.4 and 48.7%, respectively,

was observed. The number of erythrocytes in all studied groups was within the normal range, the level of hemoglobin in the control group was registered 10% lower than normal and 20.5% lower than in the second group of animals. Hematocrit in the experimental groups corresponded to the norm (35.2 and 38.7%, respectively), in the control group it exceeded the norm by 26% ($p < 0.01$). The content of sodium and potassium in the blood serum in the control group was reduced by 44 and 17.1%, respectively, and in the experimental groups it was within the reference values. In the experimental groups, positive dynamics of normalization of calcium-phosphorus ratio and iron was noted. Glucose level significantly ($p < 0.05$) increased by 2.7% in the 1st experimental group and by 3.9% in the 2nd group. The data obtained indicate high effectiveness of the preparations (90 and 100% respectively) and a positive effect on the physiological parameters of the calves, as well as on the consumer qualities of the products.

Keywords: calves, dyspepsia, leukocytes, erythrocytes, hematocrit, serum biochemistry

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Conflict of interest

The authors declare no conflict of interest.

INTRODUCTION

Diseases of the digestive organs account for 70% of the total number of diseases in young cattle, of which about 40% end in death, causing economic losses to the livestock industry. Dyspepsia (enzymatic diarrhea) disease of newborn cattle is registered in all periods of the year both on farms with high and low livestock breeding levels. This pathology is registered mainly in the period of mass calving of the dairy period calves (the first 2-5 days of life). The disease is accompanied by acute digestive disorders, indigestion, diarrhea, metabolic disorders and dehydration [1-4].

Various factors can be the causes of dysbacteriosis in the first days of life of young animals, so an important aspect in determining the etiology of dyspepsia is the simultaneous study of gastrointestinal tract microflora composition and blood parameters in newborn animals¹ [2, 3].

Chemotherapeutic drugs used in livestock farms are not always effective and may have adverse effects on calves. Drugs of plant origin

have low toxicity, high bioavailability, a wide range of regulatory effects and polyvalence of therapeutic action. They are increasingly used in medical and veterinary practice and are an important link in the prevention and treatment of diseases of young animals [2, 4].

The purpose of the study was to evaluate the effect of phytobiotic preparations on blood parameters of calves with dyspepsia.

MATERIAL AND METHODS

To achieve this goal, experimental studies were carried out in the training and experimental farm of the Trans-Baikal Agrarian Institute. Laboratory research was carried out in the laboratory of laboratory and analytical studies of the East-Siberian Research and Development Institute of Veterinary Science - branch of SF-SCA RAS. Before setting up the experiment a complex of well-known diagnostic methods was used: analysis of clinical and epizootological data of the farms; conditions of intake, feeding and housing of newborn animals; examina-

¹Abramov S.S., Belko A.A. Dietary preparations for the prevention of dyspepsia of newborn calves. All-Union Scientific Conference, dedicated to the 140th anniversary of the Kharkov Zoovet Institute. Kharkov, 1991. pp. 170-171.

tion of the clinical and physiological status of cows and calves, their feeding and housing.

The preparations developed by us are phytocomplexes (with the addition of probiotic) in the form of solutions obtained by extraction, having a synergistic, antibacterial, antitoxic, anti-inflammatory, astringent effect [5, 6]. Sample preparation № 1 includes bird cherry fruit, rhizome of eleutherococcus, chamomile flowers, probiotic (*Bacillus amyloliquefaciens* RNCIM B-10642 strain (Russian National Collection of Industrial Microorganisms) not less than 1×10^6 CFU of live microbial cells). Sample preparation № 2 contains *Bergenia* rhizome, fennel leaf, calendula flowers, probiotic (*Bacillus amyloliquefaciens* strain RNCIM B-10642 at least 1×10^6 CFU live microbial cells).

Three groups of newborn Hereford breed calves with dyspepsia signs, 10 animals in each group at the age of 2-5 days, were formed to approve complex preparations according to the principle of analogues.

Experimental group (1-st) - animals were given preparation № 1 (fruit of bird cherry, rhizome of eleutherococcus, chamomile flowers, probiotic) 2 times a day at 12-hour intervals for 2 ml/kg of the body weight.

Experimental group (2nd) - animals were fed the developed preparation № 2 (*Bergenia* rhizome, fennel leaf, calendula flowers, probiotic) 2 times a day with an interval of 12 hours at 2 ml/kg of live weight.

Control group - animals were not given any drugs, they were treated according to the scheme adopted by the farm (they were given boiled water with NaCl 9 g/l of boiled water, pancreatin, tetracycline).

In animals of all groups the clinical status of animals (temperature, pulse, breathing, examination of visible mucous membranes, level of dehydration by skin turgor, fecal state, general condition of the animal) was evaluated daily until the period of animal recovery. Feeding of calves was carried out as usual. To evaluate blood parameters when using the drug in calves on day 7 of the experiment, blood samples

were taken in Vacutainer vacuum tubes with K2EDTA for hematology and with clot activator for biochemistry. A PCE 90 Vet hematology analyzer with a set of special reagents was used for the hematological analysis². The blood serum was examined with the help of URIT 800 Vet biochemical analyzer with the use of Dia-VetTest consumables. The efficacy of the drug was evaluated according to the general state of the animals, taking into account clinical, hematological and biochemical parameters.

Biometric processing of the obtained results was performed by the method of variation statistics with the Student's test of significance on a personal computer using Microsoft Office Excel XP programs.

RESULTS AND DISCUSSION

Analysis of clinical and laboratory data showed that the incidence of dyspepsia in newborn calves on the farm varies from single cases to 15-30%, mortality reaches 10-45% of the number of diseased, in addition, the over diseased animals are characterized by a decrease in meat productivity within 10-27% [2, 3]. All experimental calves at the beginning of the experiment had signs of acute gastrointestinal tract disorder in a mild form, recovery in the 1st and 2nd groups occurred on the 4th and 3rd day, respectively. The animals in the 1st and 2nd experimental groups at the end of the experiment were more mobile, reflexes were well expressed, there were no signs of diarrhea, pulse rate and respiration rate were within the physiological norm (see Table 1).

Animals of the control group suffered from the disease in a severe form (20% of calves died), fell behind in weight compared to the experimental groups by an average of 17.5%. Signs of dyspepsia ceased on the 7th day. The main results of the experiment are shown in Tables 1, 2.

Clinical analysis of dyspeptic newborn calves showed an increase in the body temperature of $41,5 \pm 0,13$ °C ($p < 0,05$), decreased appetite, depression, decreased skin turgor, dry

²"A set of diagnostic reagents for hematological analyzers according to TS 9398-001- 85747522-2009" produced by "Clinical Diagnostic Solutions" LLC (Russia).

hair, anemic conjunctivitis, diarrhea. After application of phytobiotic preparations clinical parameters in the experimental groups were normalized, the feces on the 3-4-th day of treatment became formed, the disease in young animals was milder. In 70% of the animals of the control group there was still high temperature and dyspnea, the pulse was rapid, feces were liquid.

To assess the effect of the drug on blood parameters of the experimental groups of calves after 7-day treatment, a comparative evaluation of some of its parameters was carried out (see Table 2).

Analysis of morphological blood parameters made it possible to estimate physiological state

of animals. The number of leucocytes in the control group was registered 23,13% higher ($p < 0,01$) compared to the upper threshold of the normative range³ (leucocytosis). Since the leucocyte index serves as an indicator of pathology, the presence of an inflammatory process was noted in this group. The use of phytobiotic preparations #1 and #2 contributed to the decrease in the number of white blood cells in the blood by 60,4% and 48,7% respectively, which indicates a decrease in the level of the disease in the examined animals. The number of erythrocytes in all groups under study was within the normal range, the level of hemoglobin in the control group was 10% lower than the lower threshold of normal and 20.5% lower than that

Табл. 1. Клинические показатели новорожденных телят в опытных группах ($n = 30, M \pm m$)

Table 1. Clinical indicators of newborn calves in the experimental groups ($n = 30, M \pm m$)

Group	Body temperature, °C	Pulse rate, bpm	Respiratory rate per minute
Norm	38,5–40,0	120–160	12–30
Experimental 1-st	40,7 ± 0,34	159,1 ± 2,82	27,1 ± 0,83*
2-nd	37,4 ± 0,23*	127,6 ± 1,87**	20,3 ± 1,16*
Control	41,5 ± 0,13	165,2 ± 2,11	33,4 ± 1,42

Here and in table 2.

* $p < 0,05$.

** $p < 0,01$.

*** $p < 0,001$.

Табл. 2. Сравнительная характеристика гематологических и биохимических показателей крови телят опытных групп ($n = 30, M \pm m$)

Table 2. Comparative characteristics of hematological and biochemical parameters of blood of calves of experimental groups ($n = 30, M \pm m$)

Indicator	Norm	Experimental group of animals		Control group
		1-st	2-nd	
Leukocytes (WBC, $10^9/l$)	8–16	11,9 ± 0,6	9,6 ± 0,40	19,7 ± 0,4**
Erythrocytes (RBC, $10^{12}/l$)	6–7,5	6,8 ± 0,89	7,2 ± 0,90	6,9 ± 0,79
Hemoglobin (HGB, g/l)	90–110	95,0 ± 2,50	102,2 ± 2,45*	81,0 ± 2,63
Hematocrit (HCT, %)	36–50	35,2 ± 5,30	38,7 ± 4,66	63,3 ± 5,80**
Potassium, mmol/l	3,5–4,5	3,9 ± 0,20	4,4 ± 0,51	2,9 ± 0,32
Sodium, mmol/l	135–148	141,1 ± 7,13	154,2 ± 6,21	75,6 ± 3,23**
Calcium, mmol/l	2,5–3,3	2,1 ± 0,44	2,7 ± 0,40	1,4 ± 0,14*
Phosphorus, mmol/l	1,4–1,9	1,3 ± 0,80	1,7 ± 0,74	1,2 ± 0,22
Ferrum, mmol/l	1,0–3,4	1,9 ± 0,36	2,5 ± 1,24*	0,7 ± 0,14
Glucose, g/l	2,2–3,2	2,5 ± 0,64	2,9 ± 0,22*	1,2 ± 0,83*

³Bazhibina E.B., Korobov A.V., Sereda S.V., Saprykin V.P. Methodological bases for assessing the clinical and morphological blood parameters of domestic animals: a training manual. Moscow: Aquarium-Print Ltd. 2007. 128 p.

in the 2nd group of animals. Hematocrit value in the experimental groups corresponded to normal values, it was 35.2 and 38.7%, respectively. In the control group this index was 26% higher than the norm ($p < 0.01$), indicating cell dehydration, characteristic of dyspeptic states of the body due to water loss [6, 7].

The results of the obtained biochemical tests showed that the sodium and potassium content in the blood serum of the control group animals were below the normal physiological values by 44,0 and 17,1% respectively, while in the experimental groups they were within the reference values. In calves of the control group there was low calcium content $1,4 \pm 0,14$ mmol/l ($p < 0,05$) as a result of disturbed absorption in the intestine while developing dyspepsia. Concentrations of inorganic phosphorus and iron corresponded to the upper limit of the physiological norm⁴.

It is known that iron deficiency in young animals is the trigger mechanism of impaired erythrocyte metabolism and reduction reactions in tissues, leading to profound anemia [8-11]. In young animals of the experimental groups this element was within the physiological norm, in the control group it was reduced by 30%.

The glucose level in animals of the control group decreased by 45.5%. Such changes occur due to disorders of lipid peroxidation leading to the accumulation of unoxidized metabolic products [9-13]. After application of phytobiotic preparations in the experimental groups after 7 days, the blood serum glucose content increased significantly ($p < 0,05$) by 2,7% in the 1st experimental group and by 3,9% in the 2nd group. When comparing the indices of the control and the 2nd experimental groups - by 12,1% ($p < 0,001$).

CONCLUSION

The use of drugs developed by us contributes to the normalization of morphological blood parameters (leukocytes, hemoglobin, hematocrit) and biochemical parameters of blood serum of calves (sodium, potassium, calcium-

phosphorus ratio, iron, glucose). Application of phytobiotic compositions positively affects the regulation of redox processes, metabolism, water-salt metabolism, the overall clinical condition of the animals. The preparation № 2 (Bergenia rhizome, fennel leaf, calendula flowers, probiotic) proved to be the most effective (by 10% compared with preparation № 1 and by 40% compared with the control).

Timely complex therapy of calves dyspepsia by the given preparation at a dose of 2 ml/kg 2 times a day will reduce the level of the disease in young animals by 40% and the terms of therapy up to 3 days. Livestock farms will be able to reduce losses from lost weight and young cattle mortality without losing the consumer qualities of livestock products.

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⁴Medvedeva M.A. Clinical veterinary laboratory diagnostics. Handbook for veterinarians. Moscow: LLC Aquarium-Print, 2008. 416 p.

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МЕТОДЫ НЕИНВАЗИВНОЙ ОЦЕНКИ ПОЛОВОГО ДИМОРФИЗМА ЭМБРИОНОВ В ЯЙЦЕ ПТИЦЫ

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Показана необходимость определения пола эмбрионов в яйце птицы неинвазивными методами до инкубации и в ее период. Применение неразрушающих методов оценки полового диморфизма на практике существенно уменьшит затраты на производство яиц и мяса сельскохозяйственной птицы. Внедрение таких методов также снизит нравственные проблемы, связанные с физическим уничтожением вылупившихся цыплят в зависимости от яичного или бройлерного направления профиля птицефабрик. Рассмотрены основные методы и технические средства определения полового диморфизма, применяемые в мировой практике, основным недостатком которых является сложность реализации и связанная с ней высокая цена приобретения таких средств. Проведен анализ существующих мировых тенденций по определению полового диморфизма эмбрионов в яйце птицы. Выявлены менее затратные методы оценки их пола до инкубации и во время ее проведения. Проанализированы основные неинвазивные методы оценки полового диморфизма эмбриона яйца и показаны преимущества и недостатки этих методов. Научная новизна исследований заключается в том, что впервые предложено использовать при оценке полового диморфизма все параметры формы, включая асимметрию яйца по трем пространственным координатам, определяемым методами компьютерного зрения. Предложена экспериментальная колориметрическая установка для оценки полового диморфизма эмбрионов яиц до и во время инкубационного процесса на основе компьютерного зрения. Использование экспериментальной установки при исследованиях позволит оценить эффективность не менее трех методов определения полового диморфизма эмбрионов в яйце курицы по следующим параметрам: пространственной асимметрии яйца, структурным изменениям развития эмбриона и частоте его сердечных сокращений.

Ключевые слова: яйцо, инкубация, эмбрион, диморфизм, неинвазивные методы, компьютерное зрение

METHODS FOR NONINVASIVE ASSESSMENT OF SEXUAL DIMORPHISM OF EMBRYOS IN THE POULTRY EGG

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The necessity of determining the sex in the bird's egg by non-invasive methods before incubation and during the incubation period is shown. The use of non-destructive methods for assessing sexual dimorphism in practice will significantly reduce the cost of producing eggs and poultry meat. The

introduction of such methods will reduce the moral problems associated with the physical destruction of hatched chickens, depending on the egg or broiler direction of the poultry farm profile. The main methods and technical means for determining sexual dimorphism, used in world practice, are considered, the main disadvantage of which is the complexity of implementation and the associated high cost of acquiring such tools. Analysis of current world trends in the determination of sexual dimorphism of embryos in the poultry egg was carried out. Less costly methods of estimating their sex before and during incubation were identified. The main noninvasive methods for assessing the sexual dimorphism of the egg embryo are analyzed and the advantages and disadvantages of these methods are shown. The scientific novelty of the research lies in the fact that for the first time it was proposed to use all shape parameters, including egg asymmetries in three spatial coordinates, determined by computer vision methods, when assessing sexual dimorphism. An experimental colorimetric apparatus for assessing the sexual dimorphism of egg embryos before and during the incubation process based on computer vision was proposed. The use of an experimental setup in research will allow evaluating the effectiveness of at least 3 methods for determining the sexual dimorphism of chicken eggs in terms of parameters: spatial asymmetry of the egg, structural changes in the development of the embryo and its heart rate.

Keywords: egg, incubation, embryo, dimorphism, noninvasive methods, computer vision

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Conflict of interest

The authors declare no conflict of interest.

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INTRODUCTION

Poultry eggs contain a large number of valuable micronutrients and nutrients, so they are among the most popular and essential foods. They are the main source of protein and are used to prepare a large number of traditional dishes, indispensable for dietary and sports nutrition.

There is a gender preference in chicken production, where the male is preferable for broiler production and the female for egg production. For broiler production, hens are inferior to cockerels due to their lower growth rate compared to their peers [1, 2]. Cockerels require additional feed expenses to gain weight. The use of hens in broiler farms is not economically justified because of the lower body weight gain compared to cockerels. However, cockerels cannot be used in the production of

eggs, the only product that is 97-98% digestible by the human body.

Because of gender-oriented production, more than 7.0 billion day-old cockerels are destroyed each year worldwide, resulting in significant economic losses [3, 4].

Many researchers have tried different strategies to determine the sex of an embryo before the chicks leave the egg and even before incubation based on differences in DNA content in the blastoderm, hormonal differences (estrogens) in the allantois fluid and the fluorescent properties of embryonic blood [5]. It is accurately established that sex differences exist in the composition of organic substances of embryos during incubation, in egg odor, DNA content, blood fluorescence intensity and in Raman scattering. However, these methods are rarely used in practice be-

cause they destroy the integrity of the egg structure.

A promising and effective method for determining the sex of an embryo in an egg should not affect the integrity of the egg shell or the embryo inside and have a negative impact on embryo development after the hatching and development process. The method must be fast-acting so that it can be applied to a large number of eggs, economically feasible in terms of application not only in large poultry farms of the country, but also in the farms of the Russian Federation, and also be acceptable from an ethical point of view.

The purpose of the study is to analyze current world trends in determining the sex dimorphism of poultry eggs and to identify less costly methods of sex estimation before and during incubation.

Existing methods and tools for assessing sexual dimorphism

In the study [6], a method of gender identification on the 9th day of incubation was proposed and tested by measuring estrone sulfate in the product of metabolism in the embryo - allantois fluid. Holes were drilled in the egg shell and 20-50 ml of this fluid were taken out with an insulin syringe. The research revealed that female embryos had a higher level of hormones in the allantois fluid than male embryos. The method was used to destroy eggs with male embryos on day 9 of incubation and was only economically advantageous compared to the traditional daily culling of male chicks. However, using this manual method to collect embryo biomaterial can damage embryonic structures and even lead to embryo death of the test egg.

In the study [7], a small hole was burned into the egg shell using a laser during the 8th to 10th day of the incubation period. Then, a small amount of the egg contents solution was extracted and the estrogen content was determined.

The German company Seleggt developed in 2017 the first functional and sought-after technology for determining gender using a hormone test¹. This technology uses a laser that also burns a hole no larger than 0.3 ml into an eggshell.

This method is based on endocrinological process and allowed to determine the sex of the future chick 8-10 days after oviposition with 98% reliability. The productivity with this method was 3600 eggs per hour.

Fluorescence [8, 9] and Raman spectroscopy [10, 11], named after the Nobel Prize-winning Indian physicist Chandrasekhara Venkata Raman, are widely used in the studies of embryonic sex determination. Raman spectroscopy is based on inelastic scattering of photons, known as Raman scattering, accompanied by a noticeable change in the frequency of radiation.

Researchers at the University of Leipzig put into practice a combination of fluorescence and Raman spectroscopy methods [12]. The studies were performed at the early stages of embryonic development, when the embryo is not yet sensitive to pain [13, 14]. Indeed, when a chicken egg is incubated up to 5 days old, the embryo has a primitive blood flow providing gas exchange through the extra-embryonic vascularized area of the yolk sac. The diameter of this area is about 30 mm.

During the study, the egg was placed in a vertical position with the blunt end of the egg facing upwards (see Fig. 1). The embryo in this case was in the center of the vascularized area below the air chamber at the blunt end of the egg. In this case, the air chamber is localized between the inner shell, which directly contacts the protein, and the outer shell, which adjoins the shell. At the blunt end of the egg the outer shell was opened together with the shell. When a window is formed in the shell at the blunt end, the inner membrane of the embryo remains intact. Given that it is

¹In-ovo sexing URL: https://en.wikipedia.org/wiki/In-ovo_sexing#:~:text=In%20poultry%20farming%2C%20in-ovo%20sexing,company%20Seleggt%20in%20November%202018 (reference date 25.04.2022).

quite thin, an embryo with yolk circulation can be seen through the membrane (see Figure 2).

This fact made it possible to identify vessels under a Raman microscope, irradiate circulating blood with a near-infrared laser, and obtain a backscatter spectrum.

The laser-induced infrared radiation spectra obtained from embryonic blood vessels consisted of Raman scattering bands. The scattering bands were biased with the fluorescence arising from the laser radiation. As the researches have shown, at excitation by infrared radiation with wave length ≈ 910 nm, the intensity of fluorescence of eggs of females I_f appeared lower in comparison with the intensity of fluorescence of eggs of males I_m ($I_f = 91$, $I_m = 68$). According to the authors of this

study, the reliability of gender determination using this method was 93%.

It should be noted that this method is invasive and has a negative effect on the growth and development of hatchlings.

In Ovo company together with the University of Leiden, has created an automated method for micro-sampling fluid from an egg at a sampling rate of three samples per second. Using a fast-acting mass spectrometer Sciex Echo® MS, a robotic system has been created that can sort embryos by gender on day 9 of their development².

However, such a technological complex for sorting eggs by gender is not available to domestic consumers because of its complexity of implementation and high cost.

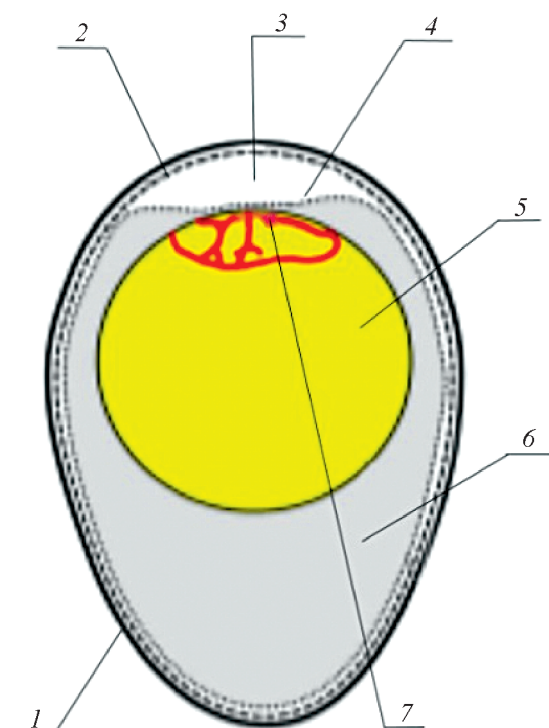


Рис. 1. Структура и расположение яйца при его препарировании:

1 – скорлупа; 2 – наружная подскорлуповая оболочка; 3 – воздушная камера (пуга); 4 – внутренняя подскорлуповая оболочка; 5 – желток; 6 – белок; 7 – эмбрион

Fig. 1. The structure and arrangement of the egg in its dissection:

1 – shell; 2 – outer shell membrane; 3 – air cell; 4 – inner shell membrane; 5 – yolk; 6 – protein; 7 – embryo



Рис. 2. Внешний вид кровеносных сосудов эмбриона на 5-й день инкубации при вскрытии наружной оболочки яйца

Fig. 2. The appearance of the blood vessels of the embryo on the 5th day of incubation when opening the outer shell of the egg

²In Ovo URL: <https://www.cbinsights.com/company/in-ovo>. Peebles E.D. In ovo applications in poultry: A review 2018. Vol. 97. Is. 7. P. 2322–2338. DOI: 10.3382/ps/pey08.

Analysis of promising low-cost methods for assessing sexual dimorphism

Currently, intensive research is underway to develop relatively uncomplicated methods for assessing the sex of embryos in a poultry egg.

The mystery of the beginnings of the chicken was attempted by the eminent ancient scientist Aristotle³. He assumed that oblong eggs would produce cockerels and rounded eggs would produce hens. Although this fact was not confirmed in practice, his idea of the difference in shape and geometric parameters of the egg in determining the sex finds its way in domestic and foreign studies. Thus, a number of Russian scientists believe that the sex can be determined before hatching by the shape index⁴, which is a ratio of the longitudinal l and transverse b geometrical size of the egg. The authors have stated by measuring this size with a laser interferometer that if the ratio $l/b = 1,2-1,3$, these are the future hens, $l/b = 1,4-1,5$, the future cockerels.

Considering that the volume and the surface area of eggs are reliable prognostic parameters of the quality characteristics of hatching hen eggs, many researchers suggest performing a geometric transformation of the actual contour of the egg into a known geometric figure whose shape most closely resembles the egg under study [15-19].

As a rule, four geometric figures were used in the analysis of all egg shapes: sphere, ellipsoid, ovoid and pear-shaped [20].

If the first three figures had a clear mathematical definition, each of which was obtained from the expression of the previous one, the formula for the pear-shaped profile was derived quite recently. In this case, in addition to the parameters of the shape index, the values of the shift of the vertical axis of the egg and the diameter of the pointed end of the egg at the length of $l/4$ were used. Consideration of

these four parameters allowed scientists V.G. Narushin, M.N. Romanov (Ukraine) and D.K. Griffin (Great Britain) to derive a universal equation for determining the contours of bird eggs existing in nature [21]. It can be expected that the analytical equations obtained in the studies will not only be the basis for research in evolutionary biology, but will also be used to predict the sex of an egg before its incubation.

Computer vision systems are intensively used to control all technological processes in poultry farming [22], the structural diagram of which is shown in Fig. 3.

Application of computer vision reduces the cost of labor-intensive processes associated with the increasing intensification of production of poultry farms, the need to create acceptable conditions for the welfare of animals. Computer vision systems can provide reliable, non-invasive and precision technology for probing and monitoring various aspects of poultry production processes. They will provide a large variety of data on the assessment of the vital functions and prognosis of the sexual characteristics of embryonic eggs for further analysis. Since computer vision uses a mathematical apparatus in image acquisition, it will be effective in developing the methods for predicting sex traits by shape parameters, including asymmetry of eggs by three spatial coordinates, before or during their incubation.

It is extremely difficult to make such predictions, despite the fact that computer methods perform identification, image classification, semantic segmentation, detection and recognition of the structural elements of the studied objects using machine learning methods based on deep neural networks [23].

The results of studies of the relationship between linear egg size in wild birds (sparrow, lapwing, canary, European thrush, lark, etc.) and sexual dimorphism are rather pessimistic

³Aristotle (384-322 BC). On the Origin of Animals, translated from Greek, Moscow; L.: USSR Academy of Sciences, 1940. 252 p.

⁴Patent (RN) № 2238643. Method of automated sorting chicken eggs by gender. N.V. Vasilenko, E.N. Ivashov, V.V. Protsenko, Stepanchikov S.V.; Application. 07.05.2002; publ. 27.10.2004; Bul. no. 30.

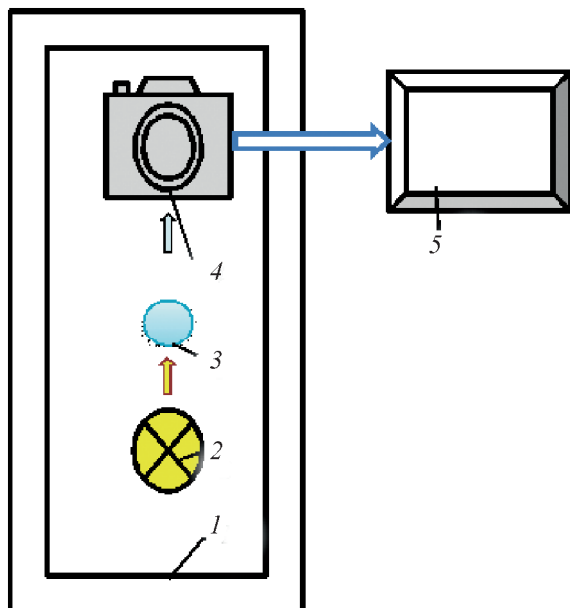


Рис. 3. Структурная схема экспериментальной установки компьютерного зрения:

1 – темная комната; 2 – RGB-осветитель; 3 – яйцо; 4 – фотоаппарат; 5 – ноутбук

Fig. 3. Structural diagram of the experimental setup for computer vision:

1 – dark room; 2 – RGB illuminator; 3 – egg; 4 – camera; 5 – laptop

and do not give an unequivocal answer [24].

In this case it is proposed to determine the asymmetry of linear dimensions of an egg relative to three spatial coordinates. The aspiration for shape symmetry in living organisms is a well-known fact, which is explained by the decrease of entropy in regulated systems [25].

As a scientific hypothesis, it is assumed that the nature of asymmetry in spatial coordinates in male and female embryos of hen eggs is different. Confidence in the confirmation of the hypothesis is provided by the preliminary results of predicting the sex of chicken eggs by the distributed parameters of the shape index in two spatial coordinates [26]. In this study, all eggs were set in the same spatial position. The distributed parameters of the egg shape index were determined on a contrasting white

and black image of the egg. The background of the egg was transformed into black. All transformations were performed using the standard MATLAB⁵ image processing library. The values of shape index distributions for each egg from the selected batch were transferred to the database as pixels. The results were obtained by intelligent data analysis using the Random-Forest machine learning algorithm.

A batch of 262 chicken eggs was examined. After incubation, 116 females and 106 males were hatched; 40 eggs were rejected. According to the authors of the study, the reliability of prediction of male and female chicks was 93 and 100%, respectively.

Computer vision methods for predicting the sex of chicks are also used during the incubation period. For example, in the work [27], using LED-type light sources in the computer vision unit, images of two batches of chicken eggs on the 3rd-6th, 8th, and 10th days of incubation, respectively, were obtained.

In Fig. 4, blood vessels are clearly enough visible in the obtained images during the initial period of incubation. In the images, 11 regions of features characterizing embryo sexual dimorphism are highlighted. After processing these feature parameters, which display the texture features of blood vessels depicting a chick embryo, a genetic algorithm was developed to optimize the initial weights and back-propagation thresholds of neural networks with different hidden layers.

Machine learning was based on the Back Propagation Neural Network (BPNN) algorithm.

The authors claim that machine vision technology provides a realistic method of determining the chicken egg sex on day 4 of incubation with a prediction accuracy of 89.74%.

Differences in the shape of hatching eggs, changes in the ambient temperature and humidity, the presence of calcium spots on the shell, etc., have an impact on the reliability of sex determination.

⁵ImageProcessingToolbox-The MathWorks-PDF Catalogs pdf.directindustry.com <https://www.mathworks.com/help/images/functionlist.html?requestedDomain=www.mathworks.com#btvphx9-1>.

There are studies on assessing the viability of bird embryos by measuring the heart rate [28-30]. For example, in the work [30] a method of assessing the activity of chicken embryos was described using near-infrared (870 nm wavelength) imaging of the egg, obtaining the embryo heart rate signal and its further processing. The heart rate during the incubation period varied from 3.8 to 4.8 Hz. Moreover, during the incubation first there was an increase in the heart rate, on the 15th day there was a decrease.

Considering that cardiac activity in the embryo is detected on the 2nd day of incubation [28], it is possible to develop a new method for early assessment of the sexual dimorphism in the incubated egg based on the hypothesis of the difference of heart rate in cockerel and hen embryos. Such method of sex dimorphism estimation was not found in the analyzed sources. The fact of the difference in the cardiac activity of the two sexes of embryos can be refuted or proved only by applying modern methods and computer vision tools.

In accordance with the structural scheme (see Fig. 3), a synthetic vision unit was created to conduct research on the selection and justification of a relative cheap method for determining the sexual dimorphism of embryos in a chicken egg both before their incubation and

during the first days of the incubation period.

Digital camera Canon EOS 2000D EF-S 18-55 III Kit with modern CMOS-matrix (22,3 × 14,9 mm) and a powerful processor was used to obtain images in the synthetic vision unit. The maximum resolution of the matrix is 6000 × 4000 pixels, color depth is 42 bits per pixel. The range of sensitivity of the sensor is from 100 to 6400 ISO, the crop factor is 1.6. Exposure is automatic with aperture and shutter priority. The viewfinder is a reflex (TTL) with a 95% field of view. Fast shooting speed is three frames per second.

All components of the camera work together to produce sharp images with minimal noise and bright, saturated colors.

The digital device is attached by a threaded connection to the removable head of the Raylab Travel telescopic tripod. The position of the tripod head is adjustable in three spatial planes. The tripod is made as three sections - supports with adjustable movable legs and a sliding central post. This design makes imaging easier and more productive. The sliding central pole guarantees the process of shooting from the right angle. The adjustable working height of the camera ranges from 0.56 to 1.6 m to the floor. The camera is mounted on a tripod on which the slide with the object under study is fixed.

The digital device is paired with a laptop model ASUS VivoBook 17 K712EA-BX467W, running the operating system Windows 11 Home Single Language. The characteristics of its screen: the diagonal of 17.3"; resolution of 1600 × 900 pixels; brightness of 200 Cd/m².

The laptop is equipped with an Intel Pentium Gold 7505 processor with 8 GB of RAM. It provides powerful performance and wide viewing angles.

When determining the geometric parameters and asymmetry of the shape of an incubated egg there is a light kit for Raylab RL-LED10 Kit 3200-6500K laptop and IEK IO spotlights with halogen light sources. The kit light allows you to smoothly change the power

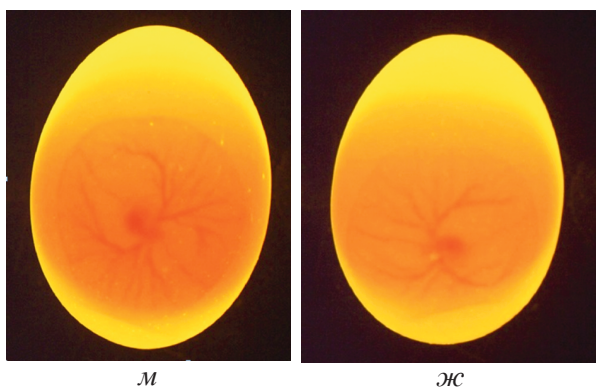


Рис. 4. Изображение зародыша куриного яйца на 4-й день инкубации:

м – петушок; *ж* – курочка

Fig. 4. Image of a chicken egg embryo on the 4th day of incubation:

м - cockerel; *ж* - female chicken

from 1 to 100%, as well as the color temperature in the range of 3200-6500K. It is equipped with a swinging bracket that helps to create the necessary light and eliminate shadows on the object of study or illuminate it.

When determining the sexual dimorphism in the first days of incubation, the Falcon Eyes ML-09 RGB LED lamp with infrared remote control is used as an illuminator. Power of the lamp is 9 W. Taking into consideration that a standard egg weighing 58 g has cross and longitudinal diameters of 42 and 57 mm respectively, the matte bulb with a diameter of 60 mm illuminates the egg with uniform white light of maximum brightness. In addition to the basic colors, the RGB mode offers 12 presets of different color shades, which are set with the control panel.

The LED lamp is placed in a light-tight cylindrical aluminum housing, which is fixed in the geometrical center on the inner side of the stage table top. A round hole 80 mm in diameter is made in advance in the table top, in which the bulb of the LED lamp enters. A removable thin black plate with a cut out oval ellipse is placed on the outer side of the table top with a pre-made round hole. Thus, when you turn on the LED lamp in a dark room a contrast illumination of the incubated egg throughout its incubation period is carried out.

The object of research (egg) is set in this oval, which limits the movement of the egg on the plane of the plate, and also gives the possibility to fix the egg when it turns along its longitudinal axis from 0 to 360 °.

A series of experimental studies are planned using this unit to identify a least-cost noninvasive method for assessing sexual dimorphism in the egg.

It should be noted that not all possible methods of sex determination in bird embryos, such as methods of hyperspectral imaging [31], impedance spectroscopy [32], etc., are considered in this article. The reason for this is the lack of information in the analyzed sources on the effectiveness of the practical implementation of these methods in the developed countries of the world.

CONCLUSIONS

1. Existing methods of determining dimorphism in poultry eggs are invasive and are usually carried out after the process of their incubation when the chicks are at least one day old.

2. Lack of nondestructive methods and means of determining the sexual dimorphism of embryos in a poultry egg raises a serious ethical problem associated with the destruction of live chicks by maceration or suffocation with carbon dioxide, and leads to significant economic losses in the poultry industry.

3. The use of the In Ovo's foreign technology, in which a micro-hole in the egg shell is burned out with a laser and the sex of the future chick is determined using the Sciex Echo® mass spectrometer, is impossible because of its high cost, despite the creation of high-performance robotic complexes using this technology, capable of sorting embryos by sex on day 9 of their development.

4. Analysis of the main promising methods for assessing the sex of embryos in a chicken egg before and during the incubation period, based on the application of vision techniques, using machine learning has been carried out.

5. An experimental unit of vision designed for further research to scientifically substantiate and create a low-cost, non-invasive method of assessing embryo sex dimorphism in a chicken egg has been manufactured and described.

6. The application of the experimental unit in the research will provide reliable results to assess the effectiveness of the method of determining the sex dimorphism by the spatial asymmetry of hen eggs before incubation. In addition, the minimum days of the incubation period for predicting the sex of embryos by their structural changes will be established, and the probability of such prediction by the heart rate of the developing egg in the embryo will be determined.

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ИНФОРМАЦИЯ ОБ АВТОРЕ

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ПРОБЛЕМЫ И ПЕРСПЕКТИВЫ ЦИФРОВИЗАЦИИ СЕЛЬСКОГО ХОЗЯЙСТВА В РЕСПУБЛИКЕ ТЫВА

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Изучены проблемы и перспективы развития цифровых технологий в аграрном секторе Республики Тыва. В Тыве использование цифровых технологий остается крайне низким. По развитию процессов цифровизации регион существенно отстает как от показателей России в целом, так и Сибирского федерального округа. Вызовами, сдерживающими развитие информационного общества в Республике Тыва, являются наличие труднодоступных населенных пунктов со сложными географическими особенностями; инфраструктурные ограничения в части оформления (согласования) необходимых документов и взаимодействия с сетевыми организациями в рамках заключения договоров на технологическое присоединение к электросетям; отсутствие покрытия и неуверенный прием подвижной радиотелефонной связи в удаленных сельских населенных пунктах Республики Тыва; нехватка квалифицированных специалистов по информационно-телекоммуникационным технологиям, слабое развитие научно-инновационного потенциала; низкие навыки использования информационных технологий; импортозависимость от зарубежного программного обеспечения, технологий и оборудования; цифровое неравенство между муниципальными образованиями. Для повышения эффективности сельского хозяйства в регионе необходимо внедрение в сельскохозяйственное производство цифровых технологий: сбор данных о посевных площадях и пастбищах, составление цифровых карт земель сельскохозяйственного назначения, использование дистанционно управляемых датчиков по мониторингу за качеством посевов сельскохозяйственных культур, над передвижением сельскохозяйственного транспорта и животных и др. Применение цифровых технологий будет способствовать продвижению продукции сельхозтоваропроизводителей до потребителя, что расширит доступ к финансовым ресурсам и рынкам за счет согласованного действия всех звеньев аграрного производства. Намечены основные направления цифровизации отрасли и обозначены перспективные цифровые технологии для применения в отрасли сельского хозяйства Республики Тыва.

Ключевые слова: сельскохозяйственное производство, цифровые технологии, ГИС-технологии

PROBLEMS AND PROSPECTS OF DIGITALIZATION OF AGRICULTURE IN THE REPUBLIC OF TYVA

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The problems and prospects of development of digital technologies in the agricultural sector of the Tyva Republic were studied. The use of digital technology remains extremely low in Tyva. The region significantly lags behind Russia as a whole and the Siberian Federal District in the development of digitalization processes. Challenges hindering the development of the information society in the Republic of Tyva are the presence of hard-to-reach settlements with complex geographical features; infrastructure restrictions in terms of registration (approval) of necessary documents and

interaction with network organizations in the conclusion of contracts for technological connection to the power grid; lack of coverage and uncertain reception of mobile radio telephone communications in remote rural settlements of the Tyva Republic; lack of qualified specialists in information and telecommunication technologies, poor development of scientific and innovation potential; low skills in using information technologies; import dependence on imported software, technologies and equipment; digital inequality between municipalities. To improve the efficiency of agriculture in the region, it is necessary to introduce digital technologies into agricultural production: collecting data on sown areas and pastures, making digital maps of agricultural land, using remote-controlled sensors to monitor the quality of crops, the movement of agricultural vehicles and animals, etc. The use of digital technologies will promote the products of agricultural producers to the consumer, which will expand access to financial resources and markets due to the coordinated action of all parts of agricultural production. The main directions of digitalization of the industry were outlined and promising digital technologies for application in the agricultural sector of the Tyva Republic were specified.

Keywords: agricultural production, digital technologies, GIS-technologies

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The authors declare no conflict of interest.

INTRODUCTION

Currently, one of the most important factors in the strategic development of agricultural production is the introduction of digital technologies in agricultural production. Digital transformation of agriculture and the planned technological breakthrough is a key factor in the development and one of the main trends in the agricultural industry in the coming years [1].

Information and digital technologies in agriculture, contribute to the formation of sustainable development of not only the agro-industrial complex, but also the development of rural areas. Thanks to these technologies, it is possible to obtain data on each agricultural object and its environment, calculate mathematically accurate algorithm of action and predict the result¹.

Digitalization has affected not only the IT sector, retail, financial institutions and social networks, where its level reaches 70-80%, but also sectors of the real economy [2]. Agriculture is no exception, because the organization of highly profitable production already objectively involves its digitalization².

The Government of the Russian Federation recognizes the need for digitalization of the agro-industrial complex, aimed at increasing agricultural production and profitability of the industry. The programs adopted by the state see their task as the consolidation of the efforts of science, federal and regional executive authorities and business for the development of domestic technologies that help to reduce import dependence and ensure sustainable development of the agro-industrial complex of the country [3].

¹Shpedt A.A., Zlotnikova V.V. Application of information and digital technologies in the agroindustrial complex of the Krasnoyarsk region. Information technologies, systems and devices in the agroindustrial complex: materials of the 7th International Scientific and Practical Conference "Agroinfo-2018". Krasnoyarsk, 2021. pp. 22-25.

²Smyshlyaev A.A., Medvedeva Zh.V. Digitalization of cattle breeding in the Altai Territory. Economic and mathematical methods of analyzing the activities of enterprises of the agroindustrial complex. pp. 260-266.

The government has developed a departmental project "Digital Agriculture"³, which provides for the implementation of a number of important areas to improve agriculture through the implementation of information technology in various constituent entities of the Russian Federation.

The purpose of the study is to examine the problems and prospects of digitalization in the agricultural sector of the economy of the Republic of Tyva.

MATERIAL AND METHODS

The study used information from the Ministry of Digital Development of the Republic of Tyva on the implementation of the state program "Development of Digital Society in the Republic of Tyva in 2014-2020", information from the department of organizational, documentary and information support of the Ministry of Agriculture of the Republic of Tyva as of 01.07.2022, the information is from the Krasnoyarskstat, as well as our own research data.

RESULTS AND DISCUSSION

The Republic of Tyva is part of the Siberian Federal District. One of the main sectors of the regional economy is agriculture. There are 327.38 thousand people, of which 149.6 thousand (45.7%) live in rural areas⁴. In this regard, the state of this industry largely predetermines the economic and social situation of the region as a whole.

The main direction of agricultural production is cattle breeding, which accounts for 83.32% of agricultural production. The Republic of Tyva has large areas of natural pastures, which allows to develop distant-pasture cattle breeding. Sheep breeding, meat herd horse breeding, cattle breeding and yak breeding are of great importance.

The region is located in the zone of risky agriculture. Here crop farming is conduct-

ed on the verge of possible farming, which makes it difficult to conduct this industry. With 2653 thousand hectares of agricultural land in Tyva only 135.5 thousand hectares is arable land, and currently a significant part of it is not cultivated. The average yield of grain crops in the last 5 years is about 13.1 cwt/ha (see footnote 4).

There are also a number of problems that hinder the development of cattle breeding. Despite the positive dynamics of growth of livestock, there is low productivity of animals and, as a consequence, the decline in livestock production.

One of the key factors in improving the efficiency of agriculture can be the introduction of digital technologies in agricultural production of the republic, which will improve the efficiency of agricultural production through the coordinated action of all parts of agricultural production. These include the introduction of elements of digital inventory of cultivated areas and pastures, making digital maps of agricultural land, etc. In the future it is possible to use remotely controlled sensors to monitor the quality of crops, the movement of agricultural vehicles and animals. In addition, agricultural producers with the introduction of digital technology will be able to trace the progress of their products to the consumer, which will increase access to financial resources and markets.

The use of digital technology in the Republic of Tyva remains extremely low. The region significantly lags behind both Russia as a whole and the Siberian Federal District in the development of digitalization processes.

Tyva ranks 78th according to the rating of the regions for the development of the information society in the Russian Federation. According to the survey, the number of households with personal computers was 58.1%, including 66% in urban areas and 47.4% in rural areas⁵.

In October 2013, the Resolution of the Government of the Republic of Tyva approved

³Departmental Project "Digital Agriculture": Official publication. Moscow: Rosinformagrotech, 2019. 48 p.

⁴Krasnoyarskstat. URL: <https://krasstat.gks.ru/folder/95045>.

⁵Program "Development of Information Society in the Republic of Tyva in 2014-2020": URL: <https://docs.cntd.ru/document/460195149/titles/133MPTQ>.

the state program "Development of Information Society and Mass Media in the Republic of Tyva for 2014-2020. (see footnote 5).

The main goals of the state policy in the sphere of information and telecommunication technologies in the Republic of Tyva are⁶:

- improving the well-being and quality of life of citizens by increasing the availability and quality of goods and services produced in the digital economy using modern digital technologies, increasing the degree of awareness and digital literacy, the quality of public and municipal services;

- modernization and development of the print and electronic media infrastructure, book publishing and printing activities in the Republic of Tyva to ensure the constitutional rights of citizens to access to information;

- introduction of digital technologies in the economy, including tourism, urban planning, housing and public utilities, and agriculture.

The challenges hindering the development of the information society in the Republic of Tyva, according to the developers of the program, are:

- availability of hard-to-reach settlements with complex geographical features;

- infrastructural constraints with regard to the execution (approval) of necessary documents and interaction with grid operators as part of the conclusion of contracts for technological connection to electrical grids;

- lack of coverage and uncertain reception of mobile radio telephone communications in the remote rural settlements of the Republic of Tyva;

- lack of qualified specialists in the field of information and telecommunication technologies (ITT), and poor development of scientific and innovation potential;

- low skills in the use of information technologies;

- import dependence on foreign software, technology and equipment;

- digital divide between municipalities.

In our opinion, the implementation of the program for the development of the information society and mass media in Tyva in 2014-2020 has not had a significant impact on the level of development of digital technologies directly in the agro-industrial complex of the republic. However, there is a trend in the region to improve the information and digital infrastructure for the provision of public services in electronic form by the authorities of the republic, which has contributed to the awareness and digital literacy of the local population.

In this regard, the public authorities of the republic need to pay attention to the development of a separate regional program for the development of the digital economy in agriculture, which includes measures to support the entities involved in agricultural production for the implementation of information systems, the purchase of server equipment, advanced training, etc.

One of the most promising areas in the digitalization of agriculture in Tyva for the first time could be the use of information systems based on geoinformation technology. The introduction of such a system in agriculture will serve as the first step towards the creation of a regional GIS, which will increase the efficiency of agricultural production in the region.

GIS technologies in the field of crop production would make it possible to centrally maintain a register of cultivated areas of crops, monitor agronomic operations, digitize crops, make crop yield forecasts, etc.

In cattle breeding, GIS-technologies can be used to select feed rations, monitor feeding and housing, and control the quality of agricultural products.

In the Republic of Tyva, where cattle breeding is the native occupation of the population of the region, the software product "Horse Breeders Assistant", information retrieval systems "IRS Kony", "IRS Kony 2", "IRS Kony 3"⁷ can be recommended for conducting meat herd horse breeding.

⁶Decree No. 482 of the Government of the Republic of Tyva dated 27.10.2017.

⁷Assistant horse breeder. URL: <http://horsemate.narod.ru/index/0-2>.

The program "Coral" and its software products: "Coral - Cattle Farm", "Coral - Feeding", "Coral - Feed base" and others, aimed at optimizing the issues of feeding, housing and creating veterinary welfare of animals using information technology, appeared in the register of complex programs of the Russian Federation⁸.

Currently, in our country there are examples of successful use of GIS-technologies in the field of veterinary medicine as well, which allows to conduct epizootological monitoring, forecast and control the epizootic process in a particular territory by promptly influencing it through GIS-technologies [4-6].

Scientists from the Institute of Experimental Veterinary Science of Siberia and the Far East and the Siberia Physical and Engineering Institute of Agrarian Issues SFSCA RAS created a computer system "Leukosis" using GIS-technology to analyze the epizootic situation on leukosis.

The developed system is intended for accumulation, systematization and subsequent analysis of epizootic and background information in order to determine the patterns of epizootic process development, on the basis of which managerial decisions for localization and elimination of bovine leukosis with the least economic and time losses will be elaborated [7].

For example, the use of the MapInfo system makes it possible to form electronic cartographic databases for creating systems of forecasting, distribution and circulation of helminth infestations of farm animals [8, 9].

The introduction of information technologies (GIS-technologies) in forecasting the epizootic process in the border territories of Tyva is also relevant. The state border between Russia and Mongolia runs through the southern territory of the republic. The length of the Tuvan section of the border is more than 1,300 km, or about 38% of the Russian-Mongolian border [10]. The successful development of

cattle breeding in this territory depends to a large extent on the epizootic welfare of the border area for infectious and invasive animal diseases. According to the veterinary service of the Republic of Tuva, for a long time in Mongolia there has been a tense epizootic situation on infectious animal diseases, where cases of unfavorable sites for anthrax, pasteurellosis, rabies, horse strangles, rinderpest, foot and mouth disease and other were registered in different years.

In this regard, the introduction of digital information technology in the practical work of veterinary professionals can provide a rapid exchange of information and create a common database for effective anti-epizootic measures in the border territories.

In the territories of the Republic of Tyva bordering Mongolia, the border service often has to stop cattle from entering "foreign" territory, which can lead to conflicts related to grazing. Here an example of successful introduction of information technology could be the procedure of implanting microchips under the skin of animals. Microchip allows to trace the movement of the animal and conduct their identification within hours, which excludes the theft and replacement of the animal.

CONCLUSION

Currently, the effective development of agriculture is impossible without the formation and implementation of elements of modern digital technologies in agricultural production. Their development in the region requires state support for agricultural producers, regardless of their form of ownership, as well as the inflow of investment in agriculture at the expense of agricultural producers themselves and other entities of agricultural business through investment funds. The creation of a digital economy in Tyva can ensure the sustainable development of the agricultural sector and increase its competitive advantage.

⁸Lukyanov B.V., Lukyanov P.B. User's Manual for the CORAL Computer Programs. URL: <https://www.korall-agro.ru/KORALL.pdf>.

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ИНФОРМАЦИЯ ОБ АВТОРАХ

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НОВЫЕ МЕСТОНАХОЖДЕНИЯ ДЕКОРАТИВНОЙ ОРХИДЕИ *SPIRANTHES SINENSIS* (ORCHIDACEAE) В ПРИМОРСКОМ КРАЕ

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Приведены сведения о скрученнике китайском *Spiranthes sinensis* (Pers.) Ames (Orchidaceae Lindl.) – перспективном виде для использования в цветоводстве в условиях Приморского края. В данном регионе скрученник китайский встречается практически во всех районах, однако известные современные местонахождения имеют низкую численность популяций. Это декоративное, длительно цветущее орхидное растение хорошо размножается вегетативно, переносит засуху, сильное уплотнение почвы и долговечно в травостое. Как и другие луговые виды орхидей, скрученник китайский может расти на солнечных местах и в полутени, в сочетании с сопутствующими некрупными растениями. К лимитирующим факторам относится нарушение естественных мест произрастания в результате экстремальных природных явлений и антропогенного воздействия. Описаны новые местонахождения *S. sinensis* в Приморском крае. Полевые исследования проведены в 2018–2021 гг. маршрутно-рекогносцировочным методом. Осуществлено описание местообитаний, а также морфометрических показателей скрученника китайского, проведен анализ гербарных образцов (VLA, VBGI, MW, MHA, LE) этого растения на Дальнем Востоке России. На территории Приморского края выявлены новые местообитания скрученника китайского, описаны пять ценопопуляций. Площадь участков составляла от 8 до 936 м², число цветущих особей – от 12 до 28 экз., плотность популяции достигала 5 экз./м². Высота цветущего растения может достигать 64 см. Вид не включен в Красную книгу Российской Федерации (2008 г.), а также в Красную книгу Приморского края (2008 г.), поэтому можно использовать его в регионе для применения в озеленении в качестве красиво цветущего травянистого растения.

Ключевые слова: *Spiranthes sinensis*, Orchidaceae, Приморский край, местообитание, сопутствующие виды, декоративное орхидное

NEW LOCATIONS OF THE DECORATIVE ORCHID *SPIRANTHES SINENSIS* (ORCHIDACEAE) IN THE PRIMORSKY TERRITORY

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The information is given about the Chinese ladies'-tresses *Spiranthes sinensis* (Pers.) Ames (Orchidaceae Lindl.), which is a promising species for use in the floriculture under conditions of the Primorsky Territory. In this region, the Chinese ladies'-tresses is found in almost all areas, but the known present-day locations have low number of populations. This decorative, long-blooming orchid propagates well vegetatively, tolerates drought, severe soil compaction, and is long-lived in grasses. Like other meadow species of orchids, the Chinese ladies'-tresses can grow in sunny places and in the

penumbra, in combination with accompanying small plants. Limiting factors include disturbance of natural habitats as a result of extreme natural phenomena and anthropogenic impact. New locations of *S. sinensis* in the Primorsky Territory are described. Field studies were conducted in 2018-2021 by route reconnaissance method. Description of habitats and morphometric parameters of Chinese ladies'-tresses, analysis of herbarium specimens (VLA, VBGI, MW, MHA, LE) of this plant in the Russian Far East was carried out. In the Primorsky Territory, new habitats of the Chinese ladies'-tresses have been identified and five cenopopulations have been described. The area of the plots ranged from 8 to 936 m², the number of flowering individuals was from 12 to 28 specimens, and the population density reached 5 specimens/m². The height of a flowering plant can reach 64 cm. The species is not included in the Red Book of the Russian Federation (2008) or in the Red Book of the Primorsky Territory (2008), so it can be used in the region for landscaping as a beautiful flowering herbaceous plant.

Keywords: *Spiranthes sinensis*, Orchidaceae, Primorsky Territory, habitat, related species, decorative orchid

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Conflict of interest

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INTRODUCTION

The *Orchidaceae* Lindl. is one of the largest plant families on Earth. The number of known species includes about 30 thousand, of which 129 grow in Russia. At present, only 40 orchid species are known in the Primorsky Territory [1]. Many outdoor orchids are rare protected plants. At present, winter-hardy orchids are cultivated by collectors for commercial purposes, besides, samples of different species are tested in botanical gardens. Specialists note difficulties with their breeding associated with narrow specialization to habitat conditions, as well as obtaining a sufficient amount of planting mate-

rial. As a result of long-term studies of vegetative and seed reproduction of some representatives of genera *Cremastra* Lindl., *Cypripedium* L., *Dactylorhiza* Nevski, *Epipactis* Zinn., *Gymnadenia* R. Br., *Oreorchis* Lindl., *Platanthera* Rich. from different geographical areas, many authors noted that a large number of temperate orchid species can grow in gardens as ornamental plants¹⁻³ [2].

At present, about 15 species of the genus *Cypripedium* are cultivated in the nurseries and gardens of Northern and Central Europe, the USA and Canada (see footnotes 2, 3) [3]. From the orchids of the Primorsky Territory, *C.*

¹Konovalova T.Yu. Species of natural orchids, the most promising for floriculture in the midland of Russia. Floriculture: History, Theory, Practice: Proceedings of the VII International Conf. (May 24-26, 2016. Minsk, Belarus). Minsk: Confido, 2016. pp. 141-143.

²Chu C.C., Mudge K.W. Propagation and conservation of native Lady's Slipper Orchids (*Cypripedium acaule*, *C. calceolus*, *C. reginae*) // North American terrestrial orchids. Propagation and production. Conference proceedings. Germantown, Maryland, 1996. P. 107–112.

³Cribb P. The Genus *Cypripedium*. Portland, 1997. 301 p.

guttatum Sw., which is considered one of the most resistant in culture, was tested in the Moscow Region [4-6]. Introduction of natural orchid plants into culture will greatly enhance the aesthetic perception of the surrounding world. Specialists are interested in the introduction of as many species of these ornamental plants as possible and in the development of their agrotechnics.

Chinese ladies'-tresses is a herbaceous plant up to 50 cm in height, a short-rhizomatous polycarpic with thickened adventitious roots [7]. Decorative, long-blooming orchid with unusual twisted inflorescence. According to our data, the height of a flowering plant in the Primorsky Territory can reach 64 cm.

Chinese ladies'-tresses is one of the most widespread species of terrestrial orchids, growing in various ecotopes, often found in disturbed habitats. Usual locations of the plants are low grass meso- and xerophytic meadows, roadsides of field roads and coastal areas, less often found in sparse woodland, on forest edges and wetlands. It survives drought and strong soil compaction well [1]. It is a hygromesophytic, light-loving, meadow-burrowing (meadow) species.

Limiting factors are the disturbance of natural habitats as a result of extreme natural phenomena and anthropogenic impact.

According to the International Union for Conservation of Nature, *S. sinensis* has no threats to extinction, but is included in the regional Red Books of Russia: in the Far East - in the Republic of Sakha (Yakutia), the Jewish Autonomous Region, Kamchatka Territory ^{4,6}.

S. sinensis has status 3, a rare species with a significant overall range, within which it occurs sporadically with small populations.

It is widespread in the All-Asian region (East and South Asia with penetration to Australia). In the Russian Far East, it grows in the Primorsky and Khabarovsk Territories, the

Amur Region, the Jewish Autonomous Region, Sakhalin, Kamchatka, and the Kuril Islands [1].

In the Primorsky Territory, the Chinese ladies'-tresses is found practically in all areas, but the known modern locations have low populations. It is found on the territories of the V.L. Komarov Ussurisky Nature Reserve, the Kedrovaya Pad (now «Land of the Leopard» National Park), and Sikhote-Alin Biosphere Reserve [8-10].

The purpose of the study is to describe new locations of *S. sinensis* in the Primorsky Territory.

MATERIAL AND METHODS

The field studies were carried out by the traditional route-reconnaissance method. Description of the identified habitats of *S. sinensis*, as well as the morphometric parameters of the plant were carried out. Analysis of herbarium specimens (VLA, VBGI, MW, MHA, LE) of Chinese ladies'-tresses in the Russian Far East was carried out.

RESULTS AND DISCUSSION

Over the period of 2018-2021, three new habitats were discovered and five *S. sinensis* cenopopulations were described in the Primorsky Territory (see Fig. 1).

Mikhailovsky district. The vicinity of the village Otradnoe, Ryabokon cl. (43°73'74" N, 132°47'87" E); 22.08.2019; mesophytic meadow. There were 25 individuals of *S. sinensis*, including 12 flowering plants, in the 936 m² plot. *Aralia elata* (Miq.) Seem., *Populus maximowiczii* A. Henry, *P. tremula* L., *Fraxinus mandshurica* Rupr. and a single stand of *Pinus koraiensis* Siebold et Zucc. are also observed on this territory. Associated plants of the herbaceous layer are *Kalimeris incisa* (Fisch.) DC, *Conyza canadensis* (L.) Cronq., *Gentiana zollingeri* Fawcett, *Ambrosia artemisiifolia* L., masses of *Ambrosia artemisiifolia* L.

⁴The Red Book of the Republic of Sakha (Yakutia). Rare and endangered species of plants and fungi. Ed. by N.S. Danilova. Moscow: Reart, 2017. Vol. 1. pp. 159.

⁵The Red Book of the Jewish Autonomous Region. Rare and threatened species of plants and fungi. Ed. by T.A. Rubtsova. Novosibirsk: ARTA, 2006. p. 106.

⁶The Red Book of the Kamchatka Territory. Plants. Ed. by O.A. Chernyagina. Petropavlovsk-Kamchatsky: Kamchatpress, 2018. Vol. 2. p. 54.

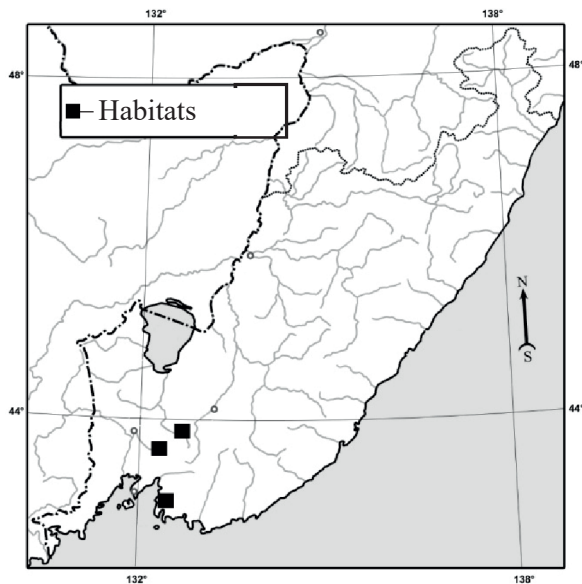


Рис. 1. Новые местообитания *S. sinensis* в Приморском крае

Fig. 1. New habitats of *S. sinensis* in the Primorsky Territory

Ussuriysk city district. The vicinity of the village Kaimananka (43°37'23" N, 132°13'50" E); 15.08.2010; 13.08.2019; agrocenosis, ecotone. Three cenopopulations of *S. sinensis* were found.

1. Absinthian grass meadow. There were 12 flowering specimens growing on an area of 25.5 m². Associated herbaceous plants – *Agrimonia striata* Michx., masses of – *Artemisia rubripes* Nakai, *A. stolonifera* (Maxim.) Kom., *A. scoparia* Waldst. et Kit., *Trifolium lupinaster* L., *T. repens* L., *Hypericum ascyron* L., *H. gebleri* Ledeb.

2. A cereal-grass meadow. There were 28 flowering specimens on an area of 16.5 m². Plant height did not exceed 35 cm and population density reached 5 specimens/m². Single small woody plants were observed on this area *Ulmus japonica* (Rehd.) Sarg. Associated herbaceous plants – *Asterager atoides* Turcz., *Miscanthus sacchariflorus* (Maxim.) Benth., *Phleum pratense* L., *Poa nemoralis* L., *P. ochotensis* Trin., *P. compressa* L., *Conyza canadensis* (L.) Cronq.

3. Absinthian grass-forb meadow. There were 20 flowering specimens growing on an area of 8 m². The plants reached a height of 50-

64 cm (see Fig. 2), with a leaf length of 17 cm and a width of 1.2 cm. Associated herbaceous plants – the above-mentioned plants from the second cenopopulation, as well as masses of *Ambrosia artemisiifolia* L., *Amphicarpaea japonica* (Oliv.) B. Fedtsch.

CATU Fokino (*Closed Administrative-Territorial Unit*). Near Fokino settlement (42°58'22" N, 132°24'31" E), 18.08.2018; area of the beach "Sandbox", a section of a sodded lawn experiencing intensive trampling. Grass stand is low, total projective cover is no more than 35%. There were 13 flowering specimens on an area of 26 m². Associated herbaceous plants – *Festuca extremiorientalis* Ohwi, *F. pratensis* Huds., *Seseli seseloides* (Turcz.) Hiroe, *Plantago depressa* Willd., *Carex arnellii* Christ, *C. bostrychostigma* Maxim., *Cyperus orthostachyus* Franch. et Savat., *Eriophorum polystachyon* L.

CONCLUSION

New habitats of the Chinese ladies'-tresses, which is relatively resistant to permanent trampling and is long-lived in herbage, were revealed in the Primorsky Territory. *S. sinensis* is very ornamental, has a long blooming period, and propagates vegetatively well. It is not listed



Рис. 2. *S. sinensis* в естественных условиях. Приморский край, окрестности с. Каймановка (43°37'23" N, 132°13'50" E):

a – вид соцветия; *б* – замеры растения

Fig. 2. *S. sinensis* in vivo. Primorsky Territory, Kaimanovka (43°37'23" N, 132°13'50" E):

a – type of inflorescence; *б* – measurements of the plant

in the Red Book of Endangered Species of the Russian Federation (2008) or in the Red Book of Endangered Species of the Primorsky Territory (2008), thus it may be used as an ornamental plant in the region. Like other meadow species of orchids, the Chinese ladies'-tresses can grow in sunny places and in the half-shade in combination with accompanying small plants.

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АМИЛОЛИТИЧЕСКАЯ АКТИВНОСТЬ ИЗОЛЯТОВ БАКТЕРИЙ *BACILLUS SUBTILIS*, ВЫДЕЛЕННЫХ ИЗ МИКРОБИОТЫ ДИКИХ ЖИВОТНЫХ*

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В настоящее время изучение ферментативного потенциала бактерий *Bacillus subtilis* является востребованным в силу того, что на их основе создаются пробиотические и ферментативные препараты для использования их в животноводстве. Представлены результаты исследования амилолитической активности 10 изолятов бактерий *B. subtilis*, выделенных из микробиоты диких животных, на амилопектин. Определение активности бактериальных ферментов производили методом Шомоди – Нельсона. В качестве ферментного препарата взяты супернатанты культуральной жидкости посевов 1, 2, 3 и 7-го дня. Изоляты с высокими качественными показателями исследовали количественно с использованием спектрофотометра. Отобраны два изолята с лучшими показателями. Концентрацию белков определяли методом Брэдфорда. Кроме изучения супернатантов изолятов, которые показали внеклеточную ферментативную активность, проведены исследования на внутриклеточные ферменты. Разрушение клеток производилось с помощью ультразвукового дезинтегратора, в котором растворенный осадок содержался в стеклянном флаконе, находящемся в колбе со льдом. Наилучшие качественные показатели выявлены у изолятов *B. subtilis* 2СП и *B. subtilis* 5СП, имеющих высокую активность по амилопектину начиная с первого дня, поэтому дальнейшие количественные исследования продолжены с ними. Концентрация белка у указанных изолятов прогрессировала по дням посевов. Удельная активность тоже соответственно увеличивалась и на 7-й день у изолята *B. subtilis* 2СП составила 2,75 ед./мг, у *B. subtilis* 5СП – 2,67 ед./мг. Изучение активности внутриклеточных ферментов у данных изолятов подтвердили аналогичные качественные показатели. По результатам исследований отобраны перспективные для разработки ферментных препаратов два изолята *B. subtilis*, выделенные из микробиоты диких животных Крайнего Севера.

Ключевые слова: дикие животные, микробиота, *Bacillus subtilis*, амилопектин, фермент, супернатант

AMYLOLYTIC ACTIVITY OF *BACILLUS SUBTILIS* ISOLATES OBTAINED FROM WILDLIFE MICROBIOTA*

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At present, the study of the enzymatic potential of *Bacillus subtilis* bacteria is in demand due to the fact that probiotic and enzymatic preparations are created on their basis for their use in animal husbandry. The results of the study of amylolytic activity of 10 isolates of *B. subtilis* bacteria iso-

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lated from wildlife microbiota for amylopectin are presented. Determination of bacterial enzyme activity was performed by the Schomodi-Nelson method. The supernatants of the culture liquid of the 1st, 2nd, 3rd, and 7th day cultures were taken as enzyme preparation. Isolates with high qualitative indices were examined quantitatively using a spectrophotometer. The two isolates with the best performance were selected. Protein concentration was determined by the Bradford method. In addition to studying supernatants of isolates that showed extracellular enzymatic activity, studies were performed on intracellular enzymes. Cell destruction was performed using an ultrasonic disintegrator, in which the dissolved sediment was contained in a glass vial in a flask with ice. The best qualitative indicators were found in *B. subtilis* 2SP and *B. subtilis* 5SP isolates, which had high amylopectin activity from day one, so further quantitative studies were continued with them. Protein concentrations in the above isolates progressed by the days of seeding. The specific activity also increased accordingly and on day 7, the *B. subtilis* 2SP isolate was 2.75 units/mg, *B. subtilis* 5SP 2.67 units/mg. The study of the activity of intracellular enzymes in these isolates confirmed similar qualitative indicators. Two *B. subtilis* isolates were selected as promising for the development of enzyme preparations isolated from the microbiota of wild animals of the Far North.

Keywords: wild animals, microbiota, *Bacillus subtilis*, amylopectin, enzyme, supernatant

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Автор заявляет об отсутствии конфликта интересов.

Conflict of interest

The author declares no conflict of interest.

INTRODUCTION

Bacillus subtilis is an aerobic gram-positive soil bacterium used for the production of heterologous proteins. It secretes numerous enzymes to degrade various substrates, which allows the bacteria to survive in the soil environment as well as in the plant rhizosphere [1]. This species and some of its closely related bacterial species have an excellent ability to secrete proteins, making them important producers of medicinal proteins and industrial enzymes. In addition, it has excellent physiological characteristics and a highly adaptable metabolism which makes it easy to cultivate on cheap substrates. *B. subtilis* grows rapidly and the fermentation cycle is short (usually about 48 h), whereas the fermentation cycle of *Saccharomyces cerevisiae* is about 180 h [2].

B. subtilis is used as a probiotic preparation in Russia and abroad [3]. The Yakut Scientific Research Institute of Agriculture (YaNIISKh)

developed the preparation "Sakhabaktisubtil" based on *B. subtilis* strains "TNP-3 DEP" and *B. subtilis* "TNP-5 DEP" isolated from the frozen soils of Yakutia. This preparation has a pronounced antagonistic effect against many pathogenic and opportunistic microorganisms, normalizes the intestinal microbiocenosis of animals, and exacerbates the immunobiological reactivity of the organism [4]. According to the literature, the microbiota of wild animals of the North is dominated by bacteria of the *Bacillus* genus [5-7].

Bacteria of the genus *Bacillus* are promising microorganisms that are widely used in animal husbandry. They are used to create feed additives in which *Bacillus* acts as a source of hydrolases. At present time to increase the nutritive value of feed, to increase its digestibility the enzyme preparations amylosubtilin, protosubtilin, monosporin and others are used. *Bacillus*-based enzyme preparations have a

positive effect on digestion and metabolism in farm birds and animals [8].

The industrial use of *B. subtilis* has developed rapidly in recent decades. It is one of the main microorganisms for the production of many industrial products. The chemicals produced by *B. subtilis* also play an important role in various fields such as food, feed additives, cosmetics, chemicals, and pharmaceuticals [9]. *B. subtilis* is classified as a Generally Recognized as Safe bacterium (GRAS), which can be used as a safe food product [10].

Deciphering the genomic determinants of starch hydrolysis based on computer modeling showed that amylolytic activity in Gram-positive bacteria depends on a complex transformation of the cell's genetic program. At the same time, it was reliably confirmed that the ability of a bacterium to break down starch depends not on the presence of the α -amylase gene in its genome but on the amylase activity [11].

The purpose of this work is to study the enzymatic activity of *B. subtilis* bacterial isolates isolated from wildlife microbiota for amylopectin.

MATERIAL AND METHODS

The studies were carried out in 2022 in the laboratory of the development of microbial preparations at YaNIISKh and in the laboratory of enzyme chemistry at the G.B. Elyakov Pacific Institute of Bioorganic Chemistry (PIBOC). Isolates of *B. subtilis* - 3, 4, 5, 8, 11, 16, 24, 25, 2sp and 5sp isolated from wild animal microbiota were taken from the working collection of the YaNIISKh microorganisms. Meat-peptone broth was used to inoculate the bacteria. It was incubated on a shaker in flasks with a cotton-gauze plug at room temperature to oxygenate it.

The Schomodi-Nelson method was used when conducting the studies to determine the activity of bacterial enzymes. Protein concentration was determined by the Bradford method.

The aliquots selected from the inoculum were centrifuged at 4500 rpm for 15 min. Mixtures of E + S (enzyme-substrate), E + H₂O (enzyme-control), S + H₂O (substrate-control), 100 and 200 μ l, respectively, were made from the obtained supernatants. The mixture of distilled

water with the supernatant shows the intensity of cleavage of its own sugars, with the substrate the possible cleavage of sugars without enzyme participation. The fermentation time in the thermostat at 37 °C was ~18 h. After fermentation, the Schomodi - Nelson method was used to determine the activity of bacterial enzymes by adding 300 μ l of Nelson A + B solution in a mixture of E + S, E + H₂O and S + H₂O. Next, they were placed in a thermostat at 99 °C for 15 min, added 300 μ l of the Nelson C solution and 500 μ l of distilled water. Qualitative parameters of the isolates were determined by the Nelson's method. High-value proteins were examined quantitatively using a spectrophotometer at a wavelength of 750 nm. Protein concentration was determined by the Bradford method at 595 nm. A solution of amylopectin in succinate buffer solution (pH 6.0) at a concentration of 1 mg/ml was used as a substrate. In addition to studying supernatants of the isolates, which showed extracellular enzymatic activity, studies were performed for intracellular enzymes. For this purpose, after centrifugation of a one-day culture, a precipitate was taken and dissolved in succinate buffer solution (pH 6.0). Destruction of cells was performed using an ultrasonic disintegrator in which the dissolved sediment was contained in a glass vial in a flask with ice. The disintegrator was switched off at 2 min intervals for 20 min.

RESULTS AND DISCUSSION

The amylolytic activity of supernatants of *B. subtilis* isolates on days 1, 2, 3, and 7 of amylopectin seeding was detected (see table). Control substrates were transparent, had no blue staining, which confirms the reliability of the results.

According to the results of our studies, the best qualitative parameters were found in *B. subtilis* 2SP and *B. subtilis* 5SP isolates. They had high amylopectin activity from the first day, so further quantitative studies were continued with them.

As shown in Figs. 1 and 2, the qualitative values of the isolates were confirmed by quantitative studies. Protein concentration of isolates progressed by days of seeding (see Fig. 1). The specific activity also increased accordingly and

was 2.75 units/mg for *B. subtilis* 2SP and 2.67 units/mg for *B. subtilis* 5SP at day 7 (see Fig. 2).

In intracellular enzyme studies, no enzyme-control activity was observed, so we can assume that isolates of *B. subtilis* 2SP and *B. subtilis* 5SP strains cleaved exactly the amylopectin substrate without impurities of their sugars. This method proved to be of little informative value, except for the fact that the isolates had no activity in the enzyme control, when the

qualitative Nelson values for amylopectin were comparable to those of the supernatants on the spectrophotometer.

CONCLUSION

Two promising isolates of *B. subtilis* were selected for further research and development of enzyme preparations *B. subtilis* 2SP and *B. subtilis* 5SP isolated from the microbiota of wild animals of the Far North with amylolytic activity of 2.75 and 2.67 units/mg, respectively.

Качественные показатели амилолитической активности супернатантов *B. subtilis* по дням посевов Qualitative indices of amylolytic activity of *B. subtilis* supernatants by days of planting

Isolate	Amylopectin	E + H ₂ O	Amylopectin	E + H ₂ O	Amylopectin	E + H ₂ O	Amylopectin	E+H ₂ O
	1-st day		2-nd day		3-rd day		7-th day	
11	-	-	-	-	+	-	±	±
25	-	-	-	-	±	-	±	±
3	-	-	-	-	±	-	±	±
5	-	-	-	-	±	-	±	±
16	-	-	±	-	++	-	±	±
2SP	+++	+	+++	+	+++	+	+++	+
24	-	-	-	-	±	-	±	±
8	-	-	-	-	+±	-	±	±
4	±	-	±	-	+±	-	±	±
5SP	+++	+	+++	+	+++	+	+++	+
Control-substrate	-	-	-	-	-	-	-	-

Note. «-» – no activity, «±» – little activity, «+» – average activity, «++» – low activity, «+++» – high activity.

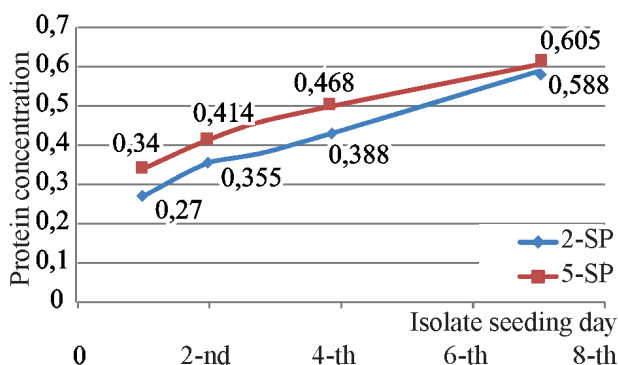


Рис. 1. Концентрация белка изолятов *B. subtilis* 2СП и *B. subtilis* 5СП по дням посевов, мг/мл

Fig. 1. Protein concentration of *B. subtilis* 2SP and *B. subtilis* 5SP isolates by days of planting, mg/ml

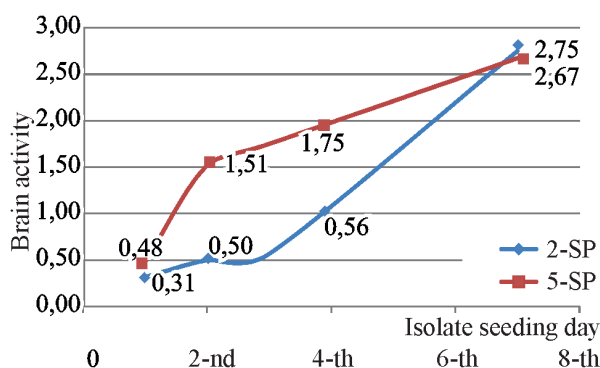


Рис. 2. Удельная активность лизатов на амилопектин по дням посевов, ед./мг белка

Fig. 2. Specific activity of lysates for amylopectin by days of planting, units/mg protein

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ИСПОЛЬЗОВАНИЕ ДНК-МАРКЕРОВ В СЕЛЕКЦИИ СОИ ДЛЯ ОТБОРА ФОТОПЕРИОДИЧЕСКИ НЕЙТРАЛЬНЫХ ЛИНИЙ

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Представлены результаты применения ДНК-маркеров в селекционном процессе сои для отбора фотопериодически нейтральных линий. Исследования проведены в предгорной зоне юго-востока Республики Казахстан на базе научно-полевого стационара в условиях 2020 г. Материалы исследования – 22 образца сои от ультраскороспелых (MG000) до средне-позд-неспелых (MGIII) групп спелости, использованных в качестве родительских форм для скрещиваний, а также 67 гибридных популяций из них (поколение F_2-F_4). ДНК-идентификация аллельной вариации генов *E1*, *E3*, *E7* проведена методом ПЦР с использованием SSR маркеров. По результатам ДНК-идентификации определены 10 образцов наиболее ценных родительских форм сои (носителей двух рецессивных аллелей в гомозиготном состоянии *ele7*) и 1 образец (сорт Малета), несущий три рецессивные аллели *ele3e7*. На основании маркер-ассоциированного отбора из 355 отдельных растений 67 гибридных популяций выявлены 9 растений, несущих рецессивные аллели *ele3e7*, из таких гибридных популяций, как Зара × Малета (номера гибридных популяций: 1, 2, 7, 15) и Бірлік × Rana (П-1), и 107 растений с рецессивными аллелями *ele7* из популяций: Зара × Малета (номера гибридных популяций: 1, 9, 7, 13), Бірлік × Hilario (П-10), Бірлік × Toury (П-6), Бірлік × Память ЮГК (П-5, M15/2, M15/3, M15/4 и M20), Бірлік × Припять (П-2), Ласточка × 234 (ЛТ44/11, ЛТ44/12). Данные образцы могут быть направлены для испытания в северные регионы Республики Казахстан как слабочувствительные к фотопериоду линии.

Ключевые слова: соя, ген чувствительности к фотопериоду, ДНК-маркер, маркер-ассоциированный отбор, селекция, линия

THE USE OF DNA MARKERS IN SOYBEAN BREEDING TO SELECT PHOTOPERIOD-NEUTRAL LINES

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The results of using DNA markers in the soybean breeding process to select photoperiod-neutral lines are presented. The studies were carried out in the foothill zone of the south-east of the Republic of Kazakhstan on the basis of the scientific field station of the Kazakh Research Institute of Agriculture and Plant Growing in the conditions of 2020. The material for the study was 22 samples of soybean from ultra-early ripening (MG000) to middle late ripening (MGIII) groups used as parental forms for crosses and their 67 hybrid populations (generation F_2-F_4). DNA identification of the allelic variation of the *E1*, *E3*, *E7* genes was carried out by PCR method using SSR markers. According to the results of DNA identification, ten samples of the most valuable parental forms of soybeans (carriers of two recessive alleles in the homozygous state *ele7*) and 1 sample (the Maleta variety)

carrying three recessive alleles *e1e3e7* were identified. Based on the marker-associated selection from 355 individual plants of 67 hybrid populations, 9 plants carrying valuable recessive alleles *e1e3e7* were isolated from such combinations as Zara x Maleta (hybrid population numbers: 1, 2, 7, 15) and Birlik x Rana (P-1) and 107 plants carrying valuable recessive *e1e7* alleles from such populations as: Zara x Maleta (hybrid population numbers: 1, 9, 7, 13), Birlik x Hilario (P-10), Birlik x Toury (P-6), Birlik x Memory YuGK (P-5, M15/2, M15/3, M15/4 and M20), Birlik x Pripyat (P-2), Lastochka x 234 (LT44/11, LT44/12). These samples can be sent for testing to the northern regions of the Republic of Kazakhstan as the lines that are weakly sensitive to the photoperiod.

Keywords: soybean, photoperiod sensitivity gene, DNA marker, marker-associated selection, selection, line

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The authors declare no conflict of interest.

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INTRODUCTION

Soybean (*Glycine max* (L.) Merr.) is the world's leading oilseed crop. Soybeans are a rich source of vegetable oil and protein. It is a multi-purpose crop for food, forage, medical and technical uses. Global soybean production was 353.4 million tons in 2020. The leading producer of soybeans in the world is the United States with 86.6 million tons, followed by Brazil (62.8 million tons) and Argentina (36.8 million tons)¹.

Soybean is a photoperiodic, highly sensitive, short-day crop [1]. Increasing the duration of daylight hours to 16–17 h has a significant impact on the growing season, plant height and productivity, significantly limiting the latitudi-

nal range of cultivation of certain soybean varieties² [2–6]. Plant breeders note that under long day conditions, photosensitive varieties bloom later, prolonging the growing season and fail to mature in time (before the autumn frosts). If the long day requirements of a variety differ very sharply from the conditions in which it is placed, the variety does not proceed to flowering and fruiting at all [7–9]. For this reason, soybean varieties, unlike cereal varieties, are confined to a narrow range of geographic latitudes. The annual expansion of the soybean growing range increases the urgency of creating varieties capable of producing high yields in conditions with limited thermal resources, to mature in more northern latitudes, where the

¹FAO data. <http://www.fao.org/faostat>

²Savelyev A.A. Methods of genotypes identification in the selection of soybean varieties with reduced photoperiodic sensitivity: Ph. D. thesis in Biology, 06.01.05. V.S. Pustovoit All-Russian Research Institute of Oil Crops. Krasnodar. 2009. 24 p. (date of reference: 01.05.2022).

duration of daylight hours between mid-May and the end of July exceeds 16 hours.

Molecular and genetic basis of soybean adaptation to different cultivation zones is provided by genes designated as *E*. Twelve major soybean genes are known to control flowering time, ripeness, and photoperiod sensitivity: *E1*, *E2*, *E3*, *E4*, *E5*, *E6*, *E7*, *E8*, *E9*, *E10*, *E11* and *J* [10]. Of the genes listed above, *E1*, *E2*, *E3*, *E4*, and *E7* have been described as quantitative photoperiodic genes [11, 12], whose recessive alleles lead to photoperiodic neutrality, while the dominant alleles delay the transition to the reproductive phase and the onset of ripeness. The wide adaptability of the crop is caused by different variations of allelic combinations of *E1*, *E2*, *E3*, *E4*, *E7* loci [13, 14]. The use of knowledge of the genetics of flowering, ripeness and sensitivity to photoperiod in the breeding process allowed the creation of soybean varieties that are currently cultivated in a variety of climatic zones [3, 15].

The basis for selection is the presence of donors and sources of early maturity and photoperiod neutrality in the soybean gene pool. Based on the phenotypic evaluation of the collection (2300 samples) in different regions of the Russian Federation, scientists from the N. I. Vavilov All-Russian Institute of Plant Genetic Resources (VIR) selected 340 ultra-ripening and early maturing varieties that reached full seed ripening in the northernmost point of experiments, the Leningrad region (59°44' N, 30°23' E). This is the most northern point of the world soybeans [8, 16]. The use of such varieties in the breeding process allows breeders to create domestic varieties of the northern ecotype.

In the Kazakh Research Institute of Agriculture and Crop Production (KazNIIZiR), located in the south-east of the Republic of Kazakhstan at 43°N, together with the agricultural research stations the breeding of ultra-ripening MG000 (maturity group) and fast-maturing MG00 soybean ripeness groups for the northern (53°N) and eastern (49°N) regions started in 2008 [17]. Currently, only one variety Ivushka (MG000) is approved for use in Pavlodar and Kostanay regions, and four soybean varieties (Zhalpak-say, Birlik KV, Vostochnaya krasavitsa, Alua)

of KazNIIZiR selection in the East Kazakhstan region. Selection for photoperiodic neutrality is complicated by the fact that phenotypic detection of the trait is possible only under long day conditions, as a result of testing in northern ecological points. The selection of the trait of photoperiod neutrality requires the use of DNA markers to track recessive alleles of the gene in splitting generations. In this regard, KazNIIZiR has begun work on the introduction of marker-associated selection in the selection process of soybean on the trait of sensitivity to the photoperiod to promote soybeans in the northern regions of the Republic of Kazakhstan.

The purpose of the research is to identify the allelic variation of *E1*, *E3*, *E7* genes in soybean varieties and lines used as parental forms and in individual plants in hybrid populations to select photoperiod insensitive lines.

MATERIAL AND METHODS

The material for the studies was 22 parental forms of soybean MG000 - MGIII (III) of ripeness groups and 67 hybrid populations (*F2-F4*) obtained using them.

For reliability of the study, DNA samples of soybean - carriers of recessive alleles of *E1*, *E7*, *E3* genes - as positive controls: Maple Amber (*e1*), Harosoy OT94-47 (*e3*), Harosoy (*e7*), as well as carriers of the dominant alleles as negative controls: Harosoy OT89-5 (*E7*), Swallow (*E1*), Harosoy (*E3*) were included in the experiment. Harosoy lines were provided by Belarusian scientist O.G. Davydenko (Soya-North LLC).

Research Methodology. Plants of the studied hybrid soybean populations *F2-F4* were grown in 2020 in the field breeding plot of KazNIIZiR located in the foothill zone of the South East of Kazakhstan (Almaty region) at 740 m above sea level (geographical location 43°15'N, 76°54'E). Sowing of 25 seeds at a depth of 4 cm at a plot of 1 linear meter. The replication is single. When making crossing schemes, flower corolla color of parental forms was considered as a marker trait: the paternal form had purple color, and the maternal form had white color. For cultivation and selection of elite hybrid plants the pedigree method was used in the soybean breeding pro-

cess. *F1* hybrid seeds were sown separately by combinations according to the scheme: ♀: *F1*: ♂. *F1* hybrids were rejected according to corolla flower color, removing plants with white flowers. Harvesting of *F1* plants was carried out strictly individually. The seeds collected from each plant, starting from *F2* generation, were sown separately into families (offspring from each plant). Phenological observations of growth and development of soybean plants were carried out during the growing season. At the end of the growing season the most productive plants were selected. The field rejection on the signs of cracking, lodging, propensity to diseases was carried out. The number of selected plants in different combinations of crosses depended on the percentage of *F1* hybridity and rejection of low-value forms.

The best hybrid populations of *F3-F4* generation were selected for DNA identification, except for *Zara* × *Major* and *Zara* × *Hilario* (*F2*). Five healthy plants were numbered and labeled from each hybrid soybean population. After reaching the phase of the 3rd pair of true leaves, DNA was extracted from each labeled plant (from leaves) using the CTAB method [18]. PCR analysis was performed in an Eppendorf Mastercycler amplifier (Germany). The following molecular markers were used: *Satt 100* and *Satt 319* to the *E7* gene [19]; *Satt 557*

[13] and *Satt 365* [19] to the *E1* gene; *Satt 229* [19] to the *E3* gene. The nucleotide sequence and PCR conditions are presented in Table 1.

The composition of the reaction mixture for PCR analysis was as follows: 100 ng (nanograms) of genomic DNA, 1 × PCR buffer, 2.5 mM MgCl₂, 200 μM of each dNTP, 0.5 μM of each primer, 0.5 units of Taq polymerase (Biosan LLC, Novosibirsk, Russia), BSA - 1 μg. Detection was performed by electrophoresis of amplification products in 8% polyacrylamide gel (Sigma Life Science, China). DNA marker *Step 100* (Biolabmix LLC, Novosibirsk, Russia) was used as a marker of molecular weights. Elite plants were selected based on the comparison of productivity, precocity, and PCR-analysis data.

RESULTS AND DISCUSSION

Identification of allelic variation in E1, E3, E7 genes in soybean varieties and lines used as parental forms in hybridization. To obtain soybean lines oriented to the northern regions of Kazakhstan, varieties and lines were selected from the working collection according to such important traits as early maturity and high productivity. In order to identify parental forms of soybean by allelic variation of genes sensitivity to photoperiod *E1, E3, E7* the PCR analysis was carried out. Using *Satt 557* and *Satt 365*

Табл. 1. Перечень маркеров и условия ПЦР, используемых для идентификации генов чувствительности к фотопериоду

Table 1. List of markers and PCR conditions used to identify photoperiod sensitivity genes

Gene	Molecular marker		
	Name	Nucleotide sequence	PCR conditions
<i>E1</i>	<i>Satt 557</i>	F: GCGGGATCCACCATGTAATATGTG R: GCGCACTAACCCCTTTATTGAA	95 °C – 5 min, 35 cycles (92 °C – 30 s, 53 °C – 30 s, 72 °C – 45 s), 72 °C – 7 min
	<i>Satt 365</i>	F: TGCTCCCCTCTGCCTTTTTTCTATTT R: AAGGATGAGTTTGATAAACATGAATGAAGAA	
<i>E7</i>	<i>Satt 100</i>	F: ACCTCATTTTGGCATAAA R: TTGGAAAACAAGTAATAATAACA	95 °C – 5 min, 35 cycles (92 °C – 45 s, 53 °C – 1 min, 72 °C – 80 s), 72 °C – 7 min
	<i>Satt 319</i>	F: CAACTCAGTAGGGGTCAATAACAA R: TGAAATAGGGAAAATAAGGGAACA	
<i>E3</i>	<i>Satt 229</i>	F: TGGCAGCACACCTGCTAAGGGAATAAA R: GCGAGGTGGTCTAAAATTATTACCTAT	95 °C – 5 min, 35 cycles (92 °C – 30 s, 53 °C – 45 s, 72 °C – 45 s), 72 °C – 7 min

markers to *E1* gene, the amplification of two fragments was recorded, which were identified as A and B alleles according to Molnar S. et al., 2003 [19]. As a result of DNA identification of 22 varieties and lines used as parental forms, 11 varieties (234, 470, 583583, Birlik KV, Ustyia, Khorol, Yaselda, Pripyat, Toury, Maleta) carrying recessive allele *E1* were identified (see Table 2).

Identification of allelic variation in the *E3* gene of parental forms allowed us to identify 2 specimens with the valuable recessive allele *e3* (cultivars Rana and Maleta) (see Table 2, Fig. 1).

In an experiment to determine the allelic variation of the *E7* gene, three fragments were amplified and identified as A, B, and C according to Rosenzweig et al. [20]. *E7* and *e7* alleles correspond to the designations A and B. An unknown allele with a 154 bp fragment corresponding to C was also detected in the *Satt 100* locus. Based on the identification, 11 samples (234, 470, 583583, Birlik KV, Ustyia, Khorol, Yaselda, Pripyat, Toury, Maleta, Jhony) carrying the recessive allele *e7* of photoperiod insensitivity were identified (see Table 2). Thus, 10 soybean parental forms carrying two recessive alleles of *e1e7* and the variety Maleta carrying all three recessive alleles of *e1e3e7* were selected from the chosen 22 parental forms.

Marker-associated selection for E1, E3, and E7 genes in hybrid soybean populations obtained from targeted crosses. Identification of E1 gene allelic variation. Based on the PCR-analysis of 335 individual plants of 67 hybrid populations, 126 plants (37.6%) were identified as carriers of the valuable recessive allele *e1*. They were identified from the following hybrid populations of F3-F4 generations: Birlik KV (*e1*) × Memory of UKG (*E1*), Birlik KV (*e1*) × Toury (*e1*), Birlik KV (*e1*) × Soer 345 (*E1*) (see Fig. 2), Lastochka (*e1*) × 234 (*e1*), Zara (*e1*) × Chorol (*e1*), Birlik SW (*e1*) × Pripyat (*e1*), Birlik SW (*e1*) × Gignon 5 (*E1*), Birlik EF (*e1*) × Hilario (*e1*), Zara (*e1*) × Maleta (*e1*), Zara (*e1*) × Soer 5 (*e1*), Zara (*e1*) × Major (*e1*) (see. Table 3). The highest percentage of selected plants with recessive allele *e1* was noted for combinations: Birlik KV (*e1*) × Hilario (*E1*)

(80%) and Birlik KV (*e1*) × Memory of UKG (*E1*) (77%). For such hybrid combinations as Zara × Chorol, Birlik KV × Toury, Birlik KV × Pripyat, where both parental forms are carriers of *e1* recessive allele, 100% occurrence of the sought *e1* allele among the studied plants was recorded (see Table 3).

Identification of the allelic variation of the E3 gene. Based on the PCR-analysis of 335 individual plants of 67 hybrid populations (5 individual plants each), 23 plants (6.9%) with the valuable allele *e3*, obtained from combinations of crosses Zara × Maleta (1, 2, 3 (see Fig. 3), 7, 10, 12, 15) and Birlik × Rana (P- 1) were identified and selected. From 23 selected plants of Zara × Maleta hybrid populations, 22 specimens (95.7%) were identified.

Identification of the E7 gene. PCR analysis to identify allelic variation of *E7* gene in 335 individual plants of 67 hybrid populations allowed to identify 135 individual plants (40.3%) carrying recessive allele *e7* from the following hybrid combinations: Birlik × Rana (P-1), Birlik × Pripyat (P-2), Birlik × Gignon 5 (P-3) (see Fig. 4), Birlik × Hilario (P-10), Birlik × Toury (P-6), Birlik × Memory of UKG (P-5, M 15/2, M 15/3, M 15/4, M 15/5, M 20), Zara × Khorol (H10/1, H10-2), Zara × Ustyia (L4/34), Zara × 470 (L11/4), Zara × Bara (P-12), Zara × Jhony (P-13), Zara × Maleta (P-19, 1, 2, 5, 6, 7, 8, 9, 13, 15), Lastochka × 234 (LT44/11, LT44/12). The highest percentage (80%) of selected plants with recessive allele *e7* was observed in the combinations: Birlik KV (*e7*) × Memory of UKG (*e7*), Birlik (*e7*) × Hilario (*e7*), Zara (*e7*) × Ustyia (*e7*), Zara (*e7*) × 470 (*e7*).

According to the results of the research, 7 out of 355 studied plants were identified that have all three valuable recessive alleles of *e1e3e7* in the genotype in the homozygous state. Testing of these samples will be continued in order to obtain constant photoperiod-neutral lines for the northern regions of the Republic of Kazakhstan.

CONCLUSION

The allelic variation of *E1*, *E3*, *E7* genes in 22 parental forms of soybean used in the cre-

Табл. 2. Результаты идентификации генов чувствительности к фотопериоду (*E1*, *E7*, *E3*) у образцов сои, используемых в качестве родительских форм при гибридизации

Table 2. Results of identification of photoperiod sensitivity genes (*E1*, *E7*, *E3*) in soybean samples used as parental forms in hybridization

Sample	Origin	Ma-turity group	Seed parent	<i>Satt</i> 557	<i>Satt</i> 365	Gene allele <i>E1</i>	<i>Satt</i> 229	Gene allele <i>E3</i>	<i>Satt</i> 100	<i>Satt</i> 319	Gene allele <i>E7</i>
<i>Control lines</i>											
Harosoy OT89-5 <i>E7</i>	Japan	00	–	<i>B</i>	<i>B</i>	<i>e1</i>	<i>B</i>	<i>e3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Harosoy OT 94-47	Japan	00	–	<i>B</i>	<i>B</i>	<i>e1</i>	<i>B</i>	<i>e3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Harosoy	Japan	0	–	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Maple Amber	Canada	000	–	<i>B</i>	<i>B</i>	<i>e1</i>	<i>B</i>	<i>e3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Lastochka	Kazakhstan	III	♀	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
<i>Seed parents</i>											
470	Not known	00	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>C</i>	–	<i>B</i>	<i>B</i>	<i>e7</i>
583583	Not known	00	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>C</i>	–	<i>B</i>	<i>B</i>	<i>e7</i>
Birlik KV	Kazakhstan	00	♀	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Zara	Kazakhstan	I	♀	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Soer 3	Russia	00	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>C</i>	–	<i>A</i>	<i>A</i>	<i>E7</i>
Soer 5	Russia	0	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Soer 345	Russia	0	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>C</i>	–	<i>A</i>	<i>A</i>	<i>E7</i>
Bara	Russia	000	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>C</i>	<i>A</i>	<i>E7</i>
Maleta	Russia	000	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>B</i>	<i>e3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
234	Russia	000	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>C</i>	–	<i>B</i>	<i>B</i>	<i>e7</i>
Ustyа	Ukraine	00	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Khorol	Canada – Ukraine	0	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Yaselda	Belarus	00	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Прыпят	Belarus	00	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Toury	Czech Republic	0	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Rana	Czech Republic	00	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>B</i>	<i>e3</i>	<i>C</i>	<i>A</i>	<i>E7</i>
Hilario	Italy	III	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Jhony	Not known	00	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>B</i>	<i>B</i>	<i>e7</i>
Major	France	0	♂	<i>B</i>	<i>B</i>	<i>e1</i>	<i>C</i>	–	<i>A</i>	<i>A</i>	<i>E7</i>
Gignon 5	France	I	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>
Memory YuGK	Kazakhstan	I	♂	<i>A</i>	<i>A</i>	<i>E1</i>	<i>A</i>	<i>E3</i>	<i>A</i>	<i>A</i>	<i>E7</i>



Рис. 1. Результаты ПЦР по идентификации аллельной вариации гена *E3* у сортов и линий сои, используемых в качестве родительских форм с использованием маркера *Satt* 229

M – маркер *Step* 100, *E3* – Harosoy (*E3*), *e3* – Harosoy OT94-47 (*e3*), 1 – Zara, 2 – Бирлік КВ, 3 – Rana, 4 – Припят, 5 – Toury, 6 – Соер 345, 7 – Hilario, 8 – Бара, 9 – Jhony, 10 – Соер 5, 11 – Major, 12 – Соер 3, 13 – Маleta, 14 – Устя, 15 – Gignon 5, 16 – Память ЮГК

Fig. 1. PCR results for the identification of the allelic variation of the *E3* gene in soybean varieties and lines used as parental forms using the marker *Satt* 229

M - *Step* 100 marker, *E3* - Harosoy (*E3*), *e3* - Harosoy OT94-47 (*e3*), 1 - Zara, 2 - Birlik KV, 3 - Rana, 4 - Pripyat, 5 - Toury, 6 - Soer 345, 7 - Hilario, 8 - Bara, 9 - Jhony, 10 - Soer 5, 11 - Major, 12 - Soer 3, 13 - Maleta, 14 - Ustyа, 15 - Gignon 5, 16 - Memory YuGK

Табл. 3. Результаты ДНК-идентификации гибридных популяций сои
Table 3. Results of DNA identification of hybrid soybean populations

Name of the crossing combination	Generation	Number of hybrid populations studied, pcs.	Number of plants analyzed, pcs.	Presence of recessive allele <i>e1</i>		Presence of recessive allele <i>e3</i>		Presence of recessive allele <i>e7</i>	
				Name of hybrid populations in which allele <i>e1</i> carriers were selected	Number of selected plants, pcs. (%)	Name of hybrid populations in which allele <i>e1</i> carriers were selected	Number of selected plants, pcs. (%)	Name of hybrid populations in which allele <i>e1</i> carriers were selected	Number of selected plants, pcs. (%)
Lastochka × 234	F ₃	3	15	LT44/11, LT44/12	10 (66,7%)	—	—	LT44/11, LT44/12	10 (66,7%)
Zara × Yaselda	F ₃	1	5	—	—	—	—	—	—
Zara × Khorol	F ₃	2	10	H 10/1, H 10/2	10 (100%)	—	—	H 10/1, H 10/2	7 (70%)
Zara × 234	F ₃	2	10	—	—	—	—	—	—
Birlık KV × Memory YuGK	F ₃	6	30	M 15/2, M 15/3, M 15/4, M 15/5, M 20	23 (77%)	—	—	M 15/2, M 15/3, M 15/4, M 15/5, M 20	22 (73%)
Zara × 583583	F ₃	4	20	L1/1	2 (10%)	—	—	—	—
Zara × Ustya	F ₃	5	25	L4/34	1 (4%)	—	—	L4/34	4 (16%)
Zara × 470	F ₃	11	55	—	—	—	—	L 11/4	5 (9%)
Birlık KV × Rana	F ₃	1	5	P-1	3 (60%)	P-1	1 (20%)	P-1	3 (60%)
Birlık × Pripyat	F ₃	1	5	P-2	5 (100%)	—	—	P-2	5 (100%)
Birlık × Gignon	F ₄	1	5	P-3	2 (40%)	—	—	P-3	1 (20%)
Birlık KV × Memory YuGK	F ₄	1	5	P-5	4 (80%)	—	—	P-5	4 (80%)
Birlık × Toury	F ₃	1	5	P-6	5 (100%)	—	—	P-6	5 (100%)
Бирік × Соєр 345	F ₃	1	5	P-9	1 (20%)	—	—	P-9	1 (20%)
Birlık × Hilario	F ₃	1	5	P-10	4 (80%)	—	—	P-10	4 (80%)
Zara × Bara	F ₃	1	5	—	—	—	—	P-12	4 (80%)
Zara × Jhony	F ₃	1	5	—	—	—	—	P-13	4 (80%)
Zara × Major	F ₂	1	5	P-14	3 (60%)	—	—	P-14	1 (20%)
Zara × Hilario	F ₂	1	5	—	—	—	—	—	—
Zara × Soer 3	F ₃	1	5	—	—	—	—	—	—
Zara × Maleta	F ₃	16	80	P-19, 1, 2, 5, 6, 7, 8, 9, 13, 15	42 (52,5%)	1, 2, 3, 7, 10, 12, 15	22 (27,5%)	P-19, 1, 2, 5, 6, 7, 8, 9, 13, 15	42 (53%)
Zara × Ustya	F ₃	1	5	—	—	—	—	P-20	1 (20%)
Zara × Soer 5	F ₃	4	20	1, 2, 4	11 (55%)	—	—	1, 2, 4	13 (65%)
Total:		67	335		126 (37,6%)		23 (6,9%)		135 (40,6%)

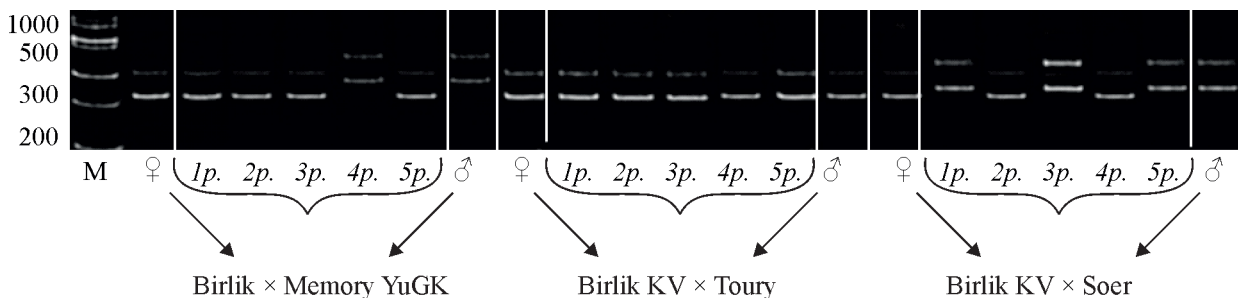


Рис. 2. Результаты ПЦР по идентификации аллельной вариации гена *E1* в гибридных популяциях сои с использованием маркера *Satt 365*

Fig. 2. PCR results for identification of the allelic variation of the *E1* gene in hybrid soybean populations using the *Satt 365* marker

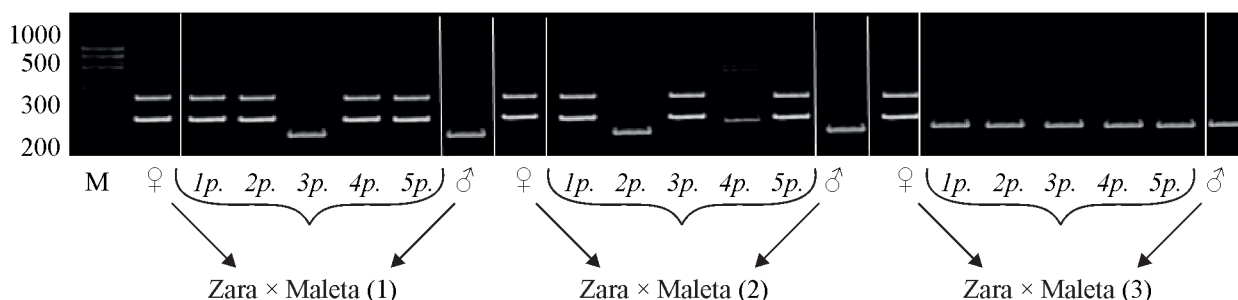


Рис. 3. Результаты ПЦР по идентификации аллельной вариации гена *E3* в гибридных комбинациях Зара x Малета с использованием маркера *Satt 229*

Fig. 3. Results of PCR for identification of the allelic variation of the *E3* gene in hybrid combinations of Zara x Maleta using the *Satt 229* marker

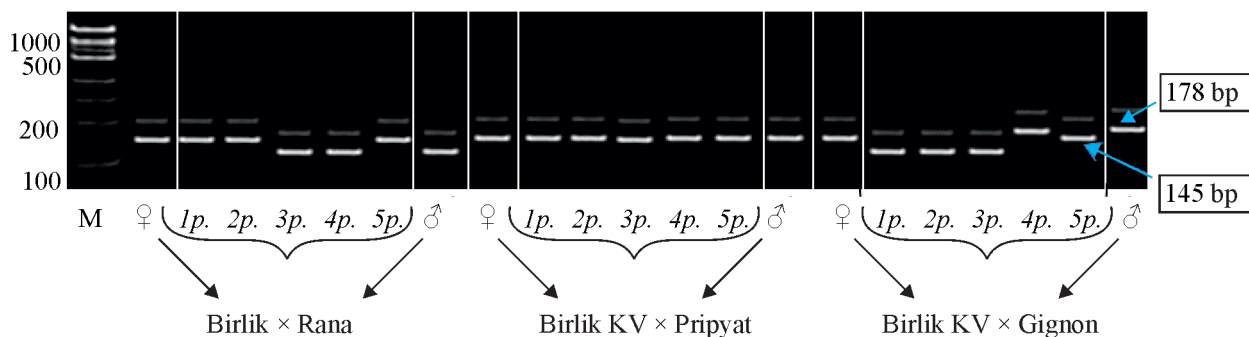


Рис. 4. Результаты ПЦР по идентификации аллельной вариации гена *E7* в гибридных популяциях с использованием маркера *Satt 100* (178 п.н. аллель *E7*, 145 п.н. аллель *e7*)

Fig. 4. PCR results for the identification of the allelic variation of the *E7* gene in hybrid populations using the *Satt 100* marker (178 bp *E7* allele, 145 bp *e7* allele)

ation of early maturing lines for the northern regions of the Republic of Kazakhstan was identified. Ten forms carrying two recessive alleles of *Ele7* and Maleta variety carrying all three recessive alleles of *Ele3e7* were identified. The results of the study of 355 plants from 67 hybrid populations (*F2-F4*), based on marker-associated selection from combinations of Zara x Male-

ta (1, 2, 7, 15) and Birlik x Rana (P-1) showed that 9 plants carrying the complex of valuable recessive alleles of *ele3e7* were identified. 107 plants carrying valuable recessive *ele7* alleles were identified from the combinations: Zara x Maleta (1, 9, 7, 13), Birlik x Hilario (P-10), Birlik x Toury (P-6), Birlik x Memory of UGK (P-5, M15/2, M15/3, M15/4 and M20), Birlik x

Pripyat (P-2), and Lastochka × 234 (LT44/11, LT44/12). These specimens are recommended for testing in the northern regions of the Republic of Kazakhstan as poorly sensitive to photoperiod lines.

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ПРАВИЛА ДЛЯ АВТОРОВ

Правила для авторов составлены на основе этических принципов, общих для членов научного сообщества, и правил публикации в международных и отечественных научных периодических изданиях, а также в соответствии с требованиями ВАК для периодических изданий, включенных в Перечень российских рецензируемых научных журналов, в которых должны быть опубликованы основные научные результаты диссертаций на соискание ученой степени доктора и кандидата наук.

Журнал публикует оригинальные статьи по фундаментальным и прикладным проблемам по направлениям:

- общее земледелие и растениеводство;
- селекция, семеноводство и биотехнология растений;
- агрохимия, агропочвоведение, защита и карантин растений;
- кормопроизводство;
- инфекционные болезни и иммунология животных;
- частная зоотехния, кормление, технологии приготовления кормов и производства продукции животноводства;
- разведение, селекция, генетика и биотехнология животных;
- технологии, машины и оборудование для агропромышленного комплекса;
- пищевые системы.

Статья, направляемая в редакцию, должна соответствовать тематическим разделам журнала «Сибирский вестник сельскохозяйственной науки»:

Наименование рубрики	Шифр и наименование научной специальности в соответствии с Номенклатурой научных специальностей, по которым присуждаются ученые степени
Земледелие и химизация	4.1.1. Общее земледелие и растениеводство
	4.1.3. Агрохимия, агропочвоведение, защита и карантин растений
Растениеводство и селекция	4.1.1. Общее земледелие и растениеводство
	4.1.2. Селекция, семеноводство и биотехнология растений
Защита растений	4.1.3. Агрохимия, агропочвоведение, защита и карантин растений
Кормопроизводство	4.1.1. Общее земледелие и растениеводство
	4.1.2. Селекция, семеноводство и биотехнология растений
	4.1.3. Агрохимия, агропочвоведение, защита и карантин растений
Зоотехния и ветеринария	4.2.3. Инфекционные болезни и иммунология животных
	4.2.4. Частная зоотехния, кормление, технологии приготовления кормов и производства продукции животноводства
	4.2.5. Разведение, селекция, генетика и биотехнология животных
Механизация, автоматизация, моделирование и информационное обеспечение	4.3.1. Технологии, машины и оборудование для агропромышленного комплекса
Переработка сельскохозяйственной продукции	4.3.3. Пищевые системы
Проблемы. Суждения Научные связи Из истории сельскохозяйственной науки Краткие сообщения Из диссертационных работ	4.1.1. Общее земледелие и растениеводство
	4.1.2. Селекция, семеноводство и биотехнология растений
	4.1.3. Агрохимия, агропочвоведение, защита и карантин растений
	4.2.3. Инфекционные болезни и иммунология животных
	4.2.4. Частная зоотехния, кормление, технологии приготовления кормов и производства продукции животноводства
	4.2.5. Разведение, селекция, генетика и биотехнология животных
	4.3.1. Технологии, машины и оборудование для агропромышленного комплекса
4.3.3. Пищевые системы	

В журнале также публикуются обзоры, краткие сообщения, хроника, рецензии, книжные обозрения, материалы по истории сельскохозяйственной науки и деятельности учреждений и ученых.

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К рассмотрению принимаются материалы от различных категорий исследователей, аспирантов, докторантов, специалистов и экспертов в соответствующих областях знаний.

Все статьи рецензируются и имеют зарегистрированный в системе CrossRef индекс DOI.

Публикации для авторов **бесплатны**.

При направлении статьи в редакцию журнала «Сибирский вестник сельскохозяйственной науки» рекомендуем руководствоваться следующими правилами.

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- статья ранее не была опубликована в другом журнале;
- статья не находится на рассмотрении в другом журнале;
- все соавторы согласны с публикацией текущей версии статьи.

Перед отправкой статьи на рассмотрение необходимо убедиться, что в файле (файлах) содержится вся необходимая информация на русском и английском языках, указаны источники информации, размещенной на рисунках и в таблицах, все ссылки оформлены корректно.

ПОРЯДОК НАПРАВЛЕНИЯ РУКОПИСЕЙ СТАТЕЙ

1. Отправка статьи осуществляется через электронную редакцию на сайте журнала <https://sibvest.elpub.ru/jour/index>. После предварительной регистрации автора, в правом верхнем углу страницы выбрать опцию «Отправить рукопись». Затем загрузить рукопись статьи (в формате *.doc или *.docx) и сопроводительные документы к ней. После завершения загрузки материалов обязательно выбрать опцию «Отправить письмо», в этом случае редакция автоматически будет уведомлена о получении новой рукописи.

Сопроводительные документы к рукописи статьи:

- скан-копия письма от организации с подтверждением авторства и разрешением на публикацию (образец на <http://sibvest.elpub.ru/>);
- скан-копия авторской справки по представленной форме (образец на <http://sibvest.elpub.ru/>), в которой должно быть выражено согласие на открытое опубликование статьи в печатном варианте журнала и его электронной копии в сети Интернет;
- скан-копия рукописи с подписями авторов. Автор, подписывая рукопись и направляя ее в редакцию, тем самым передает авторские права на издание этой статьи СФНЦА РАН;
- анкеты авторов на русском и английском языках (образец на <http://sibvest.elpub.ru/>);
- скан-копия справки из аспирантуры (для очных аспирантов).

2. Все поступающие в редакцию рукописи статей регистрируются через систему электронной редакции. В личном кабинете автора отражается текущий статус рукописи.

3. Нерецензируемые материалы (материалы научной хроники, рецензии, книжные обозрения, материалы по истории сельскохозяйственной науки и деятельности учреждений и ученых) направляются на e-mail: sibvestnik@sfcsa.ru и регистрируются ответственным секретарем.

ПОРЯДОК ОФОРМЛЕНИЯ СТАТЬИ

Текст рукописи оформляется шрифтом Times New Roman, кеглем 14 с интервалом 1,5, все поля 2,0 см, нумерация страниц внизу. Объем статьи не более 15 страниц (включая таблицы, иллюстрации и библиографию); статей, размещаемых в рубриках «Из диссертационных работ» и «Краткие сообщения», – не более 7 страниц.

Структура оформления статьи:

1. **УДК**
2. **Заголовок статьи на русском и английском языках (не более 70 знаков).**
3. **Фамилии и инициалы авторов, полное официальное название научного учреждения, в котором проведены исследования на русском и английском языках.**

Если в подготовке статьи принимали участие авторы из разных учреждений, необходимо указать принадлежность каждого автора к конкретному учреждению с помощью надстрочного индекса.

4. **Реферат на русском и английском языках.** Объем реферата не менее 200–250 слов. Реферат является кратким и последовательным изложением материала статьи по основным разделам и должен отражать основное содержание, следовать логике изложения материала и описания результатов в статье с приведением конкретных данных. Не следует включать впервые введенные термины, аббревиатуры (за исключением общеизвестных), ссылки на литературу. В реферате не следует подчеркивать новизну, актуальность и личный вклад автора; место исследования необходимо указывать до области (края), не упоминать конкретные организации.

5. **Ключевые слова на русском и английском языках.** 5–7 слов по теме статьи. Желательно, чтобы ключевые слова дополняли реферат и название статьи.

6. **Информация о конфликте интересов либо его отсутствии.** Автор обязан уведомить редактора о реальном или потенциальном конфликте интересов, включив информацию о конфликте интересов в соответствующий раздел статьи. Если конфликта интересов нет, автор должен также сообщить об этом.

Пример формулировки: «Автор заявляет об отсутствии конфликта интересов».

7. **Благодарности на русском и английском языках.** В этом разделе указываются все источники финансирования исследования, а также благодарности людям, которые участвовали в работе над статьей, но не являются ее авторами.

8. **Основной текст статьи.** При изложении оригинальных экспериментальных данных рекомендуется использовать подзаголовки:

ВВЕДЕНИЕ (постановка проблемы, цели, задачи исследования)

МАТЕРИАЛ И МЕТОДЫ (условия, методы (методика) исследований, описание объекта, место и время проведения)

РЕЗУЛЬТАТЫ И ОБСУЖДЕНИЕ

ЗАКЛЮЧЕНИЕ или **ВЫВОДЫ**

СПИСОК ЛИТЕРАТУРЫ. Количество источников не менее 15. В список литературы включаются только рецензируемые источники: статьи из научных журналов и монографии. Самоцитирование не более 10% от общего количества. Библиографический список должен быть оформлен в виде общего списка в порядке упоминания в тексте, желательны ссылки на источники 2–3-летнего срока давности. Правила оформления списка литературы – в соответствии с ГОСТ Р 7.05–2008 (требования и правила составления библиографической ссылки). В тексте ссылка на источник отмечается порядковой цифрой в квадратных скобках, например [1]. Литература в списке дается на тех языках, на которых она издана. В библиографическое описание публикации необходимо вносить всех авторов, не сокращая их одним, тремя и т.п. Недопустимо сокращение названий статей, журналов, издательств.

Если необходимо сослаться на авторефераты, диссертации, сборники статей, учебники, рекомендации, учебные пособия, ГОСТы, информацию с сайтов, статистические отчеты, статьи в общественно-политических газетах и прочее, то такую информацию следует оформить в *сноске* в конце страницы. Сноски нумеруются арабскими цифрами, размещаются постранично сквозной нумерацией.

Внимание! Теоретические, обзорные и проблемные статьи могут иметь произвольную структуру, но обязательно должны содержать реферат, ключевые слова, список литературы.

ПРИМЕРЫ ОФОРМЛЕНИЯ СПИСКА ЛИТЕРАТУРЫ, REFERENCES И СНОСК

СПИСОК ЛИТЕРАТУРЫ:

Монография

Климова Э.В. Полевые культуры Забайкалья: монография. Чита: Поиск, 2001. 392 с.

Часть книги

Холмов В.Г. Минимальная обработка кулисного пара под яровую пшеницу при интенсификации земледелия в южной лесостепи Западной Сибири // Ресурсосберегающие системы обработки почвы. М.: Агропромиздат, 1990. С. 230–235.

Периодическое издание

Пакуль А.Л., Лапишинов Н.А., Божанова Г.В., Пакуль В.Н. Технологические качества зерна мягкой яровой пшеницы в зависимости от системы обработки почвы // Сибирский вестник сельскохозяйственной науки. 2018. Т. 48. № 4. С. 27–35. DOI: 10.26898/0370-8799-2018-4-4.

REFERENCES:

Составляется в том же порядке, что и русскоязычный вариант, по следующим правилам:

Фамилии И.О. авторов в устоявшемся способе транслитерации, англоязычное название статьи, *транслитерация названия русскоязычного источника (например через сайт: <https://antrophob.ru/translit-bsi>) = англоязычное название источника*. Далее оформление для монографии: город, англоязычное название издательства, год, количество страниц; для журнала: год, номер, страницы). (In Russian).

Пример: Avtor A.A., Avtor B.B., Avtor C.C. Title of article.

Транслитерация авторов. Англоязычное название статьи

Zaglavie jurnala = Title of Journal, 2012, vol. 10, no. 2, pp. 49–54.

Транслитерация источника = Англоязычное название источника

Монография

Klimova E.V. *Field crops of Zabaikalya*. Chita, Poisk Publ., 2001, 392 p. (In Russian).

Часть книги

Kholmov V.G. Minimum tillage of coulisse-strip fallow for spring wheat with intensification of arable agriculture in southern forest-steppe of Western Siberia. *Resource-saving tillage systems*, Moscow, Agropromizdat Publ., 1990, pp. 230–235. (In Russian).

Периодическое издание

Pakul A.L., Lapshinov N.A., Bozhanova G.V., Pakul V.N. Technological grain qualities of spring common wheat depending on the system of soil tillage. *Sibirskii vestnik sel'skokhozyaistvennoi nauki = Siberian Herald of Agricultural Science*, 2018, vol. 48, no. 4, pp. 27–35. (In Russian). DOI: 10.26898/0370-8799-2018-4-4.

СНОСКИ:

Цитируемый текст¹.

¹Климова Э.В., Андреева О.Т., Темникова Г.П. Пути стабилизации кормопроизводства Забайкалья // Проблемы и перспективы совершенствования зональных систем земледелия в современных условиях: материалы науч.-практ. конф. (Чита, 16–17 октября 2008 г.). Чита, 2009. С. 36–39.

Цифровой идентификатор Digital Object Identifier – DOI (когда он есть у цитируемого материала) необходимо указывать в конце библиографической ссылки.

Пример:

Chu T., Starek M.J., Brewer M.J., Murray S.C., Pruter L.S. Assessing lodging severity over an experimental maize (*Zea mays* L.) field using UAS images // *Remote Sensing*. 2017. Vol. 9. P. 923. DOI: 10.3390/rs9090923.

Наличие DOI статьи следует проверять на сайте <http://search.crossref.org/> или <https://www.citethisforme.com>.

Для этого нужно ввести в поисковую строку название статьи на английском языке.

РИСУНКИ, ТАБЛИЦЫ, СКРИНШОТЫ И ФОТОГРАФИИ

Рисунки должны быть хорошего качества, пригодные для печати. Все рисунки должны иметь подрисуночные подписи. Подрисуночную подпись необходимо перевести на английский язык. Рисунки нумеруются арабскими цифрами по порядку следования в тексте. Если рисунок в тексте один, то он не нумеруется. Отсылки на рисунки оформляются следующим образом: «На рис. 3 указано, что ...» или «Указано, что ... (см. рис. 3)». Подрисуночная

подпись включает порядковый номер рисунка и его название. «Рис. 2. Описание жизненно важных процессов». Перевод подрисуночной подписи следует располагать после подрисуночной подписи на русском языке.

Таблицы должны быть хорошего качества, пригодные для печати. Предпочтительны таблицы, пригодные для редактирования, а не отсканированные или в виде рисунков. Все таблицы должны иметь заголовки. Название таблицы должно быть переведено на английский язык. Таблицы нумеруются арабскими цифрами по порядку следования в тексте. Если таблица в тексте одна, то она не нумеруется. Отсылки на таблицы оформляются следующим образом: «В табл. 3 указано, что ...» или «Указано, что ... (см. табл. 3)». Заголовок таблицы включает порядковый номер таблицы и ее название: «Табл. 2. Описание жизненно важных процессов». Перевод заголовка таблицы следует располагать после заголовка таблицы на русском языке.

Фотографии, скриншоты и другие нерисованные иллюстрации необходимо загружать отдельно в виде файлов формата *.jpeg (*.doc и *.docx – в случае, если на изображение нанесены дополнительные пометки). Разрешение изображения должно быть >300 dpi. Файлам изображений необходимо присвоить название, соответствующее номеру рисунка в тексте. В описании файла следует отдельно привести подрисуночную подпись, которая должна соответствовать названию фотографии, помещаемой в текст.

Следует обратить внимание на написание формул в статье. Во избежание путаницы необходимо греческие (α , β , π и др.), русские (А, а, Б, б и др.) буквы и цифры писать прямым шрифтом, латинские – курсивным (*W*, *Z*, *m*, *n* и др.). Математические знаки и символы нужно писать также прямым шрифтом. Необходимо четко указывать верхние и нижние надстрочные символы (W^1 , F_1 и др.).

ВЗАИМОДЕЙСТВИЕ МЕЖДУ ЖУРНАЛОМ И АВТОРОМ

Редакция просит авторов при подготовке статей руководствоваться изложенными выше правилами.

Все поступающие в журнал «Сибирский вестник сельскохозяйственной науки» статьи проходят предварительную проверку на соответствие формальным требованиям. На этом этапе редакция оставляет за собой право:

- принять статью к рассмотрению;
 - вернуть статью автору (авторам) на доработку с просьбой устранить ошибки или добавить недостающие данные;
 - вернуть статью автору (авторам) без рассмотрения, оформленную не по требованиям журнала;
 - отклонить статью из-за несоответствия ее целям журнала, отсутствия оригинальности, малой научной ценности.
- Переписка с авторами рукописи ведется через контактное лицо, указанное в рукописи.

Все научные статьи, поступившие в редакцию журнала «Сибирский вестник сельскохозяйственной науки», проходят обязательное двухстороннее «слепое» рецензирование (double-blind – автор и рецензент не знают друг о друге). Рукописи направляются по профилю научного исследования на рецензию членам редакционной коллегии.

В спорных случаях редактор может привлечь к процессу рецензирования нескольких специалистов, а также главного редактора. При положительном заключении рецензента статья передается редактору для подготовки к печати.

При принятии решения о доработке статьи замечания и комментарии рецензента передаются автору. Автору дается 2 месяца на устранения замечаний. Если в течение этого срока автор не уведомил редакцию о планируемых действиях, статья снимается с очереди публикации.

При принятии решения об отказе в публикации статьи автору отправляется соответствующее решение редакции.

Ответственному (контактному) автору принятой к публикации статьи направляется финальная версия верстки, которую он обязан проверить.

ПОРЯДОК ПЕРЕСМОТРА РЕШЕНИЙ РЕДАКТОРА/РЕЦЕНЗЕНТА

Если автор не согласен с заключением рецензента и/или редактора или отдельными замечаниями, он может оспорить принятое решение. Для этого автору необходимо:

- исправить рукопись статьи согласно обоснованным комментариям рецензентов и редакторов;
- ясно изложить свою позицию по рассматриваемому вопросу.

Редакторы содействуют повторной подаче рукописей, которые потенциально могли бы быть приняты, однако были отклонены из-за необходимости внесения существенных изменений или сбора дополнительных данных, и готовы подробно объяснить, что требуется исправить в рукописи для того, чтобы она была принята к публикации.

ДЕЙСТВИЯ РЕДАКЦИИ В СЛУЧАЕ ОБНАРУЖЕНИЯ ПЛАГИАТА, ФАБРИКАЦИИ ИЛИ ФАЛЬСИФИКАЦИИ ДАННЫХ

Редакция научного журнала «Сибирский вестник сельскохозяйственной науки» в своей работе руководствуется традиционными этическими принципами научной периодики и сводом принципов «Кодекса этики научных публикаций», разработанным и утвержденным Комитетом по этике научных публикаций, требуя соблюдения этих правил от всех участников издательского процесса.

ИСПРАВЛЕНИЕ ОШИБОК И ОТЗЫВ СТАТЬИ

В случае обнаружения в тексте статьи ошибок, влияющих на ее восприятие, но не искажающих изложенные результаты исследования, они могут быть исправлены путем замены pdf-файла статьи. В случае обнаружения в тексте статьи ошибок, искажающих результаты исследования, либо в случае плагиата, обнаружения недобросовестного поведения автора (авторов), связанного с фальсификацией и/или фабрикацией данных, статья может быть отозвана. Инициатором отзыва статьи может быть редакция, автор, организация, частное лицо. Отзывная статья помечается знаком «Статья отозвана», на странице статьи размещается информация о причине отзыва статьи. Информация об отзыве статьи направляется в базы данных, в которых индексируется журнал.

УВАЖАЕМЫЕ ПОДПИСЧИКИ!

Подписку на журнал «Сибирский вестник сельскохозяйственной науки»
(как на годовой комплект, так и на отдельные номера)
можно оформить одним из следующих способов:

- на сайте Почта России. Зайти в раздел «Онлайн-сервисы», затем – «Подписаться на газету или журнал». Подписной индекс издания ПМ401;
- в агентстве подписки ГК «Урал-Пресс» по индексу 46808. Ссылка на издание http://ural-press.ru/catalog/97210/8656935/?sphrase_id=319094. В разделе контакты зайти по ссылке <http://ural-press.ru/contact/>, где можно выбрать филиал по месту жительства;
- в редакции журнала (телефон 7-383-348-37-62; e-mail: sibvestnik@sfscs.ru).

Полнотекстовая версия журнала
«Сибирский вестник сельскохозяйственной науки»
размещена на сайте Научной электронной библиотеки:
<http://www.elibrary.ru>.