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РЕДКОЗЕМЕЛЬНЫЕ ЭЛЕМЕНТЫ В ПОЧВАХ ЗАСОЛЕННЫХ АГРОЛАНДШАФТОВ БАРАБИНСКОЙ РАВНИНЫ

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Изучено валовое содержание редкоземельных элементов в профиле почв катены засоленных агроландшафтов Барабинской равнины в пределах Новосибирской области. Определено валовое содержание циркония, иттрия, скандия, галлия, в том числе лантаноидов – церия, лантана и иттербия. Редкоземельные элементы крайне слабо изучены. В настоящее время влияние их на растения, организмы животных и человека активно исследуют, хотя предельно допустимые и ориентировочно допустимые концентрации для них пока не разработаны. Валовое содержание редкоземельных элементов, определяемое в почвах катены, зависит от гранулометрического состава и степени гумусированности почвенных горизонтов. Выявлено, что в изученных почвах они в основном содержатся в количестве кларков земной коры, за исключением лантана в гумусовых горизонтах, где его содержание почти в 1,5 раза (44–48 мг/кг) больше кларка в земной коре (29 мг/кг) и иттербия (в 10 раз больше кларка). По профилю почв отмечено незначительное передвижение редкоземельных элементов как в вертикальном, так и в горизонтальном направлении, что свидетельствует о малой подвижности их соединений. В профиле изучаемых почв из группы редкоземельных элементов преобладает цирконий. Его содержание в гумусовых горизонтах почв элювиальных позиций находится в пределах кларка земной коры, колебания по профилю незначительны. В больших количествах содержится иттербий – от 1,89 до 4,05 мг/кг почвы, что значительно больше кларка земной коры (0,3 мг/кг почвы). Роль лантаноидов в системе почва – растения – животное – человек требует дальнейшего глубокого изучения.

Ключевые слова: Барабинская равнина, засоленные агроландшафты, катена, редкоземельные элементы, лантаноиды

RARE EARTH ELEMENTS IN SOILS OF SALINE AGROLANDSCAPES OF THE BARABA PLAIN

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The total content of rare earth elements in the soil profile of saline agricultural landscapes catena of the Baraba plain within Novosibirsk region was studied. The total content of zirconium, yttrium, scandium, gallium, including lanthanides – cerium, lanthanum and ytterbium was determined. Rare earth elements are extremely poorly analyzed. At present, their influence on plants, organisms of animals and humans is being actively studied, although the maximum permissible and tentatively permissible concentrations for them have not yet been developed. The total content of rare earth elements, determined in the soils of the catena, depends on the granulometric composition and the degree of humus content of the soil horizons. It was revealed that in the studied soils they are mainly contained in the number of clarkes of the earth's crust, with the exception of lanthanum in the humus horizons, where its content is almost 1.5 times (44–48 mg/kg) higher than the clarke in the earth's crust (29 mg/kg), and ytterbium (10 times higher than the clarke). Along the soil profile, an insignificant movement of rare earth elements in both vertical and horizontal directions was noted, which indicates a low mobility of their compounds. Zirconium predominates in the profile of the studied soils from the group of rare earth elements. Its content in the humus horizons of soils of eluvial positions is within the clarke of the earth's crust; variations along the profile are insignificant. Ytterbium is contained in large quantities – from 1.89 to 4.05 mg/kg of soil, which is significantly higher than the clarke of the earth's crust (0.3 mg/kg of soil). The role of lanthanides in the soil – plant – animal – human system requires further in-depth study.

Keywords: Baraba plain, saline agricultural landscapes, catena, rare earth elements, lanthanides.

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INTRODUCTION

At present, due to challenging ecological conditions both in Russia as a whole and in Western Siberia, when studying the properties of agricultural landscapes special attention is paid to their microelement composition. It is important to know not only its average content for a particular territory, but also more detailed information for each agricultural landscape. The data obtained are necessary for the development of adaptive landscape farming systems: scientifically grounded systems of processing, crop rotations, fertilizers, etc. The main goal of scientifically grounded farming systems is

to obtain high and stable yields of ecologically clean and high-quality agricultural crops. This is possible only with a deep knowledge of the elemental chemical composition of soils and parent rocks. The chemical composition of soils is directly determined by the composition of the parent rocks, in which, in turn, the composition of the original rocks is preserved, which is discussed in detail in the works of V.B. Ilyina, A.I. Syso [1, 2]. The soil cover of the Novosibirsk region is characterized by a large variegation of the total content of trace elements (heavy metals), which is inherited from the parent rocks. In the process of soil formation, they are somewhat redistributed along the soil profile, relief elements, and biogenic accumulation is manifested in the humus horizons. It has been established that the main carriers of heavy metals are silt particles, humic substances and hydroxides [2].

According to A.P. Vinogradov, "... all the chemical elements of the earth's crust, including soils, may be necessary or useful for plants and living organisms. There can be no toxic elements in nature, there are their toxic concentrations "[1].

At present, a certain part of microelements in soils has been studied rather thoroughly. Hazard standards for different territories, maximum permissible concentrations, tentatively permissible concentrations, etc. have been developed. However, devices have appeared that make it possible to determine more accurately a large number of microelements. The significance of some of them for the life of plants, animals and humans has not yet been established, but the very constant presence in plants indicates their importance.

In the process of studying microelements in the soils of saline agricultural landscapes of the Baraba Plain, the content of 37 macro- and microelements was determined by atomic emission spectroscopy.

In the present paper the gross content of rare earth elements (REE), the significance of which is currently underexplored for plants and living organisms has been reviewed.

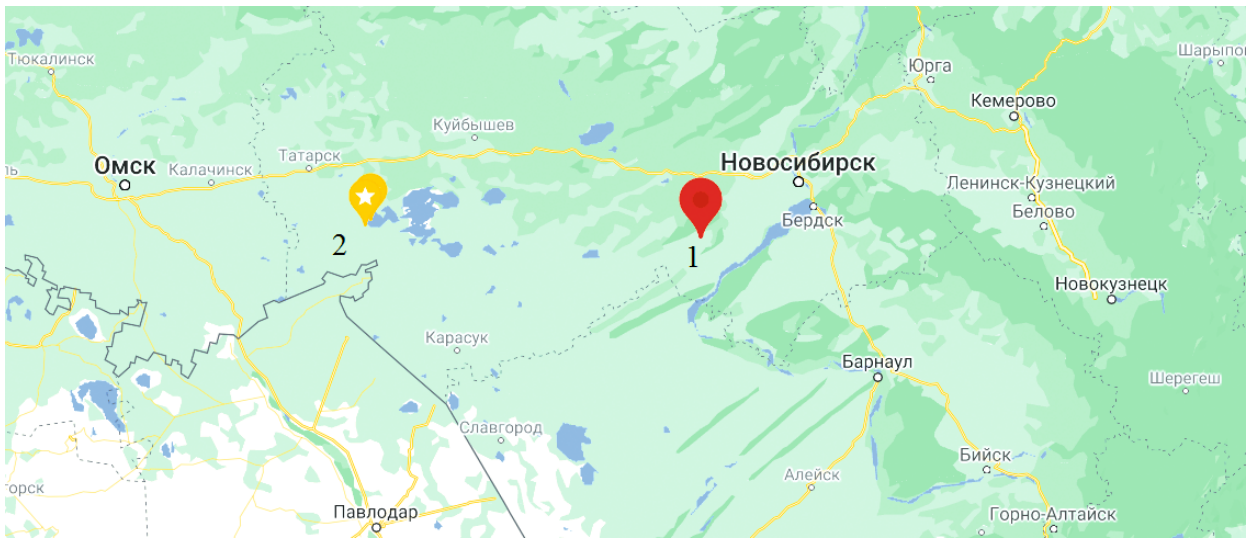
The aim of the study is to investigate the total content of rare earth elements in the soil profile of the saline agricultural landscapes catena of the Baraba Plain within the Novosibirsk region.

MATERIALS AND METHODS

The studies were carried out in two key areas of the Baraba Plain: in the wide valley of the Karasuk River and in the basin of the lake Chany. The first site is located in the Chulym district of the Novosibirsk region near the village Bolshenikolskoe. Three soil sections were laid in the saline agrolandscape along the catena. Section No. 1 is located at the eluvial position of the mesorelief (a gently sloping ridge). The coordinates are - 54˚31′28.9 ″ N. and 81˚29′45.3 ″ E, the altitude above sea level is 203.3 m. The soil is meadow-chernozemic ordinary leached medium-thick medium loamy. Section 2 was excavated in the transit zone. The coordinates are - 54 ° 35'14.5 ″ N. and 81 ° 29′45.3 ″ E. The soil is chernozem-meadow saline sandy loam, in which, at a depth of 72 cm, the profile of the buried soil with horizons A of burials and B of burials is clearly distinguished. Section No. 3 was laid in the accumulative zone. The coordinates are - 54˚35′37.4 ″ N. and 81˚29′11.5 ″ E, the altitude above sea level is 229 m. The soil is a deep solonchak heavy loamy meadow solonetz (see the figure).

The second site is located on the drying Yudinsky reach of Lake Chany. Two cuts were laid along the catena. In the eluvial position, section no. 40 was dug at the summit of the ridge at an altitude of 120 m above sea level. The coordinates are - 54˚74'58.98 ″ N and 76˚06'94.00 ″ E. Soil - meadow-chernozem ordinary, slightly salted, medium-thick sandy loam. In the accumulative zone in geochemical subordination, section No. 20 was laid. The coordinates are - 54˚78′11.33 ″ N. and 76˚83'95.28 ″ E, absolute elevation above sea level 103m. The ridge, with

[1]Vinogradov A.P. The main regularities in the distribution of trace elements between plants and the environment // Microelements in the life of plants and animals. Moscow: Publishing House of the Academy of Sciences of the USSR, 1952.



Географическое расположение объектов исследования
Geographic location of research objects

a sharp ledge, is transformed into a gently sloping surface of the dried bottom of the Yudinsky reach. Ground water was found at a depth of 60 cm. The soil is meadow-boggy, saline, heavy loamy (see the figure).

The morphological description of the soil profiles was carried out and soil samples were taken from the genetic horizons. Analyzes were carried out in them according to generally accepted methods: granulometric composition according to Kachinsky, absorbed bases - according to Schollerberger, humus - according to Tyurin, pH - potentiometrically[2]. Trace elements were determined on a two-jet atomic emission plasmatron (DAEP) by atomic spectroscopy in the laboratory of soil biogeochemistry of the Institute of Soil Science and Agrochemistry of the Siberian Branch of the Russian Academy of Sciences.

RESULTS AND DISCUSSION

The total content of seven rarely detected trace elements - zirconium (Zr), yttrium (Y), scandium (Sc) and gallium (Ga), including lanthanides - cerium (Ce), lanthanum (La) and ytterbium (Yb) has been studied in the soils of saline agricultural catena landscapes. Since they exhibit similar properties, they have been combined into one group of rare earth elements. This name is due to the fact that these elements are believed to be rare and their concentrations are widely scattered both inside the earth's crust and in soils. However, in-depth studies of recent years have shown that REEs are not rare. In terms of total prevalence, they exceed lead by 10 times, molybdenum by 50 times [5].

In the last decade, rare earth elements have begun to be used in various industries, and their release into the environment has increased accordingly. The effect of REEs on plants and organisms of animals and humans is currently being actively studied.

As our studies have shown, zirconium predominates in the soils of both catenas from the REE group (see Tables 1, 2). Its content in the humus horizons of soils in the eluvial positions of catena is 179–200 mg / kg of dry soil. Down the profile, the fluctuations are insignificant, but its amount is somewhat less - about 170 mg / kg of soil, which corresponds to the clarke of this element in the earth's crust[3].

In the transit position of the first catena in the profile of chernozem-meadow soil at a depth of 72–90 cm, horizon A of the buried soil is clearly distinguished, in which the zirconium

[2]Workshop on agrochemistry / ed. by V.G. Mineev. M.: Publishing house of Moscow State University. 2001. 687 p.
[3]Alekseenko V.A. Environmental geochemistry: textbook. M.: Logos, 2000. 627 p.

Табл. 1. Содержание редкоземельных микроэлементов в почвах засоленных агроландшафтов первой катены

Table 1. The content of rare earth trace elements in the soils of saline agricultural landscapes of the first catena

Geomorphological position, section No., soil	Horizon, cm	Trace element (heavy metal), mg / kg dry soil						
		Zr	Y	Sc	Ga	Ce	La	Yb
Eluvial, R, No. 1, meadow chernozem medium thick medium loamy	$A_{пах}$ 0–18	200	29,9	14,9	10,6	68,7	31,4	3,01
	A_1 18–45	270	41,6	18,4	10,3	92,5	44,9	4,05
	AB 45–65	198	32,4	17,8	15,7	94,7	41,4	3,38
	B 65–85	254	42,9	20,8	12,4	87,5	48,6	4,29
	$B_к$ 85–135	215	36,1	18,0	10,5	77,7	42,4	3,52
Transit, R, No. 2, chernozem-meadow slightly salty heavy loamy	A_d 0–10	208	20,7	10,3	11,7	43,8	28,2	1,94
	A_1 10–24	129	15,1	10,8	8,75	80,4	33,5	2,08
	AB_q 24–43	174	13,5	9,09	7,25	14,0	22,6	1,59
	B_q 43–72	113	15,5	9,38	5,67	54,2	24,7	1,83
	$A_{пог}$ 72–90	222	28,6	14,6	11,1	61,0	37,5	2,98
	$B_к$ 90–130	135	15,2	11,2	8,85	66,3	32,5	1,91
Accumulative, R, No. 3, deep meadow saline solonchak heavy loamy	$A_{пах}$ 0–20	276	19,4	12,2	8,39	69,3	29,7	2,18
	B_1 25–35	126	19,3	10,6	9,11	51,3	22,8	1,89
	B_{2q} 35–50	149	24,6	11,7	9,8	60,8	26,7	2,36
	B_{3q} 50–68	168	31,2	13,2	10,1	84,7	35,6	3,25
	B_{4q} 68–80	195	28,9	14,9	8,17	71,0	32,3	2,82
Clarke element in the earth's crust [5]		170	20	10	19	70	29	0,3

Табл. 2. Содержание редко определяемых микроэлементов в почвах засоленных агроландшафтов второй катены

Table 2. The content of rarely detected trace elements in the soils of saline agricultural landscapes of the second catena

Geomorphological position, section No., soil	Horizon, cm	Trace element (heavy metal), mg / kg dry soil						
		Zr	Y	Sc	Ga	Ce	La	Yb
Eluvial, P, 40, meadow chernozem solodized sandy loam	A_1 3–18	193,0	26,2	12,1	8,87	50,2	30,4	2,62
	A_1 18–30	224,0	26,9	11,4	9,93	39,7	24,8	2,72
	AB 35–45	177,0	19,1	8,4	8,41	38,7	23,3	1,86
	B_{ca} 61–93	178,0	24,5	16,9	8,65	58,2	34,1	2,28
	C_{Ca} 90–100	178,0	25,5	11,9	8,62	52,5	29,5	2,41
Accumulative, R, No. 20 meadow-swamp saline heavy loamy	A_q 0–7	179,0	21,3	24,1	14,9	69,6	49,0	3,57
	A_{1q} 10–20	171,0	30,7	21,0	13,0	59,4	39,3	3,37
	B_q 40–50	136,0	24,9	17,4	12,3	46,6	25,5	2,68
	G 65–75	174,0	31,5	22,8	12,0	55,6	39,4	3,34
Clarke element in the earth's crust [5]		170	20	10	19	70	29	0,3

content is high - 222 mg / kg. In the accumulative position, the largest amount of zirconium was found in the humus horizon of the deep solonetz - 276 mg / kg; it decreases down the profile. It is known that zirconium belongs to low-mobile elements, does not form water-soluble compounds within the landscape, and weakly migrates with organic complexes [3, 4]. In our opinion, some of its accumulation in the humus horizon of soils in the accumulative zone is associated with the erosional processes of the movement of clay fractions from the upper positions of the catena to the lower water flows. In the second catena, approximately the same zirconium content was found in the profiles of the studied soils.

The gross content of yttrium in the soil profiles of the studied catenas is also close to the clarke of this chemical element in the earth's crust. In the first catena at the eluvial position in the meadow chernozem soil, the amount of yttrium is somewhat higher than in the second, due to the heavier granulometric composition (see Table 3). V.V. Ivanov noted that there are no reliable soil clarkes of yttrium, since "soils are rarely analyzed for yttrium." He believes that

Табл. 3. Физико-химические свойства почв катены
Table 3. Physical and chemical properties of catena soils

Geomorphological position, section No., soil	Horizon, cm	Physical and chemical properties		
		pH	Humus,%	Physical clay, particles <0.01 mm,%
Eluvial, R, No. 1, meadow chernozem medium-power medium loamy	$A_{пах}$ 0–18	6,6	9,7	36,5
	A_1 18–45	6,6	7,7	31,1
	AB 45–65	6,3	1,3	53,5
	B 65–85	6,7	Not ident.	53,9
	$B_к$ 85–135	8,4	» »	56,3
Transit, R, No. 2, chernozem-meadow slightly salty heavy loamy	A_d 0–10	7,9	10,8	14,0
	A_1 10–24	8,2	4,5	8,8
	AB_q 24–43	8,3	1,0	21,4
	B_q 43–72	8,4	0,6	22,6
	$A_{пог}$ 72–90	8,9	0,9	53,3
	$B_к$ 90–130	9,1	0,6	56,1
Accumulative, R, no. 3, deep meadow saline solonchak heavy loamy	$A_{пах}$ 0–20	9,8	5,2	60,6
	B_1 25–35	10,1	2,6	60,2
	B_{2q} 35–50	10,2	0,9	41,2
	B_{3q} 50–68	10,0	Not ident.	41,2
	B_{4q} 68–80	10,0	» »	54,6
Eluvial, R, No. 40, meadow chernozem solodized sandy loam	A_1 3–18	6,8	5,7	14,8
	A_1 18–30	7,1	4,5	14,1
	AB 35–45	7,3	1,1	36,1
	B_{ca} 61–93	8,9	0,5	29,4
	C_{Ca} 90–100	9,0	0,2	26,9
Accumulative, R, no. 20, meadow-swamp saline heavy loamy	A_q 0–7	8,9	5,9	72,2
	A_{1q} 10–20	9,0	2,8	88,1
	B_q 40–50	8,6	2,8	66,1
	G 65–75	8,9	2,5	51,1

yttrium is in the clay fraction of soils, where its average content is 33 mg / kg, in sandy and calcareous soils - 18 and 8 mg / kg of dry soil, respectively [5]. This conclusion is consistent with the conclusions of other researchers [1, 2], including ours.

Scandium is a typical scattered lithophilic element. Due to its very low concentration in the earth's crust, its geochemistry is poorly studied, although it has a rather high clarke - 10 mg / kg. This is higher than tin and almost the same as lead. In the last decade, it was established that the content of scandium in soils depends on its content in the parent rock. Its lowest concentration is typical for sandstones and light soils; in heavy and clayey soils, the scandium content is much higher [6].

It should be noted that the scandium content in the soils of both catenas is equal to the clarke or is slightly higher. In the first catena, it is more abundant in the profile of the meadow-chernozemic medium loamy soil of the eluvial position, in the second, in the meadow-boggy saline heavy loamy accumulative position. Along the profile, fluctuations in the content of scandium are not so significant, which indicates a weak movement of its compounds.

The average content of this element in land plants is 0.008 mg / kg dry weight[4], in vegetables - 0.005 mg / kg, in grass - 0.07, while in barley roots - 0.63 mg / kg. The content of scandium in old leaves is higher than in young ones [7]. The biological role of scandium and its migration along biological chains is very poorly studied. It has now been established that scandium, like other REEs, is not a necessary chemical element for the life of plants, animals and humans, especially since it is not a "mineral of death" - a potent poison [8, 9].

The gross gallium content in the catena soil profile is almost 2.0–2.5 times lower than its clarke value in the earth's crust - 19 mg / kg. Its fluctuations along the profile are insignificant. In the soils of the first catena, the amount of gallium is greater in the meadow chernozemic medium loamy soil of the eluvial position than in the meadow chernozem solodized sandy loamy soil of the second catena (see Table 1). In the accumulative position, an inverse dependence of the gallium content in soils was found, which is associated with both the granulometric composition and the location of the sections along the relief. The gallium content probably depends on the humus content: with a decrease in humus in the A horizons of the soils, its amount decreases. In the buried horizon A of the soil, it is approximately the same as in the upper horizon A (see Table 1).

The gross content of cerium, the clarke of which is 70 mg / kg (see footnote 3), in the soil profile of the second catena is significantly lower than in the soil profile of the first catena, which is probably due to their lighter granulometric composition along the genetic horizons and the content of cerium in parent rock.

In the humus horizon of the chernozem-meadow soil of the transit zone, it is somewhat less than in the eluvial and accumulative ones. There is little data on the role of cerium in the life of plants, animals and humans, but recently there have been studies to study its effect on the elemental composition and development of plants [5].

The gross content of lanthanum is approximately the same in the soils of both catenas, and it is within the clarke of the earth's crust - 29 mg / kg of soil (see Tables 1, 2). Only the profile of the meadow chernozem soil of the eluvial position in the first catena is distinguished, where the total content of lanthanum along the profile is in an amount higher than the clarke - from 41.4 to 48.6 mg / kg, while in the profile of the soils of the transit and accumulative position it is slightly less - from 22.6 to 37.5 mg / kg. Much attention has been paid to this element by Yu.N. Vodyanitsky [10]. It has been established

[4]Scandium. it's Occurrence, Chemistry, Physics, Metallurgy and Technology. London, New York, Krancisco: Acad. Press, 1975. 598 p.

[5]Kotelnikova A.D., Volkov D.S., Fastovets I.A., Rogova O.B. The influence of cerium on the elemental composition of barley plants when introduced into sod-podzolic soil. Soil Science: Horizons of the Future: Proceedings of the III Intern. conf. (Soil Institute named after Dokuchaev). M., 2019.

that lanthanum in micro doses has a stimulating effect on plants, in high doses it has a depressive effect [11, 12].

It was found that the total content of ytterbium in the soil profile of catena in saline agricultural landscapes of Baraba significantly exceeds the clarke in the earth's crust. Clarke of ytterbium - 0.3 mg / kg, and in soils of catena it contains from 1.6 to 4.3 mg / kg of soil. Its greatest amount is in the meadow chernozem soil of the first catena, the smallest is here in the transit zone, which is directly related to the granulometric composition. It should be noted that plants are able to resist the accumulation of REE. They block their transfer into the human body and do not accumulate in it.

Statistical processing of the material.

To perform statistical processing, consider the physicochemical properties of catena soils, presented in table 3. The granulometric composition of soils in the first catena varies along the profile in the eluvial position from medium loamy to heavy loamy, in the transit one - from sandy loam in horizon A to heavy loamy in the lower horizons. The humus content in the studied soils is rather high in the upper horizons (9.7–10.8%). It falls sharply down the profile. The pH value in soils increases from the eluvial position to the accumulative one.

In the soils of the second catena in the eluvial position, the granulometric composition of soils in horizon A is sandy loamy, and becomes heavier downward to light loamy. In the accumulative position, it becomes heavily heavier to heavy clay. The soils of the second catena are less humus-rich than the first. The pH value in the soils of the eluvial position in the upper horizons is neutral, becoming alkaline with depth. In the accumulative one, an alkaline medium is preserved throughout the profile.

Statistical processing of the research results was carried out in the applied statistics package of standard Excel programs. In particular, a correlation analysis was carried out between

Табл. 4 Коэффициенты корреляции между распределением редкоземельных элементов по профилю и физико-химическими свойствами почв

Table 4. Coefficients of correlation between distribution of rare earth elements along the profile and the physical and chemical properties of soils

Geomorphological position, section No., soil	Physical and chemical properties	Trace element						
		Zr	Y	Sc	Ga	Ce	La	Yb
Eluvial, R, No. 1, meadow chernozem medium-power medium loamy	Physical clay	–0,34	0,01	0,45	0,51	0,12	0,37	0,08
	Humus	0,32	0,10	–0,55	–0,96	–0,73	–0,47	–0,06
	pH	–0,10	0,05	0,05	–0,48	–0,42	0,08	–0,06
Transit, R, No. 2, chernozem-meadow slightly salted heavy loamy	Physical clay	0,20	0,43	0,66	0,20	0,18	0,52	0,51
	Humus	0,37	0,14	–0,15	0,61	–0,01	–0,03	–0,10
	pH	–0,09	0,22	0,56	–0,05	0,32	0,50	0,41
Accumulative, R, No. 3, deep meadow solonchak heavy loamy	Physical clay	0,39	–0,70	–0,17	–0,81	–0,46	–0,42	–0,64
	Humus	0,85	–0,79	0,41	–0,99	0,57	0,53	–0,27
	pH	–0,93	0,19	–0,22	0,46	–0,46	–0,42	–0,06
Eluvial, R, No. 40, meadow chernozem solodized sandy loam	Physical clay	–0,78	–0,85	–0,11	–0,78	0,05	–0,06	–0,95
	Humus	0,68	0,49	–0,17	0,57	–0,32	–0,17	0,68
	pH	–0,49	0,04	0,58	–0,34	0,70	0,58	–0,21
Accumulative, R, no. 20, meadow-swamp saline heavy loamy	Physical clay	0,09	–0,10	0,32	0,43	0,32	0,16	–0,09
	Humus	0,43	–0,82	0,81	0,96	0,81	0,70	0,58
	pH	0,61	0,82	0,22	–0,21	0,22	0,34	0,37

the content of rare earth elements in soils with particle size distribution, humus and pH (see Table 4).

Certain regularities between the content of rare earth elements, granulometric composition, humus and pH are weakly manifested, although it can be noted that this relationship is absent in soils of eluvial positions. Only between zirconium and ytterbium, granulometric composition and humus is a close inverse relationship established. In the accumulative position, a close inverse relationship was also established between cerium, lanthanum, yttrium, ytterbium, scandium, and humus.

CONCLUSIONS

1. Rare earth elements zirconium, yttrium, scandium, gallium, including lanthanides - cerium, lanthanum, ytterbium are currently poorly studied, their significance in the life of plants, animals and humans has not been established. However, the constant presence of rare earth elements in the chemical composition of plants indicates their need.

2. Rare earth elements were found in all soils of the studied saline agrolandscapes. Their content is determined by the content in the parent rock, depends on the granulometric composition and the amount of organic matter in the humus horizon.

3. In the soils of saline agricultural landscapes of the Baraba Plain, rare earth elements, including lanthanides, are mainly found in the amounts of clarkes of the earth's crust, with the exception of lanthanum in the humus horizon (1.5 times the clarke value) and ytterbium (up to 10 times higher than the clarke value). Some changes in the content of rare earth elements are observed along the soil profile. However, these changes are insignificant, which indicates a weak movement of REE compounds both in the vertical and horizontal directions.

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АГРОФИЗИЧЕСКИЕ И АГРОХИМИЧЕСКИЕ СВОЙСТВА ТЕМНО-СЕРЫХ ЛЕСНЫХ ПОЧВ ПРИ РАЗЛИЧНЫХ СИСТЕМАХ ОСНОВНОЙ ОБРАБОТКИ

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Определено влияние продолжительного использования различных систем основной обработки темно-серой лесной почвы на агрофизические свойства, питательный режим и урожайность зерновых в зернопаровых севооборотах. Исследования проведены в условиях Северного Зауралья в стационарном опыте в 1996–2018 гг. Изучены традиционная отвальная и ресурсосберегающие системы основной обработки почвы. Опыт проходил в течение 3–6-й ротации двух зернопаровых севооборотов, развернутых во времени и в пространстве: чистый пар – озимая рожь – яровая пшеница – яровая вика – яровой ячмень; чистый пар – озимая рожь – яровая пшеница – яровая пшеница – яровой ячмень. При возделывании зерновой культуры, завершающей зернопаровой севооборот, в четвертом поле после пара по зернобобовому предшественнику (яровой вике) целесообразно применение систем основной обработки почвы с элементами минимизации. К ним относятся безотвальная и комбинированная системы с безотвальным рыхлением плугом со стойками СибИМЭ на 20–22 см; дифференцированная с плоскорезной обработкой на 12–14 см и дискование на 10–12 см. Изучаемые системы обработки обеспечивали близкие отвальной системе условия водного режима, сложения почвы и пищевого режима. Сформирована практически равная отвальной системе урожайность ячменя: на фоне естественного плодородия – 2,97–3,03 т/га, с применением $N_{40}P_{40}P_{40}$ – 3,47–3,65 т/га. Применение систем обработки с элементами минимизации в зернопаровом севообороте без наличия зернобобовой культуры под данную культуру по повторной зерновой культуре (пшенице) приводило к следующим результатам. Обеспеченность продуктивной влагой в слое почвы 0–1,0 м снизилась на 8,6–28,0%, значительно ухудшился пищевой режим, в особенности азотный (на 15,5–43,8%) и фосфорный (на 39,1–51,1%), с отрицательной дифференциацией плодородия почвенного профиля, урожайность зерна снизилась на 0,09–0,40 т/га.

Ключевые слова: система основной обработки, севооборот, предшественник, элементы плодородия, яровой ячмень, урожайность, эффективность производства

AGROPHYSICAL AND AGROCHEMICAL PROPERTIES OF DARK GREY FOREST SOILS UNDER DIFFERENT SYSTEMS OF BASIC TILLAGE

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The effect of long-term use of different systems of basic tillage of dark grey forest soils on the agrophysical properties, nutrient regime and yield of grain crops in grain-fallow crop rotations was determined. The study was carried out in the conditions of the Northern Trans-Urals in a stationary experiment in 1996–2018. The traditional moldboard and resource-saving systems of basic tillage were studied. The experiment took place during the third–sixth rotations of two grain-fallow crop rotations spread in time and space: bare fallow – winter rye – spring wheat – spring vetch – spring barley; bare fallow – winter rye – spring wheat – spring wheat – spring barley. When cultivating a grain crop that completes a grain-fallow crop rotation, in the fourth field after the fallow with a legume forecrop (spring vetch), it is advisable to use systems of basic tillage with elements of minimization. These include non-moldboard and combined tillage with subsurface loosening by

a plow with SibIME tines to a depth of 20–22 cm differentiated with stubble-mulch at 12–14 cm and disk harrowing at 10–12 cm. The studied tillage systems ensured the conditions of the water regime, soil composition and nutritional regime close to the moldboard tillage system. The yield of barley almost equal to the moldboard system was formed: against the background of natural land fertility – 2.97–3.03 t/ha, with the use of $N_{40}P_{40}P_{40}$ – 3.47–3.65 t/ha. Application of tillage systems with minimization elements in a grain-fallow crop rotation without planting a leguminous crop with a given crop for a repeated grain crop (wheat) led to the following results. Productive moisture availability in the soil layer 0–1.0 m decreased by 8.6–28.0%, the nutrient regime worsened significantly, especially nitrogen (by 15.5–43.8%) and phosphorus (by 39.1–51.1%), with the negative differentiation of soil fertility, and reduction of grain yield by 0.09–0.40 t/ha.

Keywords: system of basic tillage, crop rotation, forecrop, fertility elements, spring barley, yield, production efficiency.

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The authors declare no conflicts of interest.

INTRODUCTION

In the Northern Trans-Urals, as well as in other regions of the country, in field cultivation there is an increasing use of less expensive methods of cultivation than ploughing, using non-moldboard, flat-cut processing and disking in basic cultivation. This is due to economic reasons, the possibility of increasing productivity and reducing fuel costs. However, scientific studies on this topic in the region and abroad do not give an unambiguous answer about the influence of technologies of different intensity on the elements of soil fertility, as well as about their comparative efficiency in comparison with traditional plowing [1-3]. At the same time, it is important to establish the peculiarities of the influence of cultivation systems on soil fertility during the cultivation of cereals in crop rotations with precursors of different biology [4–6].

The aim of the study is to determine the effect of long-term use of various systems of the main processing of dark gray forest soil in the conditions of the Northern Trans-Urals on agrophysical properties, nutritional regime and yield when cultivating a grain crop grain using forecrop (spring wheat) and leguminous (vetch for grain) predecessors.

MATERIALS AND METHODS

The studies were carried out in 1996–2018 in the stationary experiment of the Scientific Research Institute of Agriculture of the Northern Trans-Urals - a branch of the Tyumen Scientific Center of the SB RAS. The experiment took place during the period of the 3rd-6th rotations of two grain-fallow crop rotations, deployed in time and space: pure fallow - winter rye - spring wheat - legumes (spring vetch for grain) - spring barley; pure fallow - winter rye - spring wheat - spring wheat - spring barley. Observations and counts were carried out in a barley field. The studied soil cultivation systems are presented in table 1. Techniques of the main processing were performed after harvesting the crops.

Pre-sowing treatment for all studied systems of the main treatment consisted in early spring harrowing with BZSS-1.0 tooth harrows in presowing cultivation with the Smaragd-6 cultivator to the depth of seeding. Sowing was carried

Табл. 1. Системы обработки почвы
Table 1. Soil tillage systems

Soil tillage system	Tillage methods after harvesting crops				
	Steam	Winter rye	Wheat	Grain legumes / wheat	Barley
Mouldboard	Plowing with a Lemken plow to a depth of 20-22 cm				
Non-mouldboard	Loosening with SibIME tines to a depth of 20-22 cm				
Combined	Lemken to a depth of 20-22 cm	SibIME tines to a depth of 20-22 cm	Lemken to a depth of 20-22 cm	SibIME tines to a depth of 20-22 cm	Lemken to a depth of 20-22 cm
Differentiated	Smaragd cultivator to a depth of 12–14 cm	Smaragd cultivator to a depth of 12–14 cm	Lemken to a depth of 20-22 cm	BDT-2,5 to a depth of 10–12 cm	BDT-2,5 to a depth of 10–12 cm
Flat-cut	Smaragd cultivation to a depth of 12–14 cm				
Shallow	BDT-2,5 disking to a depth of 10–12 cm				

out with a SZP-3.6 seeder. Mineral fertilizers (against a background with fertilizers) were applied before pre-sowing cultivation with a rate of $N_{40}P_{40}K_{40}$ kg ai. per 1 hectare of crop rotation area. For the destruction of weeds, herbicides were used in all studied variants.

The area of the plots (5.5–6.0 × 63 m) is 346–378 m^2, the accounting area is 100 m^2. During the years of research on the conditions of moisture supply of the growing season, 74% of the years were close to average, 26% were insufficiently provided with precipitation. The technique of bookmarking and conducting the experiment and statistical processing of the data obtained were carried out according to the method of B.A. Dospekhov[1] using O.D. Sorokin[2]. The determination of the agrophysical properties of the soil was carried out according to the method of N.A. Kachinsky[3]. Soil moisture was determined by the thermostat-weight method in each 10-cm layer to a depth of 0–100 cm. Productive moisture reserves were calculated taking into account the bulk density and wilting moisture for each 10-cm layer. The data obtained were combined by layers of 0–30 and 0–100 cm. The chemical analysis of soil samples was carried out according to generally accepted methods[4].

RESULTS AND DISCUSSION

Under the conditions of unstable moisture in the Northern Trans-Urals, the yield, as well as the possibility of effective use of intensification agents (intensive varieties, fertilizers, protective agents, biostimulants), are largely determined by the presence of moisture in the soil. The need for its maximum accumulation and preservation is due to an unfavorable balance between the intake and consumption of moisture, especially in the initial growing season [7, 8].

One of the main practical measures to influence the most rational use of available moisture resources is tillage [9]. In our studies, during the sowing - seedling period of grain, the moisture reserves of the 0–100 cm soil layer were assessed as satisfactory - 90–125 mm (60–83% of the Field Moisture Capacity), in the 0–30 cm soil layer - 32.0–35.7 mm (52–58% of the Field Moisture capacity).

The most consistently high moisture reserves during this period in grain crops seeds (barley), completing the crop rotation for leguminous crops and for repeated wheat, were provided by plowing (96.8–125.0 mm) and moldboard-free loosening by a plow with stands of the SibIME design (93.0 –122.4 mm) to a depth

[1]Dospekhov B.A. Field experiment technique: 4th ed., rev. and add. Moscow: Kolos, 1979.416 p.
[2]Sorokin O.D. Applied statistics on the computer. Krasnoobsk: EPU SB RAAS, 2004.162 p.
[3]Kachinsky N.A. Soil physics. M.: Higher school, 1965. 318 p.
[4]Arinushkina E.V. Manual for Chemical Analysis of Soils. Moscow: Moscow State University Publishing House, 1970.478 p.

of 20–22 cm. When using leguminous crops as a predecessor, resource-saving processing systems (dump-free, combined with the use of SibIME tines) provided moisture reserves in a meter layer during the sowing period - barley shoots similar to the plowing option. The decrease in moisture reserves for small treatments (flat-cut cultivator Smaragd to a depth of 12-14 cm and disking BDT-2.5 to a depth of 10-12 cm) in comparison with the option of plowing was 4.2-7.1 mm, or 4, 3-7.7% in relation to the control (see Fig. 1).

When placing the final crop rotation of a grain crop (barley) on a repeated grain crop (wheat), an increase in the negative impact of resource-saving systems and methods of processing on moisture reserves was noted in comparison with plowing. So, according to the annual plow-free tillage to a depth of 20-22 cm, the decrease in moisture reserves in the meter layer of soil in relation to the option of plowing reached 10.3 mm, or 8.2% in relation to it, for flat-cut processing - 8.6%, according to disking - 28%. In the 0–30 cm soil layer, all studied cultivation systems for both predecessors provided equal moisture conditions. The best conditions for moisture supply for deep moldboard and non-moldboard treatments are explained by the fact that in systems with predominantly shallow treatments, there was a more significant compaction of the soil layer 0–20 cm (by 0.03–0.04 g / cm^3) in the layer 10–30 cm - by 0.03–0.07 g / cm^3 [10], which worsened the conditions for assimilation of moisture from precipitation in these treatments. In addition, in most years of the research, the precipitation provision was close to the average long-term, the soil during the treatment period had good moisture. Moisture reserves before treatment were 80 mm or more. According to researchers, in these conditions, deep treatments have advantages in moisture accumulation in comparison with shallow treatments[5] [11]. The increase in the negative impact of shallow treatments on moisture accumulation in barley crops for re-wheat compared to crops for grain predecessor (re-wheat) is explained by the shorter post-harvest period.

The composition of the soils of the experimental plot does not have quite favorable properties. Their equilibrium density (1.36–1.41 g / cm^3) is far from optimal (1.00–

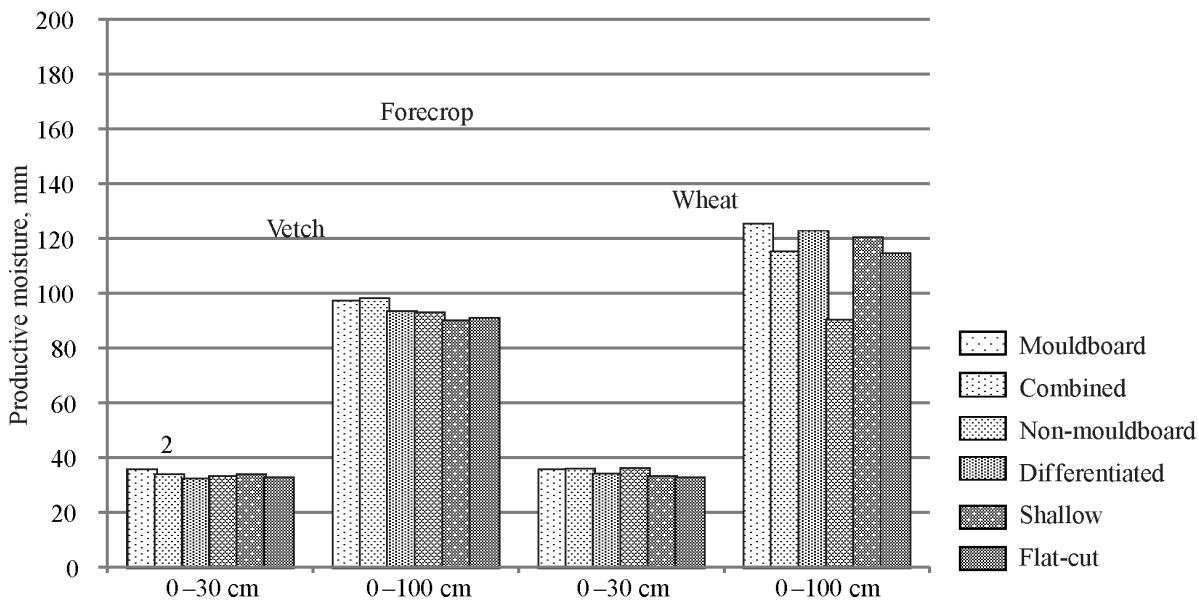

Рис. 1. Влияние основной обработки почвы на влагообеспеченность зерновых в период посев – всходы

Fig. 1. Effect of basic tillage on moisture availability of grain crops during sowing – sprouting period

[5]Moschenko Y.B. Improvement of the elements of the farming system when growing spring wheat on the chernozems of the steppe zone of Western Siberia: author. dis. Dr. Of Sciences in agriculture. Omsk, 1990.32 p.

1.25 g / cm^3) [12]. In this regard, it is important to assess the effectiveness of soil cultivation systems and cultivation methods in the studied systems on the formation of the soil composition regime.

Our data indicate that the density of the topsoil, regardless of the cultivation method and the predecessor, was close to the upper limit of optimal values (1.00–1.25 g / cm^3) for a grain crop (see Fig. 2) [13, 14].

An increase in soil density by 0.03 g / cm^3 was noted in the treatment of BDT-2.5 by disking by 10-12 cm with a differentiated system for the legume predecessor, for a repeated grain crop - by 0.04 g / cm^3 in comparison with the variant of the moldboard processing system.

The importance of the influence of the main processing systems on the peculiarities of the formation of the nutritional regime and the need to study it in relation to zonal and soil conditions have been noted by many scientists [15, 16]. Our research on the nutritional regime has also established a significant effect of tillage and its predecessor on the provision of the arable and sub-arable soil layers with available nutrients (see Table 2). It was revealed that the cultivation of cereals in a grain-fallow crop rotation with the presence of leguminous crops against the background of the application of fertilizers $N_{40}P_{40}K_{40}$ provides favorable conditions for the nutritional regime in the field that closes the crop rotation. On the contrary, in the fourth field of a grain crop after fallow in a crop rotation without a leguminous crop, the content of nitrate nitrogen in the arable layer decreased in comparison with the field with a legume predecessor by 15.5–43.8%, P_2O_5 by 39.1–52.1, K_2 About 15.1-26.4%. This is due to legumes and spring crops differing in nutrient removal, partial compensation of nitrogen removal by legumes due to symbiotic nitrogen fixation, and a longer period of nitrification in the post-harvest period.

According to both predecessors, the best nutritional conditions, both in the arable layer of 0–0.2 m and in the layer of 0.2–0.4 m, were formed with the moldboard cultivation system, which was facilitated by the better conditions for the agrophysical properties of the soil. Long-term application of treatment without formation turnover, treatment systems with predominantly small treatments led to a decrease in the content of N – NO_3 in the arable layer by 12–42%, P_2O_5 by 7.1–19.9, K_2O by 6.5–17.1%, which increased in sowing of repeated grain crops.

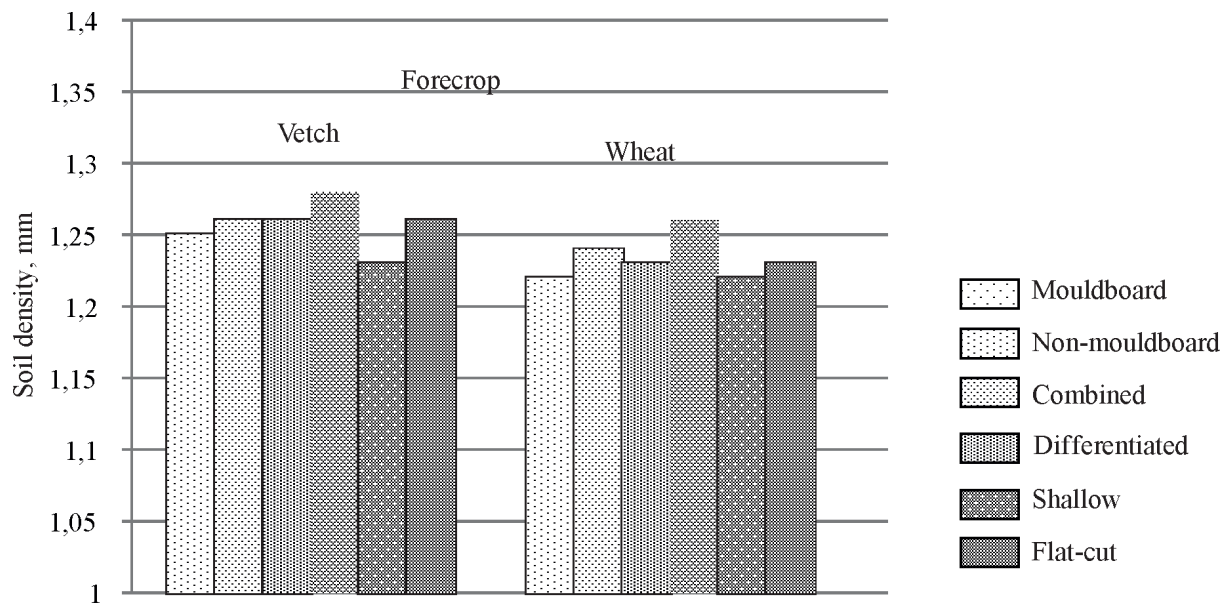

Рис. 2. Плотность пахотного (0–0,2 м) слоя почвы в период посев – всходы в среднем за 3–6 ротаций севооборотов

Fig. 2. Density of arable (0–0.2 m) soil layer, in the period of sowing – sprouting on average 3–6 crop rotations

Табл. 2. Содержание питательных элементов в почве перед посевом зерновых (в среднем за 3–6 ротаций севооборотов)

Table 2. Nutrient content before sowing of grain crops (on average for 3–6 crop rotations)

Soil tillage system	Forecrop	$N–NO_3$, mg / kg soil		P_2O_5 mg / 100 g soil		K_2O, mg / 100 g soil	
		Soil horizon 0–0,2 m					
		0–20	20–40	0–20	20–40	0–20	20–40
Mouldboard	Vetch	8,84	6,26	28,10	17,26	22,85	16,00
	Wheat	6,51	5,21	15,60	9,00	17,20	11,80
Non-mouldboard	Vetch	7,78	4,35	25,26	8,81	22,55	14,70
	Wheat	5,13	4,46	13,20	5,60	16,60	11,10
Combined	Vetch	6,24	4,15	25,30	11,09	21,25	14,90
	Wheat	5,27	4,29	15,40	6,40	17,75	10,80
Differentiated	Vetch	6,24	4,60	26,08	10,08	20,55	13,60
	Wheat	3,78	3,55	12,50	5,20	16,80	11,30
Shallow	Vetch	6,88	5,07	25,70	9,96	20,35	13,70
	Wheat	5,25	3,87	15,40	6,60	15,67	9,81
Flat-cut	Vetch	7,58	4,87	25,70	8,50	18,95	13,20
	Wheat	4,26	3,30	13,70	6,60	16,08	11,58
LSD_{05} Partial effects	On vetch	0,96	0,93	1,95	1,56	1,40	1,15
	On wheat	0,84	0,81	1,12	0,87	1,00	0,73

The general differentiation of the fertility of the arable and sub-arable soil layers for all studied cultivation systems showed the following. There was a decrease in the content of nutrients in the soil layer 0.2–0.4 m in relation to the 0–0.2 m layer: $N – NO_3$ by 13.1–44.1%, $P_2 O_5$ by 38.6–66.9 and $K_2 O$ by 28.0–39.2%. The highest negative differentiation was noted in the content of P2 O5, and it was the smallest for the dump processing system - 38.6–42.3%, while for resource-saving systems - 51.8–66.9%.

Analysis of the influence of the studied soil cultivation systems on crop yields showed that in a grain-fallow crop rotation, when placed on a legume crop, resource-saving cultivation systems provided almost equal barley yields to the moldboard cultivation system. Against the background of natural fertility, it amounted to 2.57–3.03 t / ha, with the use of fertilizers - 3.47–3.65 t / ha (see Fig. 3).

After the legume predecessor in the moldboard and resource-saving processing systems, favorable conditions for agrophysical properties, nutritional regime and, in particular, nitrogen nutrition for leguminous crops developed: nitrogen in the soil was replenished both due to the current nitrification due to a longer post-harvest period, and due to the symbiotic fixation of air nitrogen. In this regard, they obtained indicators of economic and energy efficiency, which are close to the variant of the moldboard processing system. The differences in net income between the moldboard and resource-saving processing systems did not exceed 0.6–3.7% against the background without the use of fertilizers, and 2.6–6.0% with fertilizers.

The highest efficiency was noted for the moldboard processing system: net income amounted to 14.67 thousand rubles / ha against the background without fertilizers and 22.75 thousand rubles / ha against the background of their application; the energy coefficient is 2.65 and 2.75, respectively [17].

In the crop rotation without the presence of leguminous crops, where barley was cultivated on repeated wheat, there was a significant decrease in its yield according to resource-saving processing systems: against the background of natural fertility by 0.09-0.27 t / ha (by 3.8-11.4%) , with the use of fertilizers by 0.07–0.40 t / ha (by 2.0–11.1%). This led to a decrease in net income in the cultivation of barley in comparison with the legume predecessor by 31–44.1% [17]. This decrease is due to a decrease in the content of nitrate nitrogen, mobile phosphorus and potassium in the soil by

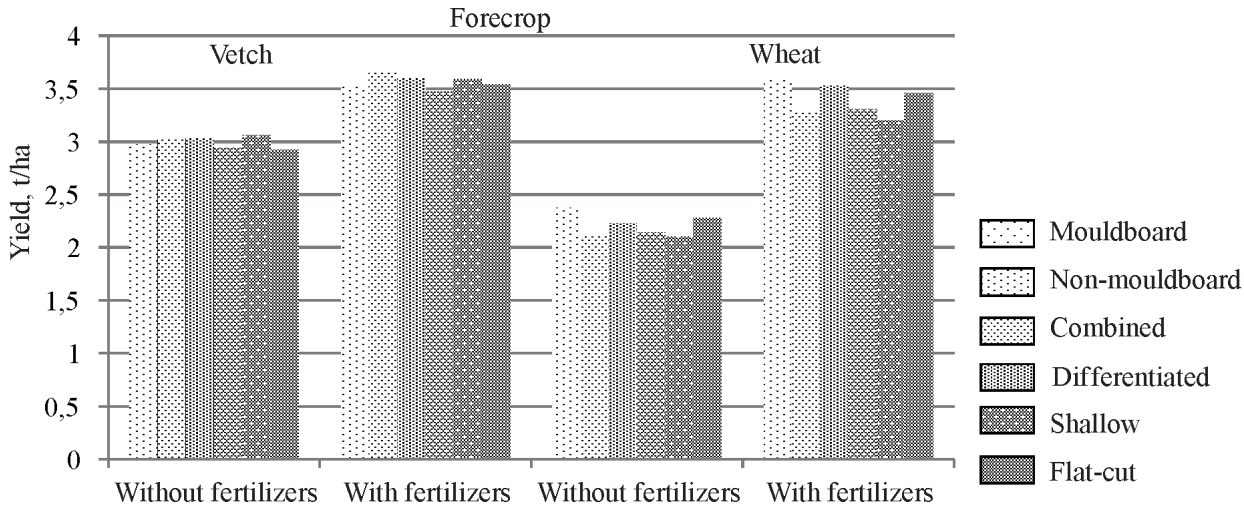

Рис. 3. Урожайность ячменя в среднем за 3–6 ротаций севооборотов
Fig. 3. Barley yield on average for 3–6 crop rotations

non-moldboard and shallow tillage, especially in crop rotation without the presence of leguminous crops. In this case, in the cultivation of barley on recycled wheat, a stable advantage of the dump system was noted, which is explained by more favorable conditions for providing moisture, soil composition and nutrient content.

CONCLUSIONS

1. On the dark gray forest soils of the northern forest-steppe of the Northern Trans-Urals in grain-fallow crop rotation during the cultivation of a grain crop - the fourth crop after steam for the legume predecessor (spring vetch) - it is recommended to use the main processing systems with minimization elements - moldboard-free and combined with moldboard-free loosening by a plow with stands SibIME by 20–22 cm, differentiated with flat-cut processing by 12–14 cm and disking by 10–12 cm. These processing systems after leguminous plants for barley provided conditions of the water regime, soil composition, nutritional regime close to the moldboard system, the formation of an almost equal moldboard system yield processing: against the background of natural fertility - 2.97-3.03 t / ha, with the use of $N_{40}P_{40}P_{40}$ - 3.47-3.65 t / ha, as well as economic efficiency.

2. The use of resource-saving processing systems in grain-fallow crop rotation without leguminous crops for grain crops for repeated wheat led to a decrease in the reserves of productive moisture in a meter layer of soil by 8.6-28.0%, a significant deterioration in the nutritional regime, especially nitrogen (by 15, 5–43.8%) and phosphorus (by 39.1–51.1%), with a negative differentiation of the fertility of the soil profile, a decrease in the yield of barley grain by 0.09–0.40 t / ha, a decrease in net income by 31– 44%.

3. To increase the potential of soil fertility and productivity of cereal crops, completing the link of crop rotation along the moldboard and resource-saving systems of basic processing, it is advisable to place and sow them on the legume predecessor.

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СОДЕРЖАНИЕ ГЛИФОСАТА В ЗЕРНЕ ПРИ ДЕСИКАЦИИ ПОСЕВОВ В ПРИОБЬЕ

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Определены остаточные количества глифосата в зерне пшеницы после десикации посевов. Глифосат – наиболее применяемый в мире гербицид. Уровень его экотоксичности широко обсуждают в научной литературе после отнесения соединения к категории «вероятно канцерогенных» пестицидов. Предполагают, что остаточное количество гербицидов может быть наиболее высоким при десикации посевов перед уборкой. Исследования проведены в 2018 г. (центральная лесостепь Новосибирского Приобья – 54°53'13,5"N, 82°59'36,7"E). В эксперименте десикацию посевов осуществили гербицидом Зеро Супер (содержание изопропиламинной соли глифосата 750 г/кг) в рекомендованной дозе, равной 1,5–2,0 кг/га. Остаточное количество гербицидов в растительной биомассе определяли при помощи тест-системы, основанной на принципе иммуноферментного анализа. Предподготовку растительных проб осуществляли согласно рекомендации фирмы Stylab, проводившей валидацию метода для определения глифосата в зерне. Остаточное количество гербицида обнаружено во всех 37 испытанных образцах. Независимо от срока отбора образцов после десикации показатель в зерне не превышал 4,4 мг/кг. Спустя 14 дней после десикации уровень минимального количества пестицида был ниже (0,5 мг/кг) в сравнении с данными, полученными спустя 7 дней (2,6 мг/кг). Содержание остаточного количества гербицидов в соломе пшеницы оказалось выше, чем в зерне. Наиболее высокие значения показателя обнаружены в биомассе сорной растительности, высохшей при десикации (до 9 мг/кг). В зерне, хранившемся один год, содержание остаточного количества гербицидов не снизилось. Полученные данные сравнили с уровнями ПДК остаточного количества гебицидов в зерне пшеницы, принятыми в мире.

Ключевые слова: глифосат, остаточные количества, зерно пшеницы, Приобье

GLYPHOSATE RESIDUES IN GRAIN AFTER DESICCATION OF CROPS IN THE OB REGION

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The residual amounts of glyphosate in wheat grain after desiccation of crops were determined. Glyphosate is the most widely used herbicide in the world. The level of its ecotoxicity is widely discussed in the scientific literature after the compound was classified as “a likely carcinogenic” pesticide. It is assumed that glyphosate residues can be highest when the crops are desiccated before harvesting. The studies were carried out in 2018 (central forest-steppe of the Novosibirsk Ob region (54°53'13.5"N, 82°59'36.7"E). In the experiment, desiccation of wheat was carried out with the herbicide Zero Super (the content of isopropylamine salt of glyphosate was 750 g/kg) at the recommended dose of 1.5– 2.0 kg/ha. Glyphosate residues in plant biomass were determined using the test system, based on the principle of enzyme immunoassay. Pre-preparation of the samples for the analysis was carried out according to the recommendation of Stylab laboratory, which validated the method for the determination of glyphosate in grain. Glyphosate residues were found in all of the 37 samples tested. Regardless of the sampling period after desiccation, herbicide residues in the grain did not exceed 4.4 mg/kg. In 14 days after desiccation, the minimum pesticide level was lower (0.5 mg/kg) compared to the data obtained in 7 days (2.6 mg/kg). The content of the herbicide residues in wheat straw was higher than in grain. The highest values of glyphosate residues were

found in the dry biomass of weed vegetation (up to 9 mg/kg). In the grain stored for 1 year, the content of GR has not decreased. The data obtained were compared with the MRL for the residual amount of glyphosate in wheat grain accepted in the world.

Keywords: glyphosate, residual amount, wheat grain, the Ob region

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INTRODUCTION

Obtaining stable output yields to provide the growing population of the planet with a sufficient amount of food and raw materials in the face of dwindling land resources is impossible without the use of plant protection chemicals, which is fraught with environmental pollution with residual amounts of pesticides [1]. To solve this problem, the world community is taking comprehensive measures: it searches for new formulas of chemicals with a low dose of use, improves approaches and methods for controlling the residual amount of pesticides in environmental objects, develops new technologies for cleaning contaminated areas, etc. [2–4].

The agrarian sector of the Siberian Federal District is in line with the global trend towards the built-up of crop protection chemicals. At the same time, the assessment of the safety of the pesticides used is based on the recommendations of the manufacturers, developed for areas with more favorable climatic conditions in comparison with Siberia. In this regard, it becomes necessary to study the features of the degradation of pesticides in crop products and environmental objects in the specific conditions of a short, often dry growing season in Siberia. The Novosibirsk Region is one of the largest grain producers in the Siberian Federal District. A significant part of the production on farms is obtained with the help of intensive technologies for growing crops, and this tendency in the development of agriculture will only intensify. In addition, in recent years, agricultural technologies have been increasingly actively mastered, implying a partial or complete abandonment of mechanical soil cultivation (direct sowing system, no-till, etc.), which requires an increased use of pesticides. One of the most widely used pesticides in such agricultural technologies are continuous herbicides based on glyphosate. In the Russian Federation, herbicides based on this compound account for 1/3 of the volume of applied herbicides and are represented by more than 40 names [5].

Glyphosate is the active ingredient in a spectrum of non-selective systemic herbicides, first patented as commercial products such as Roundup® (Monsanto Co) or Touchdown® (Syngenta Co) in 1974. Since then, these herbicides have become one of the most widely used in the world and their consumption rates are increasing [6, 7]. An increase in the sowing of transgenic crops resistant to this group of herbicides and an increase in the resistance of weeds contribute to the expansion of the market for herbicides based on glyphosate [8, 9].

The high ecotoxicity of most of the pesticides has been experimentally proven. Glyphosate (N- (phosphonomethyl) glycine) is probably no exception. In 2015, the WHO Agency for Research on Cancer (IARC) classified glyphosate as a “possibly carcinogenic” pesti-

cide[1]. The IARC announcement attracted public and scientific attention around the world. The European Union has recommended that the use of glyphosate as a desiccant be restricted and its use in public gardens and parks must be stopped[2]. Public movements have formed for a complete ban on the use of the drug in the agricultural sector. Since the publication of the document (see footnote 1), a lot of research has been done on various aspects of the problem. A great deal of very contradictory data has been obtained. In general, the authors believe that the number, duration and the design of studies are insufficient for final conclusions so far [8, 9]. To date, the use of glyphosate in the EU is allowed only until 2022, a number of countries (Belgium, Colombia, the Netherlands, Sri Lanka, France, many countries of the Middle East) have legally restricted the use of glyphosate in agriculture [10, 11].

However, the appearance of glyphosate at one time made it possible to move to a new stage in the development of soil-protective and resource-saving agriculture in the world. At present, it is difficult to imagine the agricultural sector without this drug. In recent years, possible parameters for food production have been studied if the use of glyphosate is prohibited. Calculations have shown that in this case, the gross harvest of soybeans, corn, rapeseed in the world may decrease by 18.6; 3.1; 1.5 million tons per year, respectively. To compensate for these losses, an increase in the arable land area by 0.76 million hectares will be required. At the same time, the cost of weed control will increase by $ 20-30. US / ha. The use of alternative herbicides will not lead to a decrease in the pesticide load on agrocenoses [12, 13].

As our brief review shows, despite the ecotoxicity, agriculture is unlikely to be able to stop using glyphosate. There is only one way out of the situation - strict control of the residual amount of the pesticide in the environment, primarily in products, in order to minimize its entry into the food chain.

The aim of the study is to determine the value of the residual amount of glyphosate in grain and wheat straw after desiccation of crops in the recommended doses and timing of harvesting in the Ob region, to clarify the degree of its decomposition during grain storage throughout the year.

MATERIALS AND METHODS

The studies were carried out in 2018 on the territory of the scientific and experimental base of the Siberian Research Institute of Agriculture and Chemicalization of the Siberian Federal Scientific Center of the Russian Academy of Sciences (central forest-steppe of the Novosibirsk Ob region, 54° 53'13.5 "N, 82° 59'36.7" E). Desiccation of spring wheat crops was carried out on September 3 with the herbicide Zero Super (content of isopropylamine salt of glyphosate 750 g / kg) at the recommended dose of 1.5–2 kg / ha[3]. The wheat was Novosibirskaya 31. The moisture content of grain before desiccation was 25–28%. The sprayer OP-2000. The consumption rate of the working solution is 270 l / ha. Harvesting took place 21 and 25 days (in 2017 and 2018, respectively) after desiccation. The sampling was carried out by the sheaf method (1 m^2) in triplicate on the plot and three repetitions of the experiment (9 sheaves). In the period after desiccation, the air temperature was close to the average multiyear norm. Precipitation in 2017 (harvesting 09.09.2017) 20 mm with an average annual rate of 33 mm, in 2018 (harvesting 09.13.2018) - 60 mm with a rate of 40 mm.

Grain, straw and biomass of weeds dried up under the influence of the preparation were analyzed. To determine the possibility of glyphosate decomposition during storage of

[1]IARC Monograph on Glyphosate (https://www.iarc.fr/en/.../iarcnews/.../glyphosate_IARC2016).

[2]Directive 2009/128/EC, 2009b (Article 4) of the European Parliament and of the Council of 21 October 2009. Regulation (EC) No 1107/2009 of the European Parliament and of the council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC https://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ: L: 2009:309:0001:0050: EN:PDF.

[3]State catalog of pesticides and agrochemicals approved for use on the territory of the Russian Federation.

grain products, grain of the 2017 harvest was analyzed, which was stored for a year in a cool and dry room. The residual amount of glyphosate was determined using the Abraxis Glyphosate Plate ELISA test system. The preparation of plant samples was carried out according to the recommendation (procedure R091313) of the Stylab company (Moscow), which validated the method for the determination of glyphosate in grain (www Stylab.ru). Samples of the samples were derivatized, then added together with antibodies specific to glyphosate into the wells of a microtiter plate, coated with anti-species antibodies to antibodies to glyphosate, and incubated for 30 min. The enzyme conjugate glyphosate was then added. At this point, a competitive reaction occurs between glyphosate, which may be contained in the sample, and the labeled glyphosate enzyme for binding sites with antibodies on the plate. After washing and adding the substrate (color solution), a colored signal (blue color) develops. The presence of glyphosate is detected by the addition of a colored solution that contains an enzyme substrate (hydrogen peroxide) and a chromogen (3,3`, 5,5`-tetramethylbenzidine). The antibody-bound glyphosate-enzyme conjugate catalyzes the conversion of the substrate / chromogen mixture into blue-colored reaction products. After incubation, the reaction was stopped and stabilized by adding dilute acid (stop solution). Since there was competition between labeled glyphosate (conjugate) and unlabeled (sample) for binding to antibodies, the color intensity is inversely proportional to the concentration of glyphosate in the sample.

RESULTS AND DISCUSSION

Residual amounts of glyphosate were found in all 37 samples tested. Regardless of the time of sampling after desiccation in grain, it did not exceed 4.4 mg / kg (see Table 1). At 14 days after desiccation, the minimum amount level was lower (0.5 mg / kg) compared to the data obtained after 7 days (2.6 mg / kg). The residual amount of glyphosate in wheat straw was found to be higher than in grain. The highest values of the indicator were found in the biomass of weeds dried up during desiccation.

In grain stored for a year in a cool dry room, the residual amount of glyphosate was slightly higher compared to the data obtained in the analysis of grain from the 2018 harvest.

For the first time for Siberia, data were obtained on the content of the residual amount of herbicides in wheat grain after desiccation with the recommended doses of herbicide.

It should be noted that the MRL (maximum residual level) or MRL for the residual amount of glyphosate in unprocessed wheat grain, varies significantly across the world. In the USA this value is taken equal to 30 mg / kg, in the EU - 10, in Canada - 5 mg / kg 4. This is probably due to the different background value of the content of the residual amount of pesticide in the product. As noted by the authors of the review [9], these limits are constantly growing,

Табл. 1. Содержание остаточного количества глифосата в надземной биомассе зернового агроценоза после десикации посевов

Table 1. Concentration of glyphosate residues in the aboveground biomass of grain agrocenosis after desiccation of crops

Sample selection year	*n*	The day after desiccation	Herbicide dose, kg / ha	Substrate	Glyphosate content, mg / kg	
					lim	average
2018	7	7	1,5	Grain	2,6–4,4	3,6
	12	14	1,5	»	0,5–3,9	2,5
	6	14	1,5	Straw	5,5–6,0	5,7
	6	14	1,5	Weeds	8–9	8,5
2017	6	After a year of storage	2	Grain	4,1–5,4	4,2

[4]BCGlobal Pesticide MRL Database https://www.bryantchristie.com/BCGlobal-Subscriptions/Pesticide-MRLs

apparently following an increase in the content of contaminants in products. In the Russian Federation, the MRL for herbicide residues is 20 mg / kg of grain[5]. It is difficult to understand where this figure came from. This is probably the calculated average MRL value adopted in the USA and the EU, since we did not find published information on the content of the residual amount of glyphosate in grain for the Russian Federation in the available literature. According to the above criteria, the content of the residual amount of herbicides in the grain in our experience is within acceptable limits. Based on the decrease in the minimum detectable amount of glyphosate residues in the grain, the waiting time of 14 days after desiccation is significant.

As noted by the authors of a review published in 2019 [11], there is very little documented information in the scientific literature on herbicide residues in cereal products, particularly wheat grain, after desiccation. The authors found only four sources published between 2013 and 2018 (see Table 2). From these data, it appears that herbicide residue levels in grain after desiccation exceeded the MRL adopted in Canada in one case out of four (the study was conducted in Canada). For the territory of the former USSR, data for Ukraine are available [14]. In Kiev region in 2002-2011, residual amounts of herbicides in grains of three wheat varieties during desiccation of crops with 3 l/ ha of isopropyl salt of glyphosate (480 g/l r.a.) were determined. Depending on the wheat variety the indicator in the grain was 0.5-2.5 mg/kg. In Table 2 we gave data on changes in the content of herbicide residue in processed grain products. The bulk of it remains in bran. In a special experiment by Canadian researchers [15], the proportion of herbicide residue in bran was 81% of the original content in the grain. Nutritionists usually suggest bran as a component of a healthy diet. It follows that this side of the issue should be kept in mind.

On the whole, the impression is created that the situation with the studied indicator in grain after desiccation is quite favorable. However, the question arises why the FAO / WHO MRL for glyphosate residues in wheat grain is 30 mg / kg.

If there is a lot of data on the dynamics of herbicide residues during grain processing, then there is very little information on the change in the pesticide content during grain storage [11]. We have received information on this issue. Our conclusion is based on the assumption that strict adherence to the same experimental conditions in both years of research resulted in an approximately similar level of herbicide residues in the grain before harvesting. When storing grain during the year, this indicator did not decrease. This is probably due to the well-known fact that glyphosate is well fixed in plant biomass, as a result of which the rate of its metabolism decreases [16]. Information on the herbicide residues in straw and weeds is important, since, as follows from our data, these components of the biomass can serve as a significant concentrator of pesticides in the agrocenosis.

CONCLUSIONS

1. Residual amounts of glyphosate (up to 4.4 mg / kg) were found in all samples of wheat from crops subjected to desiccation.

2. When storing contaminated grain for a year in a dry and cool room, there was no decrease in the residual amount of glyphosate in the grain.

Табл. 2. Содержание глифосата в зерне пшеницы до и после переработки [11]

Table 2. Concentration of glyphosate in wheat grain before and after processing [11]

Glyphosate content in the source grain, mg / kg	Grain processing products	
	Product type	Glyphosate content, mg / kg
Less than 0,13	Bran	0,7
0,67	Flour	0,02
1,07–1,13	Bread	0,001–0,0458
6,1–11,1	Baked goods	0,001–0,0179

[5]Hygienic standards for pesticide content in environmental objects (list) GN 1.2.3538-18 // Bulletin of regulatory and methodological documents of RSES. 2019. Issue 3 (77). Pp. 7-103.

3. A significantly higher content of the residual amount of glyphosate in straw and weed biomass (up to 9 mg / kg) was found in comparison with grain. These plant residues can be a significant source of pesticide pollution.

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ПОКАЗАТЕЛЬ ПРОНИЦАЕМОСТИ КЛЕТОЧНЫХ МЕМБРАН ПРОРОСТКОВ В ОЦЕНКЕ СТРЕССОУСТОЙЧИВОСТИ СОРТОВ ПШЕНИЦЫ

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Представлены результаты исследований применения показателя проницаемости клеточных мембран, определяемого по относительному изменению удельной электропроводности (УЭП) водных вытяжек тканей, проростков сортов яровой пшеницы Новосибирская 18, Новосибирская 44, Сибирская 21 и Омская 18 при совместном действии стрессоров. В модельных лабораторных вегетационных опытах исследована почасовая динамика УЭП водных вытяжек листьев проростков (экспозиция листьев в воде 0,5–4,5 ч) в условиях одновременного действия хлоридного засоления (1,3%) и возбудителя обыкновенной гнили злаков *Bipolaris sorokiniana* Shoem. (5000 конидий на зерно). Установлено достоверное увеличение УЭП в 1,5 раза и скорости выхода электролитов в 2 раза у менее устойчивого сорта Новосибирская 44 по сравнению с более устойчивым Омская 18. Исследована посуточная динамика УЭП 10–16-суточных проростков в условиях последовательного действия гипертермии семян (43 °С), хлоридного засоления (1,3%) и *Bipolaris sorokiniana* Shoem. (5000 конидий на зерно). Установлен протекторный эффект гипертермии у более устойчивого сорта Сибирская 21 (достоверное снижение УЭП до 1,3 раза) по сравнению с вариантом без прогрева семян. У менее устойчивого сорта Новосибирская 18 прогрев семян дестабилизировал состояние клеточных мембран (достоверное увеличение УЭП и скорости выхода электролитов в 1,5 и 1,2 раза соответственно). Экспериментально определены условия, обеспечивающие выявление максимальных различий исследуемых сортов пшеницы: возраст проростков – 10 сут, временной интервал экспозиции образцов в воде – 1,5–4,5 ч. Межсортовые различия по относительному изменению значений УЭП в варианте без прогрева семян составляли 1,9 раза и в варианте с прогревом семян – 3,7 раза с достоверностью различий на уровнях $p \leq 0,05$ и $p \leq 0,01$. Межсортовые различия по относительному изменению УЭП, установленные на интервале времени экспозиции выхода электролитов 1,5–4,5 ч, составляли 1,50–1,67 раза с достоверностью различий на уровне $p \leq 0,05$. Предложенный подход позволит разработать методику оценки новых генотипов на устойчивость к совместному действию биотических и абиотических стрессоров.

Ключевые слова: сорт, пшеница, устойчивость, стрессоры, проницаемость клеточных мембран, удельная электропроводность

THE INDICATOR OF CELL MEMBRANE PERMEABILITY OF WHEAT SEEDLINGS IN ASSESSING STRESS RESISTANCE OF WHEAT VARIETIES

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The paper presents the results of studies on the use of the permeability index of cell membranes, determined by the relative change in the specific electrical conductivity (EC) of water extracts of

tissues of spring wheat seedlings, varieties Novosibirskaya 18, Novosibirskaya 44, Sibirskaya 21 and Omskaya 18, under the combined action of stressors. In model laboratory vegetation experiments, the hourly dynamics of the EC of water extracts of seedling leaves (exposure of leaves to water for 0.5–4.5 h) was investigated under the simultaneous action of chloride salinity (1.3%) and the causative agent of common rot of cereals *Bipolaris sorokiniana* Shoem. (5000 conidia per grain). It was established that EC increased by 1.5 times and the rate of electrolyte leakage increased twofold in the less resistant variety Novosibirskaya 44 compared to the more resistant Omskaya 18. The daily dynamics of the EC of 10–16-day-old seedlings was studied under the sequential action of seed hyperthermia (43 °C), chloride salinity (1.3%), *and Bipolaris sorokiniana* Shoem. (5000 conidia per grain). The protective effect of hyperthermia was established in the more resistant variety Sibirskaya 21 (a decrease in EC up to 1.3 times) in comparison with the variant without heating the seeds. In the less resistant variety Novosibirskaya 18, heating the seeds destabilized the state of the cell membranes (increase in EC and electrolyte leakage rate by 1.5 and 1.2 times respectively). The conditions that ensure the identification of the maximum differences in the studied wheat varieties were experimentally determined: the age of seedlings 10 days, the time interval of exposure of the samples to water 1.5–4.5 h. Intervarietal differences in the relative change in the EC values in the variant without heating the seeds were 1.9 times and in the variant with heating the seeds – 3.7 times, with the significance of difference at the levels $p \leq 0.05$ and $p \leq 0.01$. Intervarietal differences in the relative change in the EC, established for the time interval exposition of electrolytes leakage of 1.5–4.5 h, were 1.50–1.67 times with the significance of difference at the level of $p \leq 0.05$. The proposed approach will make it possible to develop a methodology for assessing new genotypes for resistance to the combined action of biotic and abiotic stressors.

Keywords: wheat, variety, resistance, stressors, permeability of cell membranes, electrical conductivity

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INTRODUCTION

In crop variety breeding it is necessary to diagnose their resistance to biotic and abiotic stress factors of the environment. Performing such an assessment in the field by a direct method (by yield depression) is a laborious and time-consuming stage. In this regard, the assessment of varieties is carried out in laboratory conditions by indirect methods to change the physiological, biochemical and biophysical parameters, which reflect the process of plant adaptation to stress [1, 2]. The advantages of indirect assessment include high information content, the ability to predict a decrease in productivity under stress at the early stages of plant ontogenesis, the independence of diagnostic indicators from environmental conditions, and the possibility of automated assessment [3].

For the most part, the diagnostic methods used make it possible to assess genotypes for resistance to one acting stressor. However, in natural conditions, plant organisms are exposed to a complex of environmental stress factors in various combinations and doses at different stages of plant development. Therefore, it is important to conduct research on the resistance of crops to stressful influences, simulating their cumulative effect [4]. In Siberia, among other stressors limiting the yield, soil salinity, com-

mon root rot, and elevated air temperatures should be taken into account [5].

The main plant responses to abiotic and biotic factors are associated with the occurrence of oxidative stress, which disrupts the structure and function of cell membranes. This leads to complete cell death, inhibition or arrest of growth [6, 7]. The biological membrane serves as a primary protective barrier and provides a nonspecific component of adaptation mechanisms under the action of stress factors [8].

The most important property of biological membranes is permeability. It determines the stability of plant tissues and cells and characterizes the static, genetically determined potential resistance of the genotype [9, 10]. The higher the resistance of plants, the less the structure and properties of membranes are disturbed, and the release of electrolytes from plant tissues into the external environment decreases [11]. Changes in membrane permeability for electrolytes is one of the criteria for plant resistance to biotic and abiotic stressors. The reason for the increase in the permeability of cell membranes under stress may be the H^{+} / Ca^{2+} ratio in the membranes, a decrease in the level of SH-groups, the formation of defective regions in lipids, and an increase in the activity of endogenous phospholipases[1].

The permeability of cell membranes is determined by the conductometric method by measuring the specific electrical conductivity of electrolytes released through damaged cell membranes from plant tissues and organs into distilled water, which can be a diagnostic indicator of the stress resistance of the variety.

The results of studies on changes in the permeability of cell membranes under the action of low temperatures on plants [12, 13], drought [14, 15], infection with a pathogen [16, 17], biologically active substances [18, 19], heavy metals [20] were obtained.

The method also turned out to be in demand in studying the response of plants to the separate and combined action of stressors: low temperature and cadmium ions [21], heat shock and water deficit [22], unfavorable temperature and heavy metals [23], salinity and zinc [24].

Earlier, we experimentally proved the possibility of taking into account the change in the permeability index of cell membranes, assessed by the specific electrical conductivity (EC) of seedling tissues. On the basis of this, diagnostic methods have been developed to assess the resistance of spring wheat and barley varieties to chloride salinization and the causative agent of common rot of cereals[2, 3].

The aim of the research is to evaluate the specific electrical conductivity of aqueous extracts of leaves as an indicator of the permeability of cell membranes of seedlings to determine the stress resistance of wheat varieties to the combined action of the causative agent of root rot of cereals, chloride salinization, and hyperthermia of seeds.

MATERIAL AND METHODS

The experimental work was carried out in the laboratory for the study of physical processes in agrophytocenoses of the Siberian Institute of Physics and Technology, SFNCA RAS. The studies were carried out in laboratory conditions (vegetation experiment - water crops) on seedlings of zoned varieties of soft spring wheat Novosibirskaya 18, Novosibirskaya 44, Sibirskaya 21 of the Siberian Research Institute of Plant Production, a branch of the ICG SB RAS, and Omsk 18 of the selection of the Omsk ASC.

The study of the permeability index of seedling cell membranes was carried out to assess the resistance of varieties to the combined action of the common rot pathogen *Bipolaris sorokiniana* Shoem. (*B. sorokiniana*), chloride sa-

[1]Chirkova T.V. Physiological foundations of plant resistance: a textbook. SPb.: Publishing house S.-Pb. University, 2002.244 p.

[2]Patent RU 2446671 IPC A01G7 / 00, A01H1 / 04. The method for determining the relative resistance of soft spring wheat varieties to chloride salinity / T.A. Gurova, V.Yu. Berezina, N.S. Kutserubova. Publ. 10.04.2012.

[3]Patent RU 2625027 IPC A01C12N 1/14, A01H 5/12. The method for determining the relative resistance of soft spring wheat varieties to the causative agent of common root rot of cereals / T.A. Gurova, V.V. Alt, O.S. Lugovskaya. Publ. 11.07.2017.

linization and hyperthermia of seeds. The hourly and daily dynamics of the EC of the extracts of seedlings leaves of wheat cultivars differing in their resistance to these stressors was studied. Preliminarily, the resistance of varieties was assessed in laboratory conditions according to growth parameters, changes in dry and wet biomass, and development of the disease on seedlings [25].

The hourly dynamics of changes in the leaf extracts EC (kinetics of electrolyte release after exposure of leaf tissue in distilled water for 0.5–4.5 h) was recorded in 10-day-old seedlings of varieties Omskaya 18 (relatively stable) and Novosibirskaya 18 (relatively unstable). The stressors acted simultaneously.

Experiment options:

– control (stress-free seeds),

– infection of seeds with *B. sorokiniana* (5000 conidia per grain) + chloride salinity of 1.3%.

The daily dynamics of EC was recorded in 10–16 day old seedlings of varieties Sibirskaya 21 (relatively resistant) and Novosibirskaya 18 (relatively unstable). A sequential action of stressors was used - preliminary heating of seeds, followed by infection with *B. sorokiniana* and chloride salinization.

Experiment options:

– control (seeds without heating) and heating of seeds at 43 ° C;

– seeds without heating + infection with *B. sorokiniana* (5000 conidia per grain) + chloride salinity 1.3%;

– seed heating + *B. sorokiniana* infection (5000 conidia per grain) + chloride salinity 1.3%.

The levels of stress loads (conidial suspension of *B. sorokiniana* 5000 conidia per grain, concentration of sodium chloride (NaCl) 1.3% and temperature 43 ° C) were determined by us in specially conducted vegetation experiments. These levels make it possible to differentiate wheat varieties of Siberian selection when assessing their resistance to these stress factors (see footnotes 2 and 3) [26].

Wheat seeds were pre-sterilized with 96% ethyl alcohol for 2 min, followed by three rinsing with distilled water. The seeds were heated for 20 min in hot water in a water bath according to the VIR[4] method. After cooling, the seeds were placed in Petri dishes with moistened filter paper and germinated in a thermostat at 22 °C for three days. Simultaneously, the soaked seed samples were germinated without heating. Seed infection was carried out in the germination phase (on the third day of cultivation) with a conidial suspension of a mixture of medium pathogenic isolates of *B. sorokiniana* prepared on 0.1% aqueous agar (one drop per grain). Then the plants were grown in a roll culture on tap water (control option) and sodium chloride (*B. sorokiniana* infection + chloride salinization option) in a Biotron-8 climatic chamber with a day-night photoperiod of 16 and 8 h, respectively, illumination 20,000 lx (day), temperature 22 ºC and humidity 60%.

To measure the EC, the first leaves of the seedlings were removed, washed with tap water, and dried with filter paper. An average sample was prepared in each variant. For this purpose, cuts were made from the middle part of the leaves with a blade, they were placed in a bag made of nylon cloth, washed with distilled water, and then dried with filter paper. Samples of plant tissue were placed in a glass cup with distilled water at a certain tissue - water ratio. The cups were kept under illumination conditions for at least 1.5 h. The specific electrical conductivity of aqueous extracts of leaves was measured on an edge EC conductometric device, HANNA Instruments (Germany). The response of the cultivar was determined by the relative change in the UEP of water extracts of the leaves of seedlings after exposure of plants to stressors. The smaller the change in the parameter, the more resistant the variety[5].

[4]Diagnostics of plant resistance to stress effects: guidelines / ed. G.V. Udovenko. L., 1988.228 p.

[5]Gurova T.A., Denisyuk S.G., Lugovskaya O.S., Svezhintseva E.A., Mineev V.V. Methodological provisions for early diagnosis of the resistance of spring wheat and barley to the combined action of stressors. Novosibirsk: SFSCA RAS, 2017.62 p.
ностью различий на уровне $p \leq 0,05$.

Analytical repetition of experiments is 6-8 times, biological - 3 times. Representative sample - 200 seedlings in each variant of the experiment. The experimental data were mathematically processed using standard statistical programs. To determine the significance of differences in mean values, the Student's t-test was used. Regression analysis was carried out to reveal the relationship between the recorded parameters of varieties and their resistance. The average error did not exceed 3–5%. Three series of experiments were carried out.

RESULTS AND DISCUSSION

A study of the hourly dynamics of the release of electrolytes. One of the most essential requirements for a diagnostic method is its differentiating ability. It is determined by the reliability of the assessment of the resistance of closely related objects (varieties of the same crop, plants from the same varietal population) to various stress effects. Therefore, the choice of regimes (conditions) that would provide the greatest amplitude of the manifestation of varietal differences is necessary in the experiment.

The study of electrolyte release kinetics depending on the duration of leaf tissue exposure to water is an important methodology for establishing the time of maximum inter-varietal differences in the assessment of variety stress tolerance. The EC measurements showed a different pattern of electrolyte release from leaf tissue of wheat seedlings into distilled water under simultaneous action of chloride salinity and cereal root rot pathogen (see Fig. 1).

The release of electrolytes in seedlings of both varieties in the control variants did not differ significantly. In the experimental variant, the electrolyte yield in seedlings of the relatively resistant cultivar Omskaya 18 stabilized at a lower level of the EC value compared to the relatively unstable cultivar Novosibirskaya 44. The rapid release of electrolytes in the first 0.5–1.5 h of exposure of the samples to distilled water was replaced by a weaker one. but with a stable output over the next three hours (exposure time 1.5–4.5 h). Thus, on the curve of the total electrolyte yield, two sections can be distinguished, characterizing the time intervals of various processes (see Fig. 2).

Section 1 of the graph (exposure time 0.5–1.5 h) characterizes the release of electrolytes from the apoplast, in which ions move in accordance with the laws of diffusion and adsorption [27]. Section 2 of the graph (exposure time 1.5–4.5 h) differs in a smaller angle of inclination of the linear approximation than section 1, and reflects the functional activity of the plasma membrane, its resistance to diffusive penetration of electrolytes.

Consequently, the time interval for the release of electrolytes from the free space (apoplast) and cytoplasm (through the plasmalemma), which indirectly indicates the state of the plasmalemma, is 1.5–4.5 hours. The indicator of the relative change in the EC for the unstable variety Novosibirskaya 44 significantly exceeds (with an exposure of 1.5 h - 1.5 times) the indicator of the relative change in the EC for the resistant cultivar Omskaya 18. The slope of the linear approximation (the rate of electrolyte release) for the unstable cultivar Novosibirskaya 44 significantly exceeds (more than 2 times) the rate of electrolyte release in seedlings of the resistant cultivar Omskaya 18. Intervarietal differences established by exposure times 1.5; 3.0 and 4.5 hours were 1.5; 1.66 and 1.67 times, respectively, with the significance of differences at the level of $p \leq 0.05$.

Thus, the maximum differences between the studied varieties of spring wheat were revealed in the time interval of the experiment, 1.5–4.5 h. The time of exposure of the samples in water was established (at least 1.5 h). With a shorter exposure period, electrolytes are released only from the free space (apoplast). An exposure time of more than 4.5 hours increases the duration of the sample evaluation procedure and reduces the quality of analyzes. The more resistant cultivar Omskaya 18 has the smallest indicators of the relative change in the EC and the rate of release of electrolytes. The smaller the relative change in EC, the more resistant the cultivar to the combined effect of the conidial suspension of *B. sorokiniana* (5000 conidia per grain) and 1.3% chloride salinity.

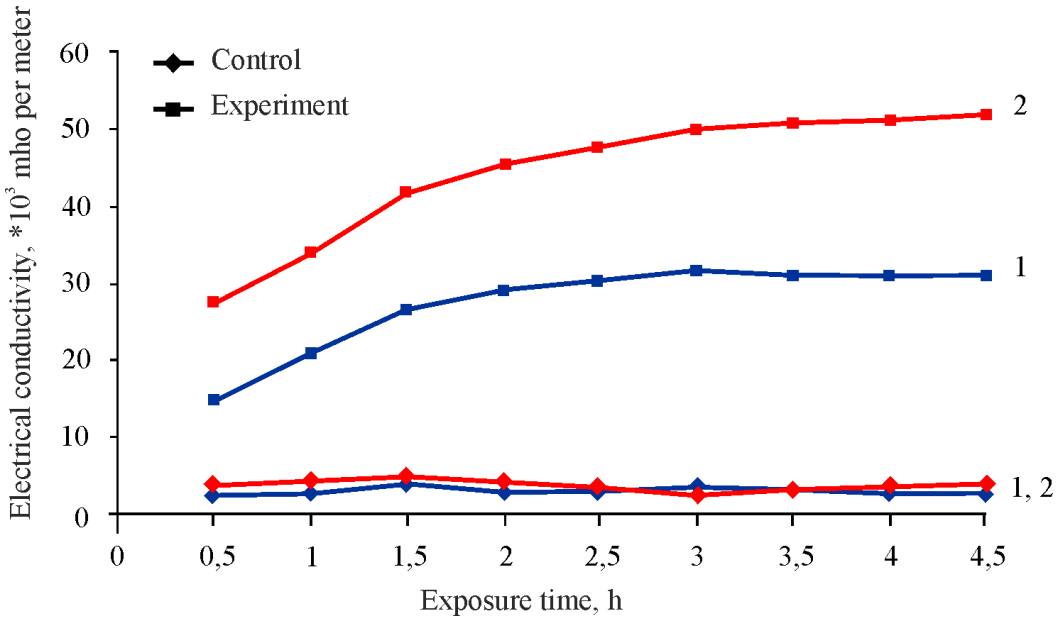

Рис. 1. Удельная электропроводность водных вытяжек листьев пшеницы в зависимости от длительности экспозиции листовой ткани в воде (стрессоры – конидиальная суспензия *B. sorokiniana* – 5000 конидий на зерно + хлорид натрия 1,3%): *1* – сорт Омская 18 (относительно устойчивый), *2* – сорт Новосибирская 44 (относительно неустойчивый). Различия с контролем достоверны на уровне $p \leq 0{,}01$

Fig. 1. Electrical conductivity of water extracts of wheat leaves depending on the duration of exposure of leaf tissue to water (stressors – conidial suspension of *B. sorokiniana* – 5000 conidia per grain + sodium chloride 1.3%): *1* – variety Omskaya 18 (relatively resistant), *2* – variety Novosibirskaya 44 (relatively non-resistant). Differences with control are reliable at the level of $p \leq 0.01$

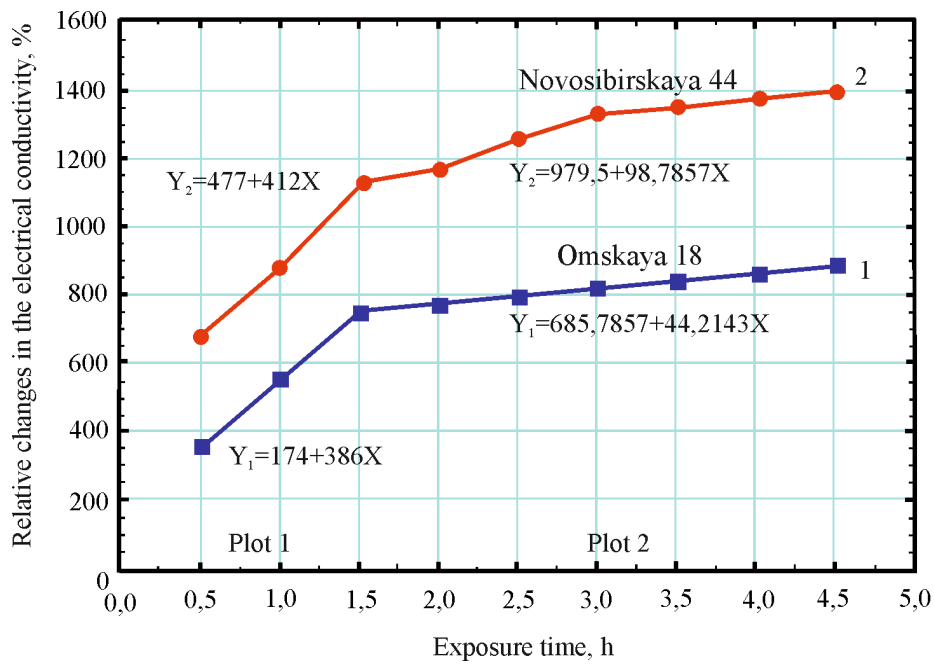

Рис. 2. Линейная аппроксимация относительного изменения удельной электропроводности водных вытяжек листьев пшеницы от длительности экспозиции листовой ткани в воде: *1* – сорт Омская 18 (относительно устойчивый); *2* – сорт Новосибирская 44 (относительно неустойчивый)

Fig. 2. Linear approximation of the relative change in the electrical conductivity of water extracts of wheat leaves on the duration of exposure of leaf tissue to water: *1* – variety Omskaya 18 (relatively resistant); *2* – variety Novosibirskaya 44 (relatively non-resistant)

A study of the daily dynamics of the release of electrolytes. The daily dynamics of the leaf extracts EC was studied in 10–16-day old seedlings of wheat cultivars Sibirskaya 21 (relatively resistant) and Novosibirskaya 18 (relatively unstable). The experiment was carried out without preliminary heating and with heating of seeds under the combined action of the following stressors: conidial suspension of *B. sorokiniana* and sodium chloride. The primary experimental data are presented in Table 1. The results obtained are approximated graphically in the form of quadratic dependences (see Fig. 3). The coefficients of quadratic equations for each variant of the experiment, respectively, are presented in Table 2. Analysis of the obtained experimental data on the study of daily dynamics and the corresponding approximating quadratic dependences made it possible to reveal the following features.

The value of the relative change in the EC sharply increases in 12-day-old seedlings with a subsequent increase in the indicator during cultivation for 16 days in both varieties in the variants with and without heating the seeds (see Table 1 and Fig. 3). In the variant without heating the seeds, the indicator of the relative change in the EC in seedlings of the Novosibirskaya 18 variety over the entire range of studies reliably ($p \leq 0.05$) exceeds the EC indicator for the resistant cultivar Sibirskaya 21 (almost 2 times on the 10th day of seedling cultivation). Preliminary heating of seeds followed by the action of two stressors (*B. sorokiniana* and sodium chloride) led to the destabilization of cell membranes and a significant ($p \leq 0.05$) increase in the EC of seedlings of Novosibirskaya 18 variety (relatively unstable). In the cultivar Sibirskaya 21, the seeds heating stimulated the formation of protective and adaptive reactions, which was reflected in a decrease in the EC for the entire period of cultivation of seedlings of this cultivar. One of the possible reasons for such a combined tolerance in seedlings of the Sibirskaya 21 cultivar is the ability of plants to adapt existing cross reactions under the action of stressors [28].

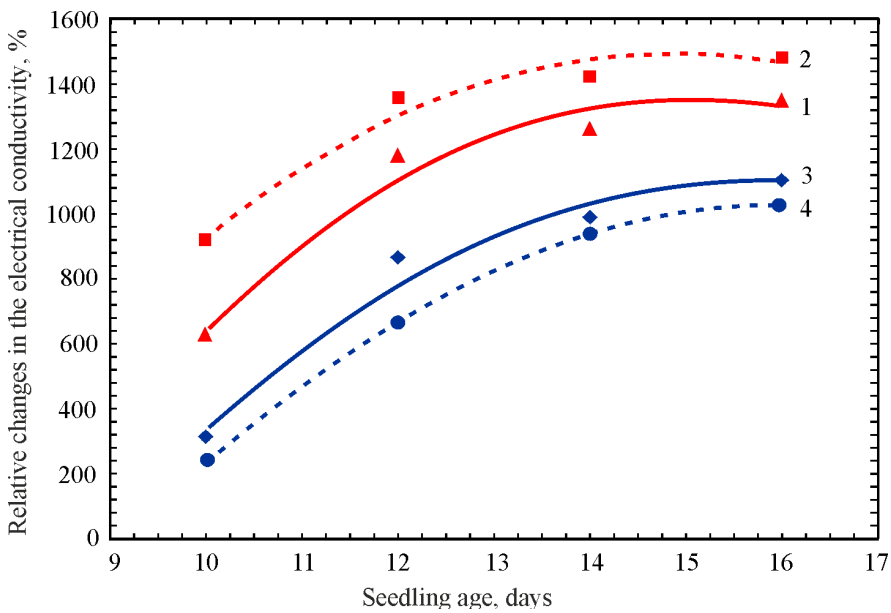

Рис. 3. Зависимость относительного изменения УЭП от возраста проростков при совместном действии стрессоров (конидиальная суспензия *B. sorokiniana* + хлорид натрия 1,3%): *1* – сорт Новосибирская 18, без прогрева семян; *2* – сорт Новосибирская 18, с прогревом семян; *3* – сорт Сибирская 21, без прогрева семян; *4* – сорт Сибирская 21, с прогревом семян

Fig. 3. Dependence of the relative change in EC on the age of seedlings under the combined action of stressors (conidial suspension of *B. sorokiniana* + chloride salinity 1.3%): *1* – Novosibirskaya 18, without seed heating; *2* – Novosibirskaya 18 variety, with seed heating; *3* – Sibirskaya 21, without seed heating; *4* – Sibirskaya 21, with seed heating

Табл. 1. Изменение удельной электропроводности сортов пшеницы в зависимости от возраста проростков при действии комплекса стрессоров, УЭП × 10^{-3} См/м

Table 1. Changes in the specific electrical conductivity of wheat varieties depending on the age of seedlings under the action of a complex of stressors, EC × 10^{-3} Cm/m

Variety	Option	Seedling age, days			
		10	12	14	16
Without heating the seeds					
Novosibirskaya 18	Control	3,5 ± 0,01	3,5 ± 0,05	3,1 ± 0,03	3,0 ± 0,05
	Conidial suspension *B. sorokiniana* + sodium chloride	25,5 ± 0,5*	44,7 ± 0,5*	42,2 ± 0,3*	43,5 ± 0,5*
Sibirskaya 21	Control	5,1 ± 0,05	4,1 ± 0,06	4,0 ± 0,01	4,1 ± 0,07
	Conidial suspension *B. sorokiniana* + sodium chloride	21,4 ± 0,7*	39,6 ± 1,1*	40,4 ± 0,9*	49,2 ± 1,2*
Heating up seeds at 43 ° C					
Novosibirskaya 18	Control	3,8 ± 0,03	3,8 ± 0,02	2,9 ± 0,03	3,0 ± 0,03
	Conidial suspension *B. sorokiniana* + sodium chloride	38,7 ± 1,0*	55,4 ± 1,2*	44,1 ± 0,8*	47,4 ± 0,9*
Sibirskaya 21	Control	6,9 ± 0,08	4,8 ± 0,04	4,2 ± 0,03	4,0 ± 0,02
	Conidial suspension *B. sorokiniana* + sodium chloride	23,8 ± 0,5*	36,8 ± 0,7*	43,6 ± 0,8*	45,4 ± 1,1*

* Differences with the control are significant at the significance level of $p \leq 0.01$.

In both cultivars, the difference in the relative change in the EC in the variants with and without heating the seeds was maximal in 10-day-old seedlings - 1.5 times (Novosibirskaya 18) and 1.3 times (Sibirskaya 21) with the significance of differences at the level of $p \leq 0, 05$. Intervarietal maximum differences were also recorded in 10-day-old seedlings; in the variant without heating the seeds, they were 1.9 times and in the variant with heating the seeds, 3.7 times, with the significance of differences at the level of $p \leq 0.05$.

One of the important characteristics of the process is the rate of its growth. To characterize the rate of release of electrolytes, an analysis of linear functions (derivatives of approximation dependences) was carried out, which made it possible to reveal a number of features (see Table 2).

The maximum electrolyte yield was achieved at a seedling age of about 15.5 days for all variants of experiments (14.86; 14.95; 15.67; 16.25 days), and in the Novosibirskaya 18 variety - a day earlier than in the Sibirskaya 21 variety. The slope of the function, which characterizes the rate of change in the rate of release of electrolytes, is also noted to be higher in the Novosibirskaya 18 variety, which indicates a greater instability of cell membranes in this variety under the combined action of stressors.

Thus, under the conditions of the sequential action of stressors, a protective effect of hyperthermia was established in the more resistant cultivar Sibirskaya 21 (a significant decrease in EC up to 1.3 times) as compared with the variant without heating the seeds. In the less resistant variety Novosibirskaya 18, heating the seeds destabilized the state of the cell membranes (a significant increase in the EC and the rate of electrolyte release by 1.5 and 1.2 times, respectively).

CONCLUSIONS

1. The informativity of leaf aqueous extracts EC as an indicator of change in cell membrane permeability of seedlings has been experimentally established to determine stress tolerance of wheat varieties to the combined action of cereal root rot, chloride salinity and seed hyperthermia pathogen.

2. The dependence of resistibility of the EC of leaf extracts of four varieties of spring wheat to simultaneous and sequential combined action of seed hyperthermia (43 °C), chloride salinity (1.3%) and common cereal rot pathogen *B. sorokiniana* (5000 conidia per grain) was

Табл. 2. Аналитические выражения аппроксимационных функций зависимости относительного изменения УЭП от возраста проростков

Table 2. Analytical expressions of the dependence approximation functions of EC relative change on the age of seedlings

Option	Analytic expression		Maximum abscissa *Xm*, days	Angle factor of function Y_1
	Function *Y*, EC	Derivative Y_1		
Novosibirskaya 18 variety				
Seeds without heating + Conidial suspension *B. sorokiniana* + sodium chloride	$Y_1 = -28{,}8125X^2 + 861{,}275X - 5080{,}45$	$-57{,}6250X + 861{,}275$	14,95	–57,6
Seeds with heating + Conidial suspension *B. sorokiniana* + sodium chloride	$Y_2 = -23{,}5625X^2 + 700{,}675X - 3713{,}65$	$-47{,}1250X + 700{,}675$	14,86	–47,1
Sibirskaya 21 variety				
Seeds without heating + Conidial suspension *B. sorokiniana* + sodium chloride	$-22{,}3125X^2 + 699{,}475X - 412{,}050$	$-44{,}6250X + 699{,}475$	15,67	–44,6
Seeds with heating + Conidial suspension *B. sorokiniana* + sodium chloride	$-20{,}3188X^2 + 660{,}353X - 4327{,}895$	$-40{,}6376X + 660{,}353$	16,25	–40,6

determined. The smallest changes in the EC value and electrolyte release rate were recorded in seedlings of relatively resistant varieties of spring wheat Omskaya 18 and Sibirskaya 21.

3. When studying the hourly dynamics of the electrolyte release (exposure of seedling leaves in water for 0.5–4.5 h) with the simultaneous action of *B. sorokiniana* (5000 conidia per grain) and chloride salinity (1.3%), there was a significant 1.5-fold increase in the relative change of the EC and a 2-fold increase in the electrolyte release in the less resistant variety Novosibirskaya 44 compared to the more resistant variety Omskaya 18.

4. In a study of the daily dynamics of electrolyte release in 10–16-day-old seedlings under the sequential action of seed hyperthermia (43° C), *B. sorokiniana* (5000 conidia per grain) and chloride salinity (1.3%), the protective effect of hyperthermia was established in a more resistant cultivar Siberian 21. There was a significant ($p \leq 0.05$) decrease in the relative change in EC by 1.3 times compared with the option without seeds heating. In the less resistant variety Novosibirskaya 18, the seeds heating destabilized the state of cell membranes, which led to a significant ($p \leq 0.05$) increase in the relative change in the EC and the rate of electrolyte release by 1.5 and 1.2 times, respectively.

5. Analytical expressions have been obtained, confirming the experimentally established methodological procedures of conductometric measurements, providing the maximum intervarietal differences: the age of seedlings - 10 days; the time interval of exposure of the samples in water was 1.5–4.5 h. The intervarietal differences in the variant without heating the seeds were 1.9 times and in the variant with heating the seeds - 3.7 times with the reliability of differences at levels $p \leq 0.05$ and $p \leq 0.01$. Intervarietal differences established at the time interval of exposure to the release of electrolytes 1.5–4.5 h were 1.50–1.67 times with the significance of differences at the level of $p \leq 0.05$.

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ЭФФЕКТИВНОСТЬ ИСПОЛЬЗОВАНИЯ БИОЛОГИЧЕСКОГО ПОТЕНЦИАЛА СОРТОВ ЛЬНА-ДОЛГУНЦА ТОМСКОЙ СЕЛЕКЦИИ ПРИ ПЕРЕРАБОТКЕ ЛЬНОТРЕСТЫ

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Приведены сведения о структуре посевных площадей льна-долгунца в России в 2016–2020 гг. Сорта томской селекции занимают почти 30% от общей площади посевов. Исследования проходили в льносеющих регионах России в 2001–2020 гг. Определена технологическая ценность льносырья изучаемых сортов льна-долгунца при переработке льнотресты различного качества на льноперерабатывающих предприятиях. Изучены показатели по общему выходу волокна, выходу длинного волокна, выходу короткого волокна, номеру длинного волокна, номеру короткого волокна, а также по комплексу указанных признаков. Установлена эффективность использования потенциальных возможностей изучаемых сортов по отношению к данным Госсортоиспытания. По результатам переработки льнотресты и рейтинговой оценки указанных сортов в сравнении с лучшими и худшими сортами установлено, что для низкокачественной льнотресты к числу сортов могут быть отнесены следующие: Томский 18 – по выходу короткого волокна (24,9%), Томский 16 – по номеру длинного волокна (11,08 N), Тост – по номеру короткого волокна (3,42 N) и комплексу признаков (средний индекс рейтинга 9,8 позиций). К группе лучших сортов по общему выходу волокна принадлежат сорта Томский 18 (33,2%) и Тост (32,2%), выходу длинного волокна Тост (13,1%), выходу (23,2%) и номеру (5,20 N) короткого волокна Томский 18 (высококачественная льнотреста). Уровень реализации биологического потенциала, установленного при Госсортоиспытании, всех представленных сортов в производственных условиях при переработке льнотресты по общему выходу волокна и выходу длинного волокна недостаточен. Его значение для сортов Томской школы селекции составляет по общему выходу волокна 65,2–86,3%, выходу длинного волокна 17,6–31,4% (низкокачественная льнотреста), 77,7–94,2 и 30,5–52,2% соответственно (высококачественная льнотреста).

Ключевые слова: лен-долгунец, льнотреста, выход волокна, льноперерабатывающие предприятия

EFFICIENCY OF USING THE BIOLOGICAL POTENTIAL OF FIBER FLAX VARIETIES OF THE TOMSK SCHOOL OF BREEDING IN FLAX STRAW PROCESSING

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The information on the structure of the areas cultivated with fiber flax in Russia in 2016–2020 is presented. The varieties of the Tomsk school of breeding occupy almost 30% of the total area under crops. The study was carried out in the flax-growing regions of Russia in 2001–2020. The technological value of flax raw material of the studied varieties of fiber flax for processing flax of various quality at flax processing enterprises was determined. The indicators for the total fiber yield, long and short fiber yield, long fiber number and short fiber number, and the combination of these features were studied. The efficiency of using the potential capabilities of the studied varieties in relation to the data of the State Variety Testing was established. According to the results of flax straw processing and the rating assessment of these varieties in relation to the best and worst ones, it was found that the following varieties can be referred to as the ones with low-quality flax straw: Tomsk 18 – by the yield of short

fiber (24.9%), Tomsk 16 – by the number of long fiber (11.08 N), Tost – by the number of short fiber (3.42 N) and the complex of features (average rating index of 9.8 positions). The group of the best varieties for the total fiber yield includes Tomsk 18 (33.2%) and Tost (32.2%), for the long fiber yield – Tost (13.1%), for the short fiber yield (23.2%) and the number of short fiber (5.20 N) – Tomsk 18 (high-quality flax). None of the varieties presented fulfil the biological potential in the processing of flax straw under production conditions in terms of the total fiber yield and the yield of long fiber, established by the State Variety Testing. Its value for the varieties of the Tomsk school of breeding is 65.2–86.3% for the total fiber yield, 17.6-31.4% for the long fiber yield (low-quality flax straw), 77.7–94.2 and 30.5–52.2%, respectively (high-quality flax straw).

Keywords: fiber flax, flax straw, fiber yield, flax processing enterprises

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INTRODUCTION

In modern conditions, the flax industry is faced with an important task related to import substitution - the creation of a reliable and stable domestic raw material base in the production of fiber flax, capable of meeting the requirements of flax processing enterprises in high-quality fiber products [1-3]. Improving the efficiency of fiber flax production and the competitiveness of its products depends on many factors [4–7]. One of the most important factors is the development of new domestic varieties that have economically valuable traits and a high degree of their manifestation during the processing of flax trees under production conditions [8–10]. Among other varieties of both domestic and foreign origin, these requirements are also met by the varieties of the Tomsk School of Breeding[1] [11–13].

The area sown under these varieties in Russia averaged almost 30% of the total area[2] from 2016 to 2020. Due to various reasons (primarily due to the insufficient amount of seeds of domestic breeding varieties) almost 30% of the area is cultivated with fiber flax varieties of foreign selection (see Table 1) [1, 2, 7]. Taking into account the high prevalence of Tomsk selection varieties in the crops, it is necessary to assess their importance from the position of technological flaxseed fleece in processing for fiber. According to the traditional technology of flax fiber processing for long and short fiber, the technological value is determined by their yield and quality[3] [14]. It is known that the biological potential of the varieties, which is determined by the State Variety Testing, in the practical activities of flax processing enterprises is far from being fully used.

The aim of the research is to determine the technological value of flax straw of the Tomsk breeding cultivars in comparison with other varieties of domestic and foreign origin in primary flax straw processing of different quality; to estimate the efficiency of using biological potential of the mentioned fiber flax varieties by main features in flax straw processing in production conditions.

[1]The State Register of Breeding Achievements Admitted to Cultivation in the Russian Federation. 2016.450 p.

[2]Varietal crops of fiber flax and hemp in 2015–2016. Statistical data "Agency" Len ". 2016–2020.

[3]Patent 2597552 C1 (Russian Federation) The method for assessing the technological value of fiber flax stems / V.A. Romanov, T.A. Rozhmina, M.M. Kovalev S.L. Belopukhov. Appl. 10.03.2015.

Табл. 1. Структура посевных площадей сортов льна-долгунца томской селекции (2016–2020 гг.)
Table 1. Structure of areas cultivated with fiber flax varieties of the Tomsk School of Breeding (2016–2020)

Crop year	Total acres, thousand ha, %	%	Domestic varieties, thousand ha, %	Foreign varieties, thousand ha, %	Grade-out crops, thousand ha,	Cropland acres, thousand ha, %										Total, thousand ha, %
						Tomsk 16	Tomsk 17	Tomsk 18	Tost	Tost 3	Tost 4	Tost 5	In memory of Krepkov	Tomich	Tomich 2	
2016	44,695		27,533	13,880	3,282	1208	9395	3938	6136	234	20	194	–	–	–	21,12
	100	To column 2	61,6	31,1	7,3	2,7	21,0	8,8	13,7	0,52	0,04	0,43	–	–	–	47,2
		To column 4	100	50,2	11,9	4,4	3,4	14,3	22,3	0,85	0,07	0,70	–	–	–	76,7
2017	44,971		24,949	11,600	8,422	500	5607	5076	–	228	64	130	–	1	–	11,596
	100	To column 2	55,5	25,8	18,7	1,1	12,5	11,3	–	0,51	0,12	0,29	–	0,002	–	25,8
		To column 4	100	46,5	33,8	2,0	22,5	20,3	–	0,91	0,26	0,52	–	0,01	–	46,5
2018	44,500		23,308	14140	7,052	723	5567	4092	–	100	47	416	–	–	–	10,945
	100	To column 2	52,4	31,8	15,8	1,6	12,5	9,2	–	0,22	0,11	0,94	–	–	–	24,6
		To column 4	100	60,7	30,2	3,1	23,9	17,6	–	0,43	0,20	1,78	–	–	–	47,0
2019	49,70		26,972	13600	9,128	–	6945	4474	–	407	32	725	24	182	–	12,640
	100	To column 2	54,2	27,4	18,4	–	14,0	9,0	–	0,82	0,06	1,46	0,05	0,04	–	25,4
		To column 4	100	50,4	33,8	–	25,7	16,6	–	1,51	0,12	2,69	0,09	0,07	–	46,9
2020	53,200		32,043	12700	8,457	–	7446	4749	–	438	152	1004	149	3	14	13,940
	100	To column 2	60,2	23,9	15,9	–	14,0	8,9	–	0,82	0,29	1,9	0,28	0,01	0,03	26,2
		To column 4	100	39,6	26,4	–	23,2	14,8	–	1,37	0,47	0,01	0,46	0,01	0,05	43,5
Mean	47,413		26,961	13,184	7,268	486	6978	4466	1227	281	63	494	35	37	3,0	14,048
	100	To column 2	56,9	27,8	15,3	1,1	14,8	9,4	2,7	0,6	0,12	1,00	0,07	0,01	0,01	29,6
		To column 4	100	49,5	27,2	1,9	25,9	16,7	4,5	1,0	0,22	1,14	0,11	0,02	0,01	52,1

MATERIAL AND METHODS

The studies were carried out from 2001 to 2020 in the conditions of flax-growing farms and flax-processing enterprises of the Tver, Smolensk, Pskov, Kostroma, Vologda regions, as well as on the basis of the All-Russian Research Institute of Flax (currently the Federal Scientific Center for Bast Crops). According to a special methodological program, which was developed to establish the standards for the transfer of flax straw of new varieties of fiber flax into fiber, batches of flax of different quality for each variety under study were prepared[4] [5, 7]. The weight of each batch was at least 2 tons. Some batches were represented by low-quality flax-straw (numbers 0.50–0.75), and some were high-quality flax-straw (numbers 1.00 and more). When carrying out control developments at flax processing enterprises, in accordance with the rules of technical operation of flax factories, the optimal operating mode of the equipment was established. According to the current GOSTs, the quality of flax straw, long and short fibers were determined at all production transitions. With normalized humidity and contamination of flax straw and fibers, the total yield, the yield of long and short fibers from each batch were established. The following equipment was used: SKP-10LU flax-straw dryer, MTL-1 crushing and scutching unit, TL-4-2 machine for processing loose pieces, SKP-10KU short fiber dryer, KPAL flax tow scutcher. State variety trials at variety plots of flax-growing regions of the Russian Federation were carried out according to the existing methodology[5]. The experimental data were statistically processed using the methods of mathematical statistics [15].

RESULTS AND DISCUSSION

During the research, the technological value of flax-straw was determined when processing 18 varieties of domestic selection and 12 varieties of foreign ones for fiber. The varieties of Tomsk selection are Tomsky 16, Tomsky 17, Tomsky 18, Toast. The total fiber yield, the yield and the number of long and short fibers were determined in the course of control developments for two groups of flax stands: low-quality and high-quality, separately for each batch of flax of a certain type. 6 varieties were identified with the maximum and minimum values for the total fiber output, long fiber output, short fiber output, long fiber number, and short fiber number according to the average values of the traits. The data are given in table 2, 3 (low-quality flax straw) and table 4, 5 (high quality flax straw). The tables show that both the best and the worst varieties in terms of individual traits do not include the same varieties. For example, the maximum average value of the total fiber yield from low-quality flax straw (see Table 2) was recorded for the Alexandrite variety (30.3%), the long fiber yield - for the Lenok variety (11.1%), the short fiber - for the Diplomat variety (25%), the number of the long fiber - in the Tomsky 16 variety (11.08 N), the short - in the Tverskoy variety (4.0 N) (see Table 3). For the varieties of Tomsk selection, the following pattern is observed: with the maximum value of the output of short fiber, Tomsky 18, the number of the long fiber - Tomsky 16, with the minimum value of the total yield of fiber - Tomsky 16, the yield of long fiber - Tomsky 17, Tomsky 18 (see Table 2, 3). These varieties occupy an intermediate position for the rest of the characteristics. From the data presented in table 4, 5 (high-quality flax straw), it follows that the best varieties in terms of total fiber yield include varieties Tomsky 18 (33.2%), Toast (32.2%), long fiber yield - Toast (13.1%), short - Tomsk 18 (23.2%), short fiber number - Tomsk 18 (5.20 N). The worst varieties in terms of total fibre yield (27.6%) and long fibre yield were Tomsk 16 (16.9%) and short fibre yield was Toast (17.4%).

[4]Order of the Ministry of Agriculture of the Russian Federation No. 23-r dated March 10th, 2016 “The procedure for determining the standards for the conversion of flax straw and hemp into fiber” (As amended by the Decree of the Government of the Russian Federation No. 450 dated 12.06. 2008). 7 p.

[5]The methodology for state variety testing of agricultural crops. Moscow: Kolos, 1985.263 p.

Табл. 2. Сорта льна-долгунца с минимальным и максимальным значением общего выхода, выхода длинного и короткого волокна (низкокачественная льнотреста)

Table 2. Varieties of fiber flax with a minimum and maximum value of the total yield, the yield of long and short fiber (low-quality flax straw)

Variety	Total fiber yield, %	Variety	Long fiber yield, %	Variety	Short fiber yield, %
Varieties with the highest trait value					
Alexandrite	30,3	Lenok	11,1	Diplomat	25,0
Diplomat	29,7	Zaryanka	9,6	Tomsk 18	24,9
A 93	29,5	Alexandrite	9,4	Sursky	23,5
Alpha	29,0	Leader	9,0	Elektra	23,4
Escalina	28,9	Grant	8,7	A 93	23,2
Grant	28,6	Alpha	8,6	Escalina	23,0
Tomsk varieties					
Tomsk 16	22,7		4,0		18,8
Tomsk 17	26,5		3,6		22,9
Tomsk 18	27,6		2,7		24,9
Tost	27,1		6,2		20,8
Varieties with minimum trait values					
Agatha	21,0	Elektra	2,7	Agatha	14,0
Caesar	21,1	Tomsk 18	2,7	Zaryanka	15,7
Susanna	22,4	Susanna	3,1	Caesar	16,8
Tomsk 16	22,7	Lira	3,4	Lenok	17,0
Lira	23,4	Sursky	3,6	Vasilek	18,0
Mogilevsky 2	23,7	Tomsk 17	3,6	Leader	18,7

Табл. 3. Сорта льна-долгунца с минимальным и максимальным значением номера длинного и короткого волокна (низкокачественная льнотреста)

Table 3. Varieties of fiber flax with a minimum and maximum value of the long and short fiber number (low-quality flax straw)

Variety	Long fiber number, N	Variety	Short fiber number, N
Varieties with the highest trait value			
Tomsk 16	11,08	Tverskoy	4,00
Zaryanka	11,00	Aleksim	3,75
Alpha	11,00	Alpha	3,71
Lenok	10,80	Leader	3,50
Vasilek	10,70	Tost	3,42
Tost	10,67	Alexandrite	3,38
Tomsk varieties			
Tomsk 16	11,08		3,00
Tomsk 17	10,24		3,00
Tomsk 18	10,08		3,00
Tost	10,67		3,42
Varieties with minimum trait values			
Sursky	8,96	Praleska	2,00
Alexandrite	9,60	Universal	2,00
Leader	9,70	Susanna	2,25
Impulse	9,73	Mogilevsky 2	2,50
Grant	9,88	Escalina	2,50
Escalina	10,00	Caesar	2,50

Табл. 4. Сорта льна-долгунца с минимальным и максимальным значением общего выхода, выхода длинного и короткого волокна (высококачественная льнотреста)

Table 4. Varieties of fiber flax with a minimum and maximum value of the total yield, the yield of long and short fiber (high-quality flax straw)

Variety	Total fiber yield, %	Variety	Long fiber yield, %	Variety	Short fiber yield, %
Varieties with the highest trait value					
Alpha	33,6	Lira	15,3	Praleska	26,0
Tverskoy	33,3	Alpha	14,5	Elektra	25,5
Tomsk 18	33,2	Alexandrite	13,6	Veralin	23,2
Veralin	33,2	Lenok	13,2	Tomsk 18	23,2
Praleska	33,1	Tost	13,1	A 93	22,6
Tost	32,2	Sursky	12,6	Impulse	22,3
Tomsk varieties					
Tomsk 16	27,1		6,9		20,4
Tomsk 17	31,0		11,2		19,8
Tomsk 18	33,2		9,9		23,2
Tost	32,2		13,1		17,2
Varieties with minimum trait values					
Aleksim	25,3	Elektra	6,3	Susanna	14,4
Susanna	25,5	Tomsk 16	6,9	Aleksim	15,2
Tomsk 16	27,1	Praleska	7,1	Lira	16,1
Smolich	28,2	Sofia	7,7	Tost	17,2
Dashkovsky	28,4	Dashkovsky	8,2	Lenok	17,4
Mogilevsky 2	28,6	Smolich	8,3	Zaryanka	17,6

Табл. 5. Сорта льна-долгунца с минимальным и максимальным значением номера длинного и короткого волокна (высококачественная льнотреста)

Table. 5. Varieties of fiber flax with a minimum and maximum value of the long and short fiber number (high-quality flax straw)

Variety	Long fiber number, N	Variety	Short fiber number, N
Varieties with the highest trait value			
Alpha	12,10	Tomsk 18	5,20
Zaryanka	11,95	Tverskoy	4,00
Leader	11,60	Zaryanka	4,00
Dashkovsky	11,52	Leader	4,00
Aleksim	11,48	Impulse	4,00
Agatha	11,47	Veralin	4,00
Tomsk varieties			
Tomsk 16	11,23		3,20
Tomsk 17	11,02		3,40
Tomsk 18	10,06		5,20
Tost	10,66		3,54
Varieties with minimum trait values			
Grant	10,00	Praleska	2,07
Veralin	10,04	Universal	2,80
Tomsk 18	10,06	Mogilevsky 2	2,84
Sofia	10,46	Grant	2,84
Sursky	10,50	Sofia	3,00
Alexandrite	10,50	Diplomat	3,11

The results of the analysis were obtained by conditionally dividing flax straw by quality into two groups: low-quality and high-quality. The group of high-quality flax straw includes nine numbers (1.00; 1.25; 1.50; 1.75; 2.00; 2.50; 3.00; 3.50; 4.00), a low-quality one consists of only two numbers (0.50; 0.75). In this regard, a more detailed differentiated analysis was carried out with the determination of the average index of the rating assessment of varieties according to the indicated characteristics across the entire rating scale GOST 24383–89 "Flax straw. Requirements for harvesting" to clarify the results. (see Fig. 1, 2).

In this case, the rating was understood as the place (position) occupied by the variety for each characteristic in terms of flax straw numbers, as well as for a complex of characteristics. In this case, the maximum value of the trait was assigned a rating of 1, the minimum value corresponded to a rating equal to the number of studied varieties. The average rating index was calculated as a weighted average, taking into account the number of flax-straw batches rated with a certain number.

Graphical analysis made it possible to distinguish the best varieties by a set of characteristics (a + b + c + d + e): for low-quality

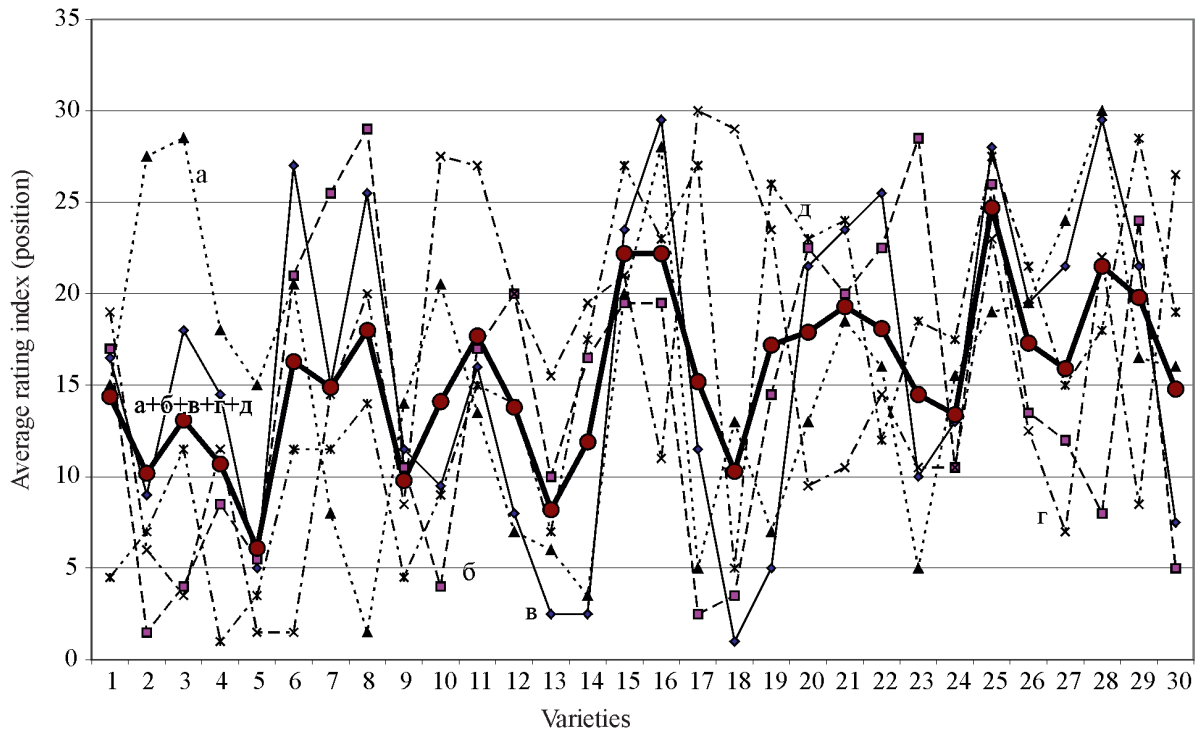

Рис. 1. Средний индекс рейтинговой оценки сортов льна-долгунца:
а – общий выход волокна, %; б – выход длинного волокна, %; в – выход короткого волокна;
г – номер длинного волокна, N; д – номер короткого волокна, N;
а + б + в + г + д – комплекс признаков (низкокачественная льнотреста).
Сорта: 1 – Алексим; 2 – Ленок; 3 – Зарянка; 4 – Тверской; 5 – Альфа; 6 – Томский 16; 7 – Томский 17;
8 – Томский 18; 9 – Тост; 10 – Лидер; 11 – Импульс; 12 – Смолич; 13 – А 93; 14 – Дипломат;
15 – Универсал; 16 – Цезарь; 17 – Сурский; 18 – Александрит; 19 – Эскалина; 20 – Дашковский;
21 – Могилевский 2; 22 – Лира; 23 – Электра; 24 – Веролин; 25 – Сюзана; 26 – София; 27 – Василек;
28 – Агата; 29 – Пралеска; 30 – Грант

Fig. 1. Average rating index of fiber flax varieties by:

a – total fiber yield, %, б – long fiber yield, %, в – short fiber yield, %, г – long fiber number, N, д – short fiber number, N; (а+б+в+г+д) complex of characteristics (low-quality flax straw)
Varieties: 1. Aleksim; 2. Lenok; 3. Zaryanka; 4. Tverskoy; 5. Alpha; 6. Tomsk 16; 7. Tomsk 17; 8. Tomsk 18; 9. Tost; 10. Leader; 11. Impulse; 12. Smolich; 13. A93; 14. Diplomat; 15. Universal; 16. Caesar; 17. Sursky; 18. Alexandrite; 19. Escalina; 20. Dashkovsky; 21. Mogilevsky 2; 22. Lira; 23. Elektra; 24. Veralin; 25. Susanna; 26. Sofia; 27. Vasilek; 28. Agatha; 29. Praleska; 30. Grant

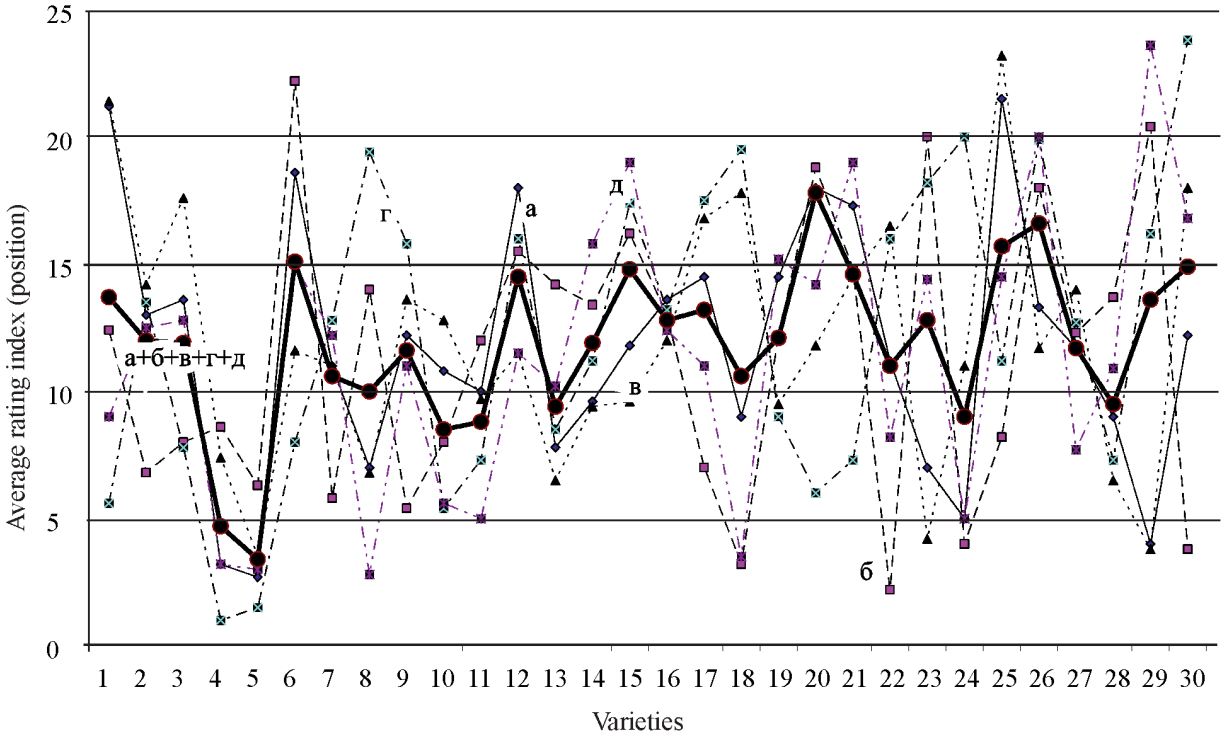

Рис. 2. Средний индекс рейтинговой оценки сортов льна-долгунца (высококачественная треста):
а – общий выход волокна, %; б – выход длинного волокна, %; в – выход короткого волокна; г – номер длинного волокна, N; д – номер короткого волокна, N; а + б + в + г + д – комплекс признаков (высококачественная льнотреста). Обозначение сортов см. на рис. 1

Fig. 2. Average rating index of fiber flax varieties (high-quality flax) by:
а – total fiber yield, %, б – long fiber yield, %, в – short fiber yield, %,
г – long fiber number, N, д – short fiber number, N; (а+б+в+г+д) complex of characteristics (high-quality flax), names of the varieties see fig.1

flax - Alfa, A 93, Toast, Lenok, Aleksandrite, Tverskoy with an average rating index of 6.1, respectively; 8.2; 9.8; 10.2; 10.3; 10.5 positions. Average rating index for high-quality flax of the best varieties: Alpha - 3.4, Tverskoy - 4.7, Leader - 8.5, Impulse - 8.8, Veralin - 9.0, A 93 - 9.4 positions.

Among the varieties of the Tomsk school of selection by a complex of characteristics, the variety Toast (average index - 9.8 positions) (low-quality flax straw) stood out. Varieties Tomsky 18, Tomsky 17, Toast (high-quality flax straw) (average index 10.6, 10.0, 11.6 positions) were only slightly inferior in this indicator to the best varieties.

An equally important issue in determining the significance of a variety in the processing of flax is not only the obtained actual values of the yield and quality of fiber, but also the disclosure of the potential of the variety, established during the State Variety Test, for these characteristics in the practice of flax processing enterprises. The results of flax-straw processing in terms of the total yield and the yield of long fiber obtained under production conditions, as a percentage in relation to the data of the State Variety Test, taken as 100% are presented in fig. 3 (low quality flax seed) and fig. 4 (high-quality). It can be seen from the figures that the level of realization of the potential possibilities of varieties, especially in terms of the yield of long fiber, is clearly insufficient, for low-quality flax straw it is lower than for the high-quality flax straw.

Statistical processing of the data confirmed the significance of the differences between the data of the State Variety Test and the results of processing flax straw in production conditions at a 5% significance level. According to the total fiber yield from low-quality flax-straw, the

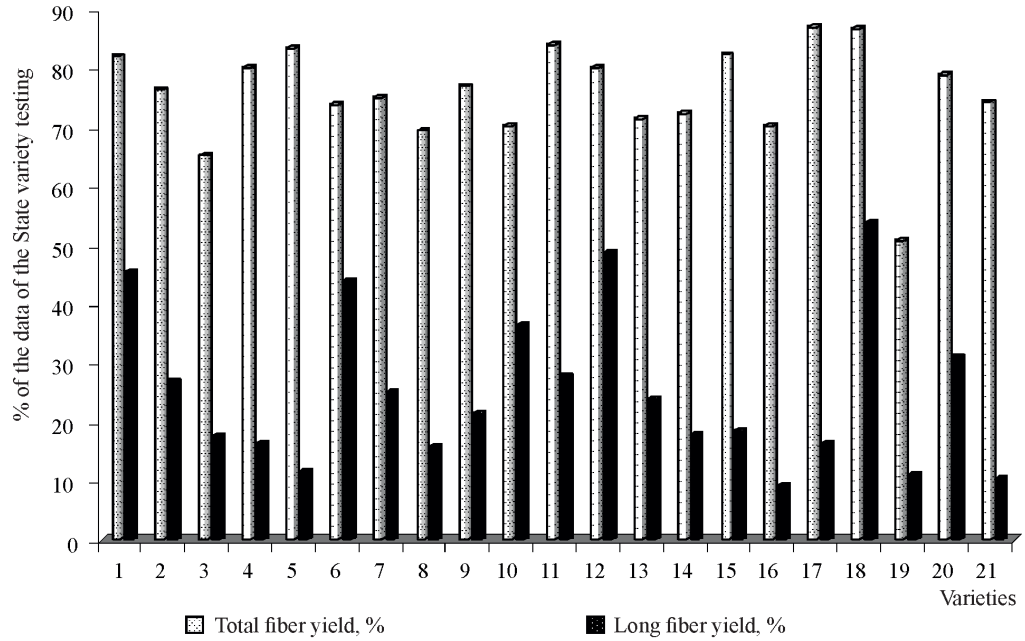

Рис. 3. Уровень реализации потенциала сортов льна-долгунца по общему выходу волокна и выходу длинного волокна из низкокачественной льнотресты, % к данным Госсортоиспытания.

Сорта: 1 – Зарянка; 2. Тверской; 3 – Томский 16; 4 – Томский 17; 5 – Томский 18; 6 – Тост; 7 – Импульс; 8 – Пралеска; 9 – Смолич; 10 – Василек; 11 – Алексим; 12 – Ленок; 13 – А 93; 14 – Дашковский; 15 – Могилевский 2; 16 – Дипломат; 17 – Универсал; 18 – Александрит; 19 – Цезарь; 20 – Грант; 21 – Сурский

Fig. 3. The level of implementation of the potential of fiber flax varieties by total fiber yield and long fiber yield from low-quality flax straw, % of the data of the State variety testing

1. Zaryanka; 2. Tverskoy; 3. Tomsk 16; 4. Tomsk 17; 5. Tomsk 18; 6. Tost; 7. Impulse; 8. Praleska; 9. Smolich; 10. Vasilek; 11. Aleksim; 12. Lenok; 13. A 93; 14. Dashkovsky; 15. Mogilevsky 2; 16. Diplomat; 17. Universal; 18. Alexandrite; 19. Caesar; 20. Grant; 21. Sursky

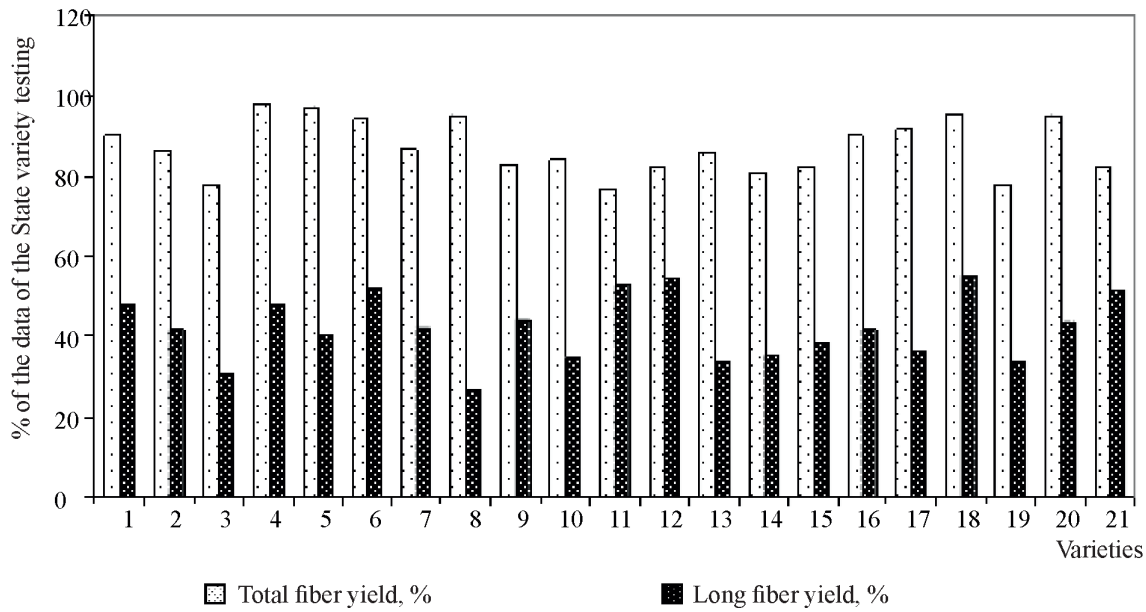

Рис. 4. Уровень реализации потенциала сортов льна-долгунца по общему выходу волокна и выходу длинного волокна из высококачественной льнотресты, % к данным Госсортоиспытания. Обозначение сортов см. на рис. 3

Fig. 4. The level of implementation of the potential of fiber flax varieties by total fiber yield and long fiber yield from high-quality flax straw, % of the data of the State variety testing (names of varieties see fig. 3)

error of the mean difference (Sd) was 0.88%, the Student's coefficient t f = 7.16> t t = 2.05; from high quality - Sd = 0.90, t f = 4.22> t t = 2.05, respectively. As for the long fiber yield, the significance of the differences is much higher: Sd = 1.25, t f = 15.80> t t = 2.05 (low-quality flax tree); Sd = 1.20, t f = 13.40> t t = 2.05 (high quality).

The level of realization of the biological potential of varieties of the Tomsk school of breeding is as follows: from low-quality flax-tree in the total fiber yield Tomsky 16 - 65.2%, Tomsky 17 - 80.1, Tomsky 18 - 83.3, Toast - 73.8%, from high quality - 77 , 7; 98.0; 97.0; 94.2% respectively; for the yield of long fiber for the same varieties and quality of flax - 17.6; 16.2; 11.4; 31.4%; 30.5; 48.3; 40.1; 52.2% respectively.

It is obvious that the existing reserve in increasing the efficiency of using the potential capabilities of varieties consists in improving the quality of flax raw materials, technical re-equipment of the industry, improving the organization of labor both in the production of fiber flax and in the processing of fiber products.

CONCLUSIONS

1. The importance of flax varieties of the Tomsk school of breeding was confirmed in terms of obtaining various numbers of high fiber yield (long and short), characterized by good quality, during the processing of flax straw.

2. Variety Tomsky 18 belongs to the group of varieties with the maximum value of short fiber yield from low quality flax (24.9%), total fiber yield (33.2%), yield (23.2%) and short fiber number (5.2 N) from high quality flax straw. Tomsky 16 is one of the best in terms of long fiber number (11.08 N) (low quality flax straw), Toast has a maximum value in total fiber yield (32.2%), long fiber yield (13.1%) (high quality flax straw), as well as by the number of short fiber (3.42 N) and a complex of features (average rating index 9.8 positions) (low-quality flax straw).

3. The potential possibilities of the varieties of the Tomsk School of Breeding in terms of the total fiber yield are realized in production conditions by 65.2–83.3%, by the yield of long fiber by 17.6–31.4% (flax straw numbers 0.50–0.75) and, accordingly by 77.7–94.2%; 30.5–52.2% (flax straw number 1.00 and more).

4. With the rational organization of the processing of flax raw materials with a simultaneous increase in its quality, a more complete realization of the inherent biological potential in the varieties of the Tomsk School of Breeding in relation to obtaining the optimal amount of fibrous products of the best quality can be achieved.

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ФОРМИРОВАНИЕ УРОЖАЙНОСТИ СОРТОВ ГОЛОЗЕРНОГО ОВСА ПРИ РАЗЛИЧНЫХ УСЛОВИЯХ ВОЗДЕЛЫВАНИЯ

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Представлены результаты изучения показателей урожайности голозерного овса при различных сроках посева и нормах высева. Исследования проведены в 2016–2020 гг. в полевом опыте в условиях Западной Сибири. Изучены образцы разных групп спелости культуры: среднеранний сорт Гаврош и среднеспелый сорт Офеня. На продолжительность вегетационного периода голозерного овса оказали влияние метеорологические факторы. Отмечена тенденция уменьшения продолжительности межфазных периодов и вегетационного периода в целом от раннего срока посева к более позднему на 4–10 дней у сорта Гаврош и на 8–10 дней у сорта Офеня. Урожайность у сорта Гаврош при раннем сроке посева достоверно превышала аналогичный показатель при среднем сроке на 17,6% и при позднем сроке – на 19,0%, у сорта Офеня – на 10,9% и на 16,2% соответственно. Увеличение урожайности раннего срока посева среднераннего сорта Гаврош относительно среднего и позднего сроков посева определялось большей крупностью зерна (r = 0,6929…0,9535 при R = 0,5140). У среднеспелого сорта Офеня в годы исследований на всех вариантах большее значение имело число продуктивных стеблей с единицы площади (r = 0,7444…0,9054 при R = 0,5140) и масса 1000 зерен (r = 0,5350…0,8297 при R = 0,5140). Наиболее оптимальная норма высева для сортов голозерного овса – 5,0–6,0 млн всхожих зерен/га. При данной норме отмечено не максимальное проявление отдельных показателей структурных элементов урожайности, а совокупность их средних значений.

Ключевые слова: голозерный овес, урожайность, срок посева, норма высева

FORMATION OF PRODUCTIVITY OF NAKED OAT VARIETIES UNDER DIFFERENT CULTIVATION CONDITIONS

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The results of studying naked oat yield indicators depending on different sowing dates and seeding rates are presented. The study was carried out in 2016–2020 in a field experiment in Western Siberia. Samples of different groups of the crop ripeness were studied: mid-early variety Gavrosh and mid-ripening variety Ofenya. The duration of the growing season of naked oats was influenced by meteorological factors. There was a tendency observed for a decrease in the duration of interphase periods and the growing season as a whole from an early sowing date to a later one by 4-10 days for the Gavrosh variety and by 8-10 days for the Ofenya variety. The yield of the Gavrosh variety at an early sowing period significantly exceeded the same indicator at a mid-period by 17.6% and at a late period – by 19.0%, of the Ofenya variety – by 10.9% and 16.2%, respectively. The increase in the yield for the early sowing period of the mid-early variety Gavrosh relative to the middle and late sowing dates was determined by a larger grain size (r = 0.6929 ... 0.9535 at R = 0.5140). For the mid-

ripening variety Ofenya, the number of productive stems per unit area (r = 0.7444 ... 0.9054 with R = 0.5140) and the weight of 1000 grains (r = 0.5350 ... 0.8297 at R = 0.5140) were of more importance in the years of research on all variants. The most optimal seeding rate for naked oat varieties is 5.0–6.0 million germinating grains/ha. At this rate, it was not the maximum manifestation of individual indicators of the yield structural elements, but a combination of their average values was noted.

Keywords: naked oats, yield, sowing time, seeding rate

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The authors declare no conflict of interest.

INTRODUCTION

At present, the Russian Federation is the world's largest producer of oats. The world harvest of oats in Russia is 22%, in Canada - 14, in the USA - 7, in Poland - 6, in Australia - 5%. In terms of production, oats ranks seventh in the world, bending to corn, rice, wheat, barley, sorghum and millet. In Russia, the cultivation of oats is on the fourth place (3.0%) after wheat (36.8%), barley (10.7%) and corn for grain (3.6%) (according to Rosstat data as of 2020[1]). At the same time, there is a downward trend in the gross harvest of this crop; in 2015 it amounted to 4.3% of the total gross harvest, in 2020 - 3.1%.

The Kemerovo region (Kuzbass) produces 11 thousand tons of seeds of spring oats of Siberian breeding varieties, including 6.8 thousand tons of seeds of varieties selected by the Kemerovo Research Institute of Agriculture - a branch of the SFSCA RAS, of which - 1.1 thousand tons of naked oats.

The naked forms of oats are the most valuable of cereals in terms of the biochemical composition of the grain, which determines its dietary and therapeutic and prophylactic properties. This makes it possible to use them in the development of products for children, functional and specialized purposes [1]. Naked oats are a unique crop in its properties. The grain of naked oats contains a large amount of proteins (up to 18–20%), oils (up to 8–12%), water-soluble polysaccharides of beta-glucans (up to 6–8%). Also, naked oats are rich in micro and macro elements (potassium, magnesium, calcium, silicon, phosphorus, sodium, etc.). In terms of the content of vitamins of group A, B, E, K, naked forms of oats demonstrate the highest rates. Oats are the only cereal containing globulins or avenins with a mass fraction of 80% of the total protein. Globulins are water-soluble, so that oats can be converted into a milky liquid. The proteins of naked oats are characterized by an increased content of arginine and an essential acid, lysine, which is almost 2 times higher in comparison with other cereals, including chalky oats [2–7].

The potential of the naked grain of oats, determined by such indicators as hoodness (0.5–0.9%), kernel content (99.1–99.5%), indicates the possibility of processing such oats for food production. The high proportion of the kernel in comparison with the grain of the scarious varieties and the uniformity make it possible to determine a potentially higher yield of finished products at correspondingly lower production costs [8–10].

Naked oat varieties are more demanding in terms of growing conditions than the scarious oats, which is due to their morphological

[1]Federal State Statistics Service. Agriculture, hunting and forestry URL: https: // rosstat. gov.ru/enterprise_economy (date of access 04/27/2021).

and biological characteristics. Technological methods of cultivation allow regulating the water and temperature conditions [11, 12].

An essential place in a number of technological methods of cultivation is occupied by the correct choice of the seeding rate and sowing time. The seed application rate determines the density of plants, the provision of agrophytocenosis with nutrients, moisture and light, its productive properties, the yield of conditioned seeds and their sowing qualities [13]. The creation of an optimal crop density is determined by the following factors: biological (potential productivity, bushiness, lodging resistance), agrotechnical (fertilization, predecessors, terms and methods of sowing), natural (natural soil fertility, physical soil properties, field relief), economic (weediness of fields, the purpose of crops - for grain, hay, green mass), agrometeorological (provision with light, heat, moisture during the growing season, depending on the changing needs of plants in ontogenesis) [14].

The choice of the sowing time is an important technological indicator, which is largely influenced by the weather conditions. It significantly changes the conditions of germination and growth of spring oats at all stages of the growing season and the formation of the yield of agricultural crops [15, 16].

The aim of the research is to study the indicators of productivity of naked oats varieties under different cultivation conditions.

MATERIAL AND METHODS

The studies were carried out in the northern forest-steppe of the Kemerovo region on the experimental field of the Kemerovo Research Institute of Agriculture - a branch of the Siberian Federal Scientific Center of Agrobiotechnology of the Russian Academy of Sciences (Kemerovo Research Institute of Agriculture - a branch of the SFSCA RAS) in 2016–2020. The objects of research were the naked oats varieties: the mid-early variety Gavrosh and the mid-season variety Ofenya. The research conditions were unstable both over the years and within the same growing season. So, 2016 and 2017 were characterized as arid (HC May - August = 0.9 and 1.0), 2018 and 2020 - excessively humidified (HC May - August = 1.6 and 1.4), 2019 - sufficiently moisture supplied, with an optimal temperature regime (HC May - August = 1.2), which influenced the formation of the harvest of naked oats.

The predecessor was pure fallow. Sowing was carried out on three dates: the first was at the onset of physical maturity of the soil (27.04-14.05), the subsequent dates were sown at 8-10-day intervals (9-21.05 and 21-28.05). The following seeding rates were studied for each date: 4.0; 4.5; 5.0; 5.5; 6.0 million germinated grains/ha. Sowing was carried out with a SN-10C seeder on a plot area of 10 m^2 in quadruple replications. The plot arrangement was randomized. Records, observations, statistical processing of data were performed in accordance with the approved guidelines[2-4].

RESULTS AND DISCUSSIONS

During the study of naked oats varieties, the differences in the duration of the passage of ontogenetic phases from weather conditions were noted. A tendency was noted for a decrease in the duration of both interphase periods and the entire growing season as a whole from an early sowing date to a later one by 4–10 days in the Gavrosh variety and by 8–10 days in the Ofenya variety. On average, over the years of research, the duration of the growing season of the medium-early variety Gavrosh at early sowing dates was 86 days, at average - 83 days, at later - 78 days. The duration of the growing season of the mid-ripening variety Ofenya at different sowing dates was 93, 88 and 83 days, respectively.

Of no small importance in the structure of the growing season is the time of emergence of seedlings, i.e. the number of days from the moment of sowing the seeds to the emergence

[2]Methodology for state variety testing of agricultural crops. M., 1985.270 p.
[3]Dospekhov B.L. Field experiment technique. M.: Agropromizdat, 1985.352 p.
[4]Sorokin O.D. Applied statistics on the computer. Krasnoobsk: GUP RPO SO RASKhN, 2004.162 p.

of seedlings. It affects the further growth and development of plants and largely depends on the meteorological conditions during this period. The time of emergence of seedlings had a greater influence on the plant height (r = –0.7123 ... –0.8004 at R = 0.5140), panicle length (r = –0.6980… –0.7365 at R = 0.5140), the number of flowers in a panicle (r = –0.6507… –0.8408 at R = 0.5140). With early sowing dates, the duration of the sowing - sprouting period for the Gavrosh variety averaged 13 days, for the Ofenya variety - 15 days. In the middle and late stages, with an increase in air temperature and humidity, the period of seed germination was reduced to 12–10 days in the Gavrosh variety, and to 14–13 days in the Ofenya variety. When analyzing the obtained experimental data, the influence of the duration of the germination time on the formation of the yield of varieties (r = –0.8798 ... –0.9982 at R = 0.5140) was noted, therefore, when calculating the duration of the entire growing season as a whole, it is necessary to take into account the value of this subperiod.

The change in the duration of the growing season was more influenced by the interphase period of seedlings - peeling (r = 0.8255 ... 0.9875 at R = 0.5140), the value of which averaged 42 days in the Gavrosh variety at early sowing, with an average of 39 , with a late one - 38 days, with the Ofenya variety - 46, 44 and 39 days, respectively. The determining criterion was the nature of soil moisture during this period (r = 0.6934 ... 0.9795 at R = 0.5140) in order to pass this period.

The duration of the interphase period between flowering and ripening in the variants differed slightly and was 44-40 days in the Gavrosh variety and 47-44 days in the Ofenya variety.

The highest yields of the studied varieties Gavrosh and Ofenya were formed in 2019. (2.89 and 3.07 t/ha) under optimum moisture conditions (GTC = 1.2). The minimum yield was obtained in the drought year 2017. (GTC = 1.0) - 1.50 and 1.70 t/ha, respectively.

In the mid-early variety Gavrosh the advantage of early sowing dates with an average yield of 2.73 t / ha was noted, while with an average period it was 2.25 t / ha, with a late one - 2.21 t / ha. At the same time, for all sowing terms, the best results were obtained at increased seeding rates of 5.0 and 6.0 million germinating grains / ha (see Table 1).

In the mid-ripening variety Ofenya, with an early sowing period, the yield was 3.03 t / ha, with an average - 2.70, with a late sowing - 2.54 t / ha. A higher yield of this variety was formed at a seeding rate of 5.5 and 6.0 million germinating grains / ha at all sowing dates (see Table 2). On average, according to experience, the yield of the Ofenya variety exceeded the productivity of the Gavrosh variety by 15%.

On average, over the years of research, a significant advantage of early sowing dates was noted: by 17.6% to the average date and by 19.0% to the late date in the Gavrosh variety; by 10.9% by the average term and by 16.2% by the late term in the Ofenya variety.

The excess yield of the early sowing period of the medium early variety Gavrosh was determined by the larger grain size (r = 0.6929 ... 0.9535 at R = 0.5140) relative to the average and late sowing dates, especially in drier years. On average, according to the experiment, the mass of 1000 grains in the Gavrosh variety at an early period was 23.2 g, with an average - 22.6, with a late one - 21.5 - 24.1 g (see Table 3).

At the same time, at an early and medium term, a high yield was noted on variants with an increased number of productive stems per unit area (r = 0.6437 ... 0.7899 at R = 0.5140), which characterized variants with increased seeding rates - 5.5 and 6.0 million germinating grains / ha. However, high values of individual elements of the yield structure, such as productive tillering, the number of grains in the panicle, and the mass of grain from the main panicle were observed at low seeding rates - 4.0 and 4.5 million germinating grains / ha. The yield of the variants of the late sowing period was formed with average indicators of structural elements.

The number of productive stems per unit area (r = 0.7444 ... 0.9054 at R = 0.5140) and the mass of 1000 grains (r = 0.5350 ... 0, 8297 at R = 0.5140) was of the decisive importance in the formation of the yield of the mid-ripening variety Ofenya in the years of research on all the

Табл. 1. Урожайность голозерного овса Гаврош, т/га, 2016–2020 гг.
Table 1. Yield of naked oats Gavrosh, t/ha, 2016–2020

Sowing time	Year of study	Seeding rate, million germinating grains / ha					Average by term
		4,0	4,5	5,0	5,5	6,0	
Early	2016	2,43	2,50	2,55	2,57	2,55	2,52
	2017	1,41	1,37	1,37	1,29	1,51	1,39
	2018	3,22	3,20	3,07	3,28	2,80	3,11
	2019	4,27	4,20	4,27	4,09	4,14	4,19
	2020	2,28	2,30	2,43	2,41	2,73	2,43
	Average by norm	2,72	2,71	2,74	2,73	2,75	2,73
Medium	2016	2,55	2,57	2,69	2,71	3,00	2,70
	2017	1,54	1,30	1,29	1,41	1,30	1,37
	2018	2,61	2,45	2,58	2,44	2,26	2,51
	2019	2,60	2,44	2,49	2,54	2,52	2,52
	2020	1,99	2,07	2,13	2,36	2,27	2,16
	Average by norm	2,26	2,17	2,24	2,29	2,27	2,25
Late	2016	2,50	2,40	2,40	2,47	2,76	2,51
	2017	1,54	1,66	1,76	1,80	1,67	1,69
	2018	2,45	2,16	2,65	2,66	2,34	2,45
	2019	1,87	1,94	2,03	2,00	2,00	1,97
	2020	2,24	2,50	2,35	2,42	2,65	2,43
	Average by norm	2,12	2,13	2,24	2,27	2,28	2,21

Note. LSD 0.5 factor A (year) = 0.08; factor B (grade) = 0.04; factor C (sowing time) = 0.05; factor D (seeding rate) = 0.07.

Табл. 2. Урожайность голозерного овса Офеня, т/га, 2016–2020 гг.
Table 2. Yield of naked oats Ofenya, t/ha, 2016–2020

Sowing time	Year of study	Seeding rate, million germinating grains / ha					Average by term
		4,0	4,5	5,0	5,5	6,0	
Early	2016	3,24	3,26	3,45	3,40	3,55	3,38
	2017	1,71	1,68	1,88	1,83	1,77	1,77
	2018	3,60	3,74	3,61	3,62	3,93	3,70
	2019	3,56	3,37	3,60	3,69	3,43	3,53
	2020	2,39	2,84	2,78	2,86	3,05	2,78
	Average by norm	2,90	2,98	3,06	3,08	3,15	3,03
Medium	2016	3,12	3,22	3,60	3,50	3,67	3,42
	2017	1,61	1,69	1,83	1,67	1,63	1,69
	2018	2,71	2,82	3,09	2,88	2,57	2,81
	2019	2,92	3,15	3,17	3,22	3,21	3,13
	2020	2,46	2,34	2,62	2,39	2,52	2,47
	Average by norm	2,56	2,64	2,86	2,73	2,72	2,70
Late	2016	3,22	3,07	3,00	3,12	3,31	3,14
	2017	1,62	1,90	1,73	1,74	1,66	1,73
	2018	2,33	2,21	2,37	2,21	2,56	2,33
	2019	2,37	2,64	2,34	2,71	2,66	2,54
	2020	2,63	3,06	3,18	2,98	3,04	2,98
	Average by norm	2,43	2,58	2,52	2,55	2,65	2,54

Note. LSD 0.5 factor A (year) = 0.09; factor B (grade) = 0.04; factor C (sowing time) = 0.07; factor D (seeding rate) = 0.09.

Табл. 3. Агробиологическая характеристика голозерного овса сорта Гаврош, 2016–2020 гг.
Table 3. Agrobiological characteristics of naked oats of the Gavrosh variety, 2016–2020

Sowing time	Seeding rate, million germinating grains / ha	Number of productive stems, pcs / m^2	Productive bushiness, pcs.	The number of grains in a panicle, pcs.	Splitting of filmy grains, %	Grain weight from the main panicle, g	Weight of 1000 grains, g	Natural weight, g / l
Early	4,0	344	1,8	40,8	2,2	0,89	23,1	626
	4,5	374	1,8	36,3	2,3	0,80	23,1	628
	5,0	380	1,7	34,8	2,4	0,76	23,4	627
	5,5	372	1,5	29,7	2,2	0,69	23,2	628
	6,0	392	1,5	32,9	2,3	0,73	23,1	621
	Average	372	1,7	34,9	2,3	0,77	23,2	626
Me-	4,0	344	1,9	43,6	2,8	0,96	22,6	609
dium	4,5	329	1,9	41,7	2,9	0,91	22,7	607
	5,0	353	1,9	37,7	2,5	0,86	22,4	618
	5,5	413	1,9	32,7	2,6	0,72	22,5	608
	6,0	411	1,8	34,2	2,9	0,72	22,6	617
	Average	370	1,9	38,0	2,7	0,83	22,6	612
Late	4,0	392	2,0	40,5	1,3	0,88	21,3	597
	4,5	404	2,0	41,0	1,6	0,86	22,0	594
	5,0	391	1,8	40,0	1,5	0,84	21,6	603
	5,5	398	1,8	34,0	1,7	0,71	21,4	597
	6,0	403	1,6	34,3	1,7	0,73	21,2	594
	Average	398	1,8	38,0	1,6	0,80	21,5	597

variants. Thus, a greater number of productive stems was noted at early sowing dates of 334 pcs / m^2, with medium and late dates - 302 and 312 pcs / m^2, respectively. The weight of 1000 seeds in terms of sowing time was 27.7; 26.5 and 24.6 g (see Table 4).

The best indicators of structural elements in the Ofenya variety were also noted at seeding rates of 4.0 and 4.5 million germinating grains / ha. At the same time, a high yield on options 5.5 and 6.0 million germinating grains / ha was formed with a combination of average values of structural elements.

Табл. 4. Агробиологическая характеристика голозерного овса сорта Офеня, 2016–2020 гг.
Table 4. Agrobiological characteristics of naked oats of the Ofenya variety, 2016–2020

Sowing time	Seeding rate, million germinating grains / ha	Number of productive stems, pcs / m^2	Productive bushiness, pcs.	The number of grains in a panicle, pcs.	Splitting of filmy grains, %	Grain weight from the main panicle, g	Weight of 1000 grains, g	Natural weight, g / l
Early	4,0	274	1,8	45,7	4,7	1,38	27,7	605
	4,5	314	1,8	44,9	5,1	1,21	27,8	611
	5,0	342	1,9	40,8	4,6	1,22	27,6	612
	5,5	382	1,8	41,2	5,8	1,14	27,7	607
	6,0	357	1,8	41,8	6,0	1,11	27,5	603
	Average	334	1,8	42,9	5,3	1,21	27,7	608
Me-	4,0	292	1,6	41,1	3,1	1,12	26,6	605
dium	4,5	289	1,6	40,6	3,0	1,10	26,5	598
	5,0	327	1,9	34,7	3,1	0,96	26,5	604
	5,5	337	1,7	37,2	3,8	0,97	26,9	606
	6,0	302	1,6	36,2	4,1	0,95	25,2	600
	Average	309	1,7	38,0	3,4	1,02	26,5	603
Late	4,0	273	1,7	46,4	1,4	1,09	24,3	567
	4,5	327	1,8	44,6	1,5	1,02	24,3	569
	5,0	308	1,7	45,2	1,3	1,04	24,7	572
	5,5	327	1,7	40,2	1,3	0,95	25,0	574
	6,0	327	1,6	43,8	1,7	1,01	24,8	571
	Average	312	1,7	44,0	1,4	1,02	24,6	571

Different variants of the experiment also influenced the plumpness and density of the grain of naked oats, which are characterized by the full-scale weight of the grain. This indicator was influenced by the conditions of moisture supply during the tillering period – forming into a tube (r = 0.7344 ... 0.8147 at R = 0.5140) and ripening (r = 0.6004 ... 0.6442 at R = 0.5140). There was a tendency to decrease the natural weight from early to late sowing in both varieties. A plump caryopsis of a higher quality was observed in the varieties Gavrosh and Ofenya on the variants of 5.0 and 5.5 million germinating grains / ha: 603–628 and 574–606 g / l, respectively.

CONCLUSION

As a result of a long-term study of naked oat varieties Gavrosh and Ofenya under various cultivation conditions elements of varietal technology have been identified that ensure high productivity of varieties in Western Siberia. The advantage of sowing naked oats in the early stages was noted, the yield exceeded by later sowing dates in the mid-early variety Gavrosh is 17-19%, in the mid-ripening variety Ofenya - 11-16%. The most optimal seeding rate for naked oat varieties is 5.0–6.0 million germinating grains / ha.

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ПРОДУКТИВНОСТЬ ЗЕМЛЯНИКИ САДОВОЙ И СЕЛЕКЦИОННЫЕ ВОЗМОЖНОСТИ ЕЕ ПОВЫШЕНИЯ

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На основе литературных источников обобщены сведения о признаке продуктивности земляники садовой *Fragaria* × *ananassa* (Duchesne ex Weston) Duchesne ex Rozier. Признак продуктивности имеет сложный полигенный характер. Независимое наследование отдельных компонентов продуктивности позволяет моделировать оптимальное их сочетание в новом сорте. В основе создания высокопродуктивных сортов земляники садовой лежит внутривидовая гибридизация. Установленная положительная корреляция между размером ягод и урожаем свидетельствует о возможности повышения продуктивности сортов путем использования крупноплодных форм в селекции. На современном этапе в различных зонах садоводства исследователи выделяют крупноплодные сорта отечественной селекции: ‘Атлас’, ‘Берегиня’, ‘Витязь’, ‘Гренада’, ‘Забелинская’, ‘Кемия’, ‘Наше Подмосковье’, ‘Нелли’, ‘Первоклассница’, ‘Русич’, ‘Солнечная Полянка’, ‘Соловушка’, ‘Фестивальная Ромашка’, ‘Фруктовая’, ‘Царица’, ‘Японка’, и зарубежной селекции: ‘Alba’, ‘Asia’, ‘Elsanta’, ‘Finesse’, ‘Florense’, ‘Vivaldi’, ‘Galia’, ‘Jive’, ‘Joly’, ‘M. Champion’, ‘Merced’, ‘Murano’, ‘Onda’, ‘Roxana’, ‘Rumba’, ‘Tarda Vicoda’, ‘Vima Kimberly’, ‘Vima Tarda’, ‘Vima Rina’, ‘Vima Xima’. Отмечена возможность достижения гетерозисного эффекта по признаку продуктивности при использовании метода инбридинга в селекции. Метод генетических модификаций растений имеет большое значение для селекции сложных количественных признаков продуктивности, включающих урожайность. Расширение генетической базы земляники садовой за счет октоплоидных и полиплоидных форм диких видов обеспечит увеличение продуктивности новых сортов благодаря включению в генотип признаков адаптивности к биотическим и абиотическим факторам. Использование современных методов исследований (скрининг метаболомного и биохимического профиля, ДНК-паспортизация, молекулярное маркирование) повышает объективность исследований и эффективность селекционного процесса.

Ключевые слова: земляника, продуктивность, селекция, признак, сорт

PRODUCTIVITY OF GARDEN STRAWBERRY AND BREEDING POSSIBILITIES TO IMPROVE IT

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The information on the productivity trait of garden strawberry Fragaria × ananassa (Duchesne ex Weston) Duchesne ex Rozier has been summarized on the basis of literary sources. The productivity trait has a complex polygenic character. Independent inheritance of individual productivity components allows modeling their optimal combination in a new variety. The creation of highly productive varieties of strawberries is based on intraspecific hybridization. The established positive correlation between the size of berries and the yield indicates the possibility of increasing the productivity of varieties by using large-fruited forms in breeding. At present, in various zones of horticulture, researchers distinguish large-fruited varieties of domestic breeding: ‘Atlas’, ‘Bereginya’, ‘Vityaz’, ‘Grenada’, ‘Zabelinskaya’, ‘Kemiya’, ‘Nashe Podmoscovie’, ‘Nelly’, ‘Pervoklassnitsa’, ‘Rusich’, ‘Solnechnaya Polyanka’, ‘Solovushka’, ‘Festivalnaya Romashka’, ‘Fruktovaya’, ‘Tsaritsa’, ‘Yaponka’, and foreign breeding: ‘Alba’, ‘Asia’, ‘Elsanta’, ‘Finesse’,

‘Florense’, ‘Vivaldi’, ‘Galia’, ‘Jive’, ‘Joly’, ‘M. Champion’, ‘Merced’, ‘Murano’, ‘Onda’, ‘Roxana’, ‘Rumba’, ‘Tarda Vicoda’, ‘Vima Kimberly’, ‘Vima Tarda’, ‘Vima Rina’, ‘Vima Xima’. It was noted that when using the inbreeding method, it is possible to achieve a heterotic effect on the basis of the productivity trait. The method of genetic modifications of plants is of great importance for breeding of complex quantitative traits of productivity, including yield. Expansion of the genetic base of garden strawberry by means of octoploid and polyploid forms of wild species will ensure an increase in the productivity of new varieties due to the inclusion of traits of adaptability to biotic and abiotic factors in the genotype. The use of modern research methods (screening of the metabolomic and biochemical profile, DNA certification, molecular labeling) increases the objectivity of research and the efficiency of the breeding process.

Keywords: strawberry, productivity, breeding, trait, variety

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Garden strawberry is the most widespread berry crop in the world production. Its popularity is due to the high economic efficiency of cultivation, consumer demand, and the rich biochemical composition of fruits [1].

In 2019, 8.9 million tons of strawberries were produced in the world. The production leaders are China (3.21 million tons), the USA (1.02 million tons), Mexico (0.86 million tons), Turkey (0.47 million tons), Egypt (0.46 million tons). Russia ranks seventh in the world rating of producers - 0.21 million tonnes were produced in 2019[1].

The profitability of cultivation of garden strawberries is determined by the ratio of many factors, of which the most important at present is the yield [2].

The yield of a variety depends on the productivity potential inherent in the genotype, as well as on many external factors that affect its manifestation and the possibility of implementation in specific conditions [3, 4].

Increasing productivity in modern conditions is one of the main tasks of all breeding programs, regardless of the conditions of the zone for which the variety is being developed [5, 6].

The purpose of the research is to summarize information about the productivity trait in garden strawberries on the basis of domestic and foreign literary sources analysis to identify ways to increase the priority trait at the present stage of breeding development.

A sign of productivity, the components of productivity in garden strawberries. In biology productivity refers to the amount of biomass produced per unit of time[2]. Plant productivity in relation to agricultural fruit crops is the yield per plant[3]. Ultimately, the yield of the variety depends on the productivity - the amount of products obtained from a unit of area. The productivity of a plant is determined by its genotype and environmental conditions that influence the disclosure of the potential of a particular plant, variety [2, 3, 7]. The level of manifestation of the productivity trait in strawberry cultivars depends on the type of fruiting (short daylight cultivars, fall-bearing types, neutral day cultivars) and cultivation technologies (controlled conditions, open ground) [8–12].

The following components of productivity are distinguished in garden strawberry: the number of horns (shortened annual shoots), the number of peduncles, flowers, the number and weight of fruits

[1]FAO. FAOSTAT— Food and Agriculture Organization Corporate Statistical Database. [accessed May 13, 2021] URL: http: // www.fao.org/faostat/en/#data/QC (accessed May 13, 2021).

[2]Biological dictionary on-line //http://bioword.ru/B/B178.htm (accessed: 04/29/2021).

[3]Kudryavets R.P. Encyclopedic Dictionary of the Gardener / ed. I. M. Kulikov. Moscow: Ed. House of SMEs, 2007. 605 p.

per plant[4,5] [13]. Moreover, the components are specific for each variety. The number of fruits per plant and their average weight are of decisive importance [14]. Currently, varieties that are capable of producing 500–600 g of berries and more per bush are considered highly productive [15, 16].

With an optimal combination of productivity components, the number of flowers and ovaries on one peduncle is of great importance. Among the existing variety of the assortment, varieties with a large, medium and small number of flowers and ovaries on a peduncle stand out. Moreover, the more flowers and ovaries on one peduncle, the less the average weight of berries, as a rule. The number of peduncles per bush more than 10 in the second year of planting characterizes the variety as potentially highly productive [17, 18]. A large number of peduncles in strawberry plants can be caused both by the formation of many peduncles on a small number of horns, and by a large number of horns with several peduncles on each [19].

In most strawberry varieties, the first berry is much larger than the rest, which is a biological feature and is explained by the structure of the peduncle [20]. Considering the independent nature of the inheritance of individual components of productivity (the number of horns, the number of peduncles, the number of flowers and ovaries, the weight of the fruit), it is possible to identify donor forms of a high level of these traits (both individually and in combination) and include them in the breeding process as initial parent forms. In order to identify the optimal combination of productivity components in a new variety during the development of a breeding program, a model of the future variety is theoretically substantiated, which serves as the basis for subsequent selection in hybrid offspring[6] [21].

Selection of garden strawberry for high productivity. Among industrial berry crops, strawberry is one of the youngest domesticated plants [22]. The modern cultivated strawberry (Fragaria × ananassa) is the result of an accidental hybridization between Fragaria virginiana Mill. and Fragaria chiloensis (L.) Mill. in the 18th century. [23]. Interest in strawberries as an agricultural crop increased with the development of agricultural technologies, the processing industry, and the identification of the nutritional and dietary value of fruits. Professional researchers replaced amateur breeders from the beginning of the XX century. [24]. The high ecological plasticity of garden strawberry plants contributed to the rapid spread of the crop to various ecological-geographical zones and to the increased demand of producers for high-yielding varieties.

The creation of highly productive varieties of garden strawberry is based on the traditional method - intraspecific hybridization. The tasks of research in the implementation of breeding programs are to identify the mechanisms of inheritance of the studied traits, to develop methods for accelerating the breeding process and managing it. With the help of modern research methods, including biochemical, metabolomic analysis, genetic screening, DNA marking of traits, it was possible to significantly increase the objectivity of the assessment of both the initial material for selection and the resulting hybrid populations. The use of statistical methods for data analysis makes it possible to predict the breeding process and increase its efficiency. Attracting large-fruited varieties in the breeding process is the most frequently used method of increasing productivity in strawberries. The established positive correlation between the size of berries and productivity in garden strawberries indicates the possibility of breeding for high productivity by using large-fruited forms [25]. It is effective to cross large-fruited varieties with each other to increase large-fruitedness [26, 27]. Large-fruited varieties include varieties with an average berry weight of 9–12 g or more[7]. In the studies of V.I. Lapshin and V.V. Yakovenko [28], studying the coefficients of heritability h2 of the large-fruited trait for parental strawberry varieties, a high relationship between genotype and phenotype in the variability of the trait and pronounced additive genetic effects was established.

Such method of breeding for high productivity is inbreeding. However, this method has not found wide application due to the length and complexity of the process. At the same time, positive results were obtained - a heterotic effect was achieved in

[4]Zubov A.A. Genetic characteristics and selection of strawberries (guidelines). Michurinsk, 1990.81 p.

[5]A.A. Zubov, I.V. Popova Strawberry breeding. Program and methodology for breeding fruit, berry and nut crops / under the general editorship of E.N. Sedova. Oryol: VNIISPK; 1995.S. 387–416.

[6]Gorelikova O.A. Improvement of the assortment of garden strawberries for intensive cultivation technologies in the Krasnodar Territory: PhD in agriculture. Krasnodar: FGBNU SKZNIISVV; 2017.

[7]Shokaeva D.B., Zubov A.A. Wild strawberries, garden strawberries, zemklunika. Program and methodology for the variety study of fruit, berry and nut crops / under the general editorship of. E.N. Sedova, T.P. Ogoltsova. Oryol: VNIISPK, 1999.S. 417-443.

terms of productivity when crossing inbred seedlings (see footnote 4).

The rate of creation of new varieties of strawberries is quite high. Currently, there are about 15 thousand varieties, lines and forms in the world[8]. A comprehensive study of the genetic resources of the species Fragaria × ananassa has led to the fact that in recent years breeders have paid attention to the need to expand its genetic base [29]. In this regard, distant hybridization is of particular importance. Hybridization of pineapple strawberry varieties with forms of other octoploid species has the greatest effect [30].

At the present stage, instrumental research methods are widely integrated into selection. Molecular markers are actively used to identify DNA polymorphism, genetic diversity, and population structure of the germplasm set in Fragaria × ananassa [31]. Polygenic inheritance of economically valuable traits in Fragaria × ananassa (2n = 56) complicates the implementation of genetic control when planning breeding programs (see footnote 4). For selection based on characters with complex polygenic control, the method of genomic selection is effective, eliminating the need for many years of genetic research preceding the selection process [32].

Genetic modifications of plants, involving the insertion of genes for the transfer of certain traits that are not found in nature in a given species, are used for selection for resistance to abiotic and biotic factors. However, this technique currently has limited application for the selection of complex quantitative traits, which includes yield [33]. No high yielding varieties have been created by molecular methods alone [34].

The manifestation of potential productivity due to the genotype of the variety is significantly influenced by abiotic and biotic factors, technologies and methods of growing crops. In this regard, standards of high productivity are distinguished for various production zones. According to domestic researchers, currently in the Krasnodar Territory the most productive varieties are: ‘Nelly’ (1725.8 g / bush); ‘Vivaldi’, ‘Rumba’, ‘Joly’, ‘Jive’ (1044.7 g / bush) [16]. High productivity in combination with large fruit, density of berry pulp and the amount of sugars in this zone of horticulture are found in the following varieties: 'Vima Tarda', 'Onda', 'Vima Xima', 'Florense', 'Tarda Vicoda', 'Galia', 'Kemiya' , 'Nelly' [6]. According to the results of the assessment of biological productivity, the following varieties are considered the most productive in the Oryol region: 'Alba' (346 g / bush), 'Asia' (435), 'Tsaritsa' (463), 'Solovushka' (959 g / bush) [13]. In the conditions of the Central Black Earth Region of the Russian Federation, varieties are distinguished for high productivity in combination with other economically valuable traits: 'Roxana' (up to 675 g / bush), 'Alba', 'Vima Kimberly', 'Elsanta' (up to 540), ' Vima Rina ',' Florence '(486-513 g / bush) [35]. For the Non-Chernozem zone, the following are distinguished as the most productive varieties: 'Bereginya' (690.9 g / bush), 'Our Podmoskovye' (527.0), 'Rusich' (501.8), 'Solovushka' (467.5), 'Vityaz' (437.6 g / bush) [36]. The most productive varieties in the forest-steppe conditions of the Altai Territory are: 'Zabelinskaya' (up to 580 g / bush), 'First-grader' (up to 769), 'Solnechnaya Polyanka' (up to 602), 'Festivalnaya Daisy' (up to 583 g / bush) [37]. In the conditions of the Kamchatka Territory, varieties with high productivity potential have been identified: ‘Japanese’, ‘First-grader’, ‘Atlas’, ‘Fruit’, ‘Grenada’ (308.0–869.4 g / bush) [38].

Scientists from Brazil, based on the results of studying nine new varieties, identified the most productive ones: ‘Merced’ (453–547 g / bush) and ‘Camarosa’ (434–537 g / bush) [39]. The new European variety 'Rendezvous' is capable of producing up to 1300 g of fruits per plant [40]. According to East Malling researchers, the following varieties are highly productive: ‘Murano’ (613 g / bush), ‘M. Champion " (747), 'Finesse' (854 g / bush)[9].

The role of breeding in increasing the productivity of garden strawberries is obvious, since back in the 1980s. varieties with a yield of 250-350 g / bush (‘Festivalnaya’, ‘Komsomolskaya Pravda’, ‘Zarya’, ‘Purple’, ‘Generous’, ‘Pennant’, etc.) were classified as highly productive [10].

Independent inheritance of traits - productivity components - in garden strawberry allows selection according to the planned parameters of the combination of components.

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Expansion of the genetic base of Fragaria × ananassa with the involvement of other octoploid species and polyploid forms of wild species provides a further increase in productivity in new varieties due to the realization of the potential due to the inclusion of traits of adaptability to biotic and abiotic factors in the genotype.

The widespread involvement of modern methods of studying the phenotype and genotype (screening of the metabolomic and biochemical profile, DNA certification, molecular labeling) contributes to increasing the objectivity of research and the efficiency of the breeding process.

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ПРОМЫШЛЕННОЕ СКРЕЩИВАНИЕ КОРОВ МОЛОЧНОГО СКОТА С БЫКАМИ МЯСНЫХ ПОРОД В ЗАПАДНОЙ СИБИРИ

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Представлены результаты промышленного скрещивания коров молочного скота с быками мясных пород. Научно-хозяйственные опыты проведены в Омской и Новосибирской областях. Для эксперимента отобраны выранжированные коровы красной степной породы, которых искусственно осеменили семенем быков красной степной, калмыцкой и герефордской пород. От народившихся телят по методу групп-аналогов отобрали бычков каждого генотипа и сформировали три группы: 1-я контрольная – красные степные, 2-я опытная – помеси калмыцкая × красная степная, 3-я опытная – помеси герефордская × красная степная. Во втором опыте из бычков-кастратов симментальской породы и герефорд × симментальских помесей сформированы две группы: 1-я контрольная симментальской породы и 2-я опытная герефорд × симментальские помеси. Выявлено высокодостоверное превосходство по живой массе молодняка 2-й и 3-й опытных групп. С 9- и до 15-месячного возраста по сравнению с контрольными животными оно составило 16,5–77,3 кг ($p < 0,05$–$0,001$). У помесей группы красная степная × герефордская убойный выход составил 58,6%, что выше, чем у сверстников двух первых групп, на 1,9 и 1,8% ($p < 0,05$), масса туши 209,3 кг, у красных степных – 172,2 кг ($p < 0,01$). Во втором опыте в возрасте 8, 12, 15 и 18 мес бычки 2-й опытной группы превосходили сверстников 1-й контрольной на 15,2–29,4 кг ($p < 0,05$–$0,001$). Убойный выход у опытного молодняка был выше, чем у контрольного, – 57,8%. По двум опытам помесные группы животных характеризовались лучшей мясной продуктивностью. Промышленное скрещивание коров молочного скота с быками мясных пород позволит повысить мясную продуктивность и увеличить поголовье мясного скота.

Ключевые слова: промышленное скрещивание, мясная продуктивность, порода, живая масса

COMMERCIAL CROSS BREEDING OF DAIRY CATTLE WITH BEEF BULLS IN WESTERN SIBERIA

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The results of commercial cross breeding of dairy cattle with meat bulls are presented. Scientific and economic experiments were carried out in Omsk and Novosibirsk regions. Ranked cows of the red steppe breed were selected for the experiment. They were artificially inseminated with the semen of bulls of the red steppe, Kalmyk and Hereford breeds. Bulls of each genotype were selected from the calves born and three groups were formed by the method of analogue groups: 1st control – the red steppe, 2nd experimental – crossbreed of Kalmyk × the red steppe, 3d experimental – crossbreed

of Hereford × the red steppe. In the second experiment two groups were formed from castrated bulls of Simmental and Hereford breeds × Simmental hybrids: 1st control group of Simmental breed, 2nd experimental group – Hereford × Simmental hybrids. A highly reliable superiority in the live weight of young animals of the 2nd and 3d experimental groups was revealed. From the age of 9 to 15 months, it was 16.5–77.3 kg ($p < 0.05$–0.001) compared to animals in the control group. In the group of the red steppe × Hereford, slaughter yield was 58.6%, which is higher than that of the first two groups, by 1.9 and 1.8% ($p < 0.05$), the carcass weight was 209.3 kg, the red steppe – 172.2 kg ($p < 0.01$). In the second experiment at the age of 8, 12, 15 and 18 months, the bulls of the 2nd experimental group outperformed the peers of the 1st control group by 15.2–29.4 kg ($p < 0.05$–0.001). Their slaughter yield was higher than that of the control group, and accounted for 57.8%. In two experiments, crossbred groups of animals were characterized by a better meat productivity. Commercial crossbreeding of dairy cows with beef breeds of bulls allows to increase meat productivity and increase the population of the meat cattle.

Keywords: commercial crossbreeding, meet productivity, breed, live weight

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The authors declare no conflicts of interest.

INTRODUCTION

The development of the specialized meat cattle industry in Russia is a priority task of the agricultural sector. From 2010 to 2019, the production of large cattle for slaughter in live weight decreased from 3030.0 thousand tons up to 2827.1 thousand tons or 202.9 thousand tons (-6.7%), which is the result of the reduction in the number of dairy cows and over-represented young cattle in the share of fattening cattle[1] [1]. According to experts, they can only be replaced by animals of beef breed. According to forecasts, by 2025 the number of beef cattle of specialized beef breeds should be 10 million heads. [2]. The development of beef breeds occurs both when using reputable and foreign species, as a rule, regionalized to the conditions of a specific region [3–10].

It should be noted that it takes a long time to increase the number of beef cattle through own reproduction, and it is very expensive to buy animals from other countries. In this connection, we consider it expedient for Siberian producers to use selected (culled) dairy cows for industrial crossbreeding with bulls of beef breeds. This will make it possible to create a contingent of crossbred herds for beef production in contrast to purebred beef cattle [11-15]. The potential for beef cattle breeding in Siberia with its vast agricultural land is very high, so beef cattle breeding can become a profitable industry.

The aim of the study was to examine the productive features and meat qualities of young cows from industrial crossbreeding of bred dairy cows with bulls of specialized meat breeds in Western Siberia.

[1]Dunin I.M., Butusov D.V., Shichkin G.I., Safina G.F., Chernov V.V., Lastochkina O.V., Tyapugin S.E., Bogolyubova L.P., Nikitina S V.V., Matveeva E.A., Tyapugin E.E. The state of meat cattle breeding in the Russian Federation. Yearbook of pedigree work in meat cattle breeding in the farms of the Russian Federation, 2019. Moscow, 2020, 3 p.

MATERIALS AND METHODS

Two research experiments have been conducted. The first one was carried out in APC "Uraly" of the Omsk region by commercial crossbreeding of red steppe cows with Kalmyk and Hereford bulls to increase the quantity and improve the quality of beef in the farms of the tundra northern zone of Western Siberia. For this purpose, 62 red steppe cows were selected and then artificially inseminated with semen from red steppe, Kalmyk and Hereford bulls.

From the 60 calves born, 30 steers of 10 animals of each genotype were selected. Three groups were formed using the method of peer groups: 1st control - red steppe, 2nd experimental group - Kalmykian × red steppe, 3rd experimental group - Hereford × red steppe.

The second experiment was conducted in ZAO Kozinskoe, Novosibirsk Region on steer cattle of Simmental breed and Hereford × Simmental mixtures. Two groups of 10 animals each were formed: the 1st (control) of Simmental breed, and the 2nd (experimental) of Hereford × Simmental mixtures. Feeding and maintenance of the young animals in both experiments were the same. The steers were weighed monthly in the morning before feeding. The dynamics of their live weight were determined up to 15 months of age in the first experiment and up to 18 months of age in the second.

VASKHNIL, VIZH and VNIIMP methods were used to study the meat productivity and meat quality. For this purpose, control slaughter of steers was conducted at 15-months of age, 3 animals from each group in the first experiment and at 18-months of age in the second experiment. Pre-slaughter weight, carcass weight, carcass yield, weight of internal fat, slaughter weight and slaughter yield were taken into account. The data were processed by the method of variation statistics using the Snedecor software.

RESULTS AND DISCUSSION

The influence of Hereford and Kalmyk bulls in cross-breeding with red steppe cows on the variations of live-weight of young animals in the age dynamics during their breeding from 9- to 15-months of age has been established. At birth bulls of the red steppe breed and crossbreds of the 2nd group had almost the same weight (26,0 and 27,2 kg) in contrast to the 3rd group. The live weight of Group 3 steers was 2.7 kg more than that of Group 1 and 1.5 kg more than Group 2 ($p < 0.05$) (see Table 1).

The difference in live weight in the benefit of the place in comparison with the peers of the control group was especially significant after weaning from mothers. From this age and up to 15 months, the differences between the 2nd and 3rd groups in comparison with the control high are excellent. At the same time, a more intensive increase in live weight took place in mixed bred cattle. The differences in the 1st control and 2nd experimental groups by 15 months amounted to 41.6 kg in favor of the experimental animals, in the 1st and 3rd - 52.6 kg ($p < 0.001$). Probably, the high unpretentiousness of the Hereford crosses to keeping conditions in winter and early spring periods deter-

Табл. 1. Динамика живой массы бычков, кг ($M \pm m$)
Table. 1. Dynamics of live weight of bulls, kg ($M \pm m$)

Age, months	Group (n = 10)		
	1-st	2-nd	3-rd
At birth	26,0 ± 0,82	27,2 ± 0,73	28,7 ± 0,87^{*1}
9	219,8 ± 3,75	236,3 ± 5,92^{*1}	244,5 ± 2,36^{***1}
12	273,6 ± 5,46	296,0 ± 6,67^{*1}	317,8 ± 3,80^{***1*2}
15	333,5 ± 6,82	375,1 ± 4,92^{***1}	386,1 ± 4,13^{***1}

Hereinafter: 1, 2 - means the group number.
*$p < 0,05$.
**$p < 0,01$.
*** $p < 0,001$.

mined a higher rate of live weight gain of steers of the 3rd group.

To determine the slaughter performance of young red steppe bulls and their mixtures with Kalmyk and Hereford bulls, control bulls were slaughtered at the age of 15 months. Three bulls from each group were selected for slaughter, with relatively similar live weights and the same fatness before slaughter (see Table 2).

The slaughter yield of crossbred steers of Group 2 was 56.8% and that of Group 3 was 58.6%.

At 15 months of age the carcasses of crossbred Hereford steers were more complete - 209.3 kg, or 55.2%, and those of red steppe steers - 172.2 kg, or 52.6%.

The weight of the paired carcass of Kalmyk mixed breeds was 197.0 kg, which was higher than that of their red steppe counterparts. In general, the slaughter yield of Kalmyk mixed bred bulls was equal to that of red steppe peers (56.7% and 56.8%). In red steppe x Hereford steppe bulls, the slaughter yield was 58.6%, which was 1.9% and 1.8% higher than in the first two groups, respectively.

In the second experiment, at the age of 8 months, the bulls of the 2nd experimental group excelled in the live weight of the counterparts of the 1st control by 22.9 kg (10.8%) at $p < 0.001$ (see Table 3).

The tendency for the advancement in the live weight of gobies of the 2nd group by 15.2–29.4 kg ($p < 0.05$–0.001) was observed up to 18 months of age.

The indicators of the meat production of young animals according to the results of the control slaughter of gobies are given in table. 4.

The gobies of the 1st group had a high pre slaughter weight in the experience. They excelled their peers from the 2nd group by 34.1 kg. A similar situation was observed for the slaughter mass and the mass of a pair of carcasses. The advantage in terms of slaughter output was on the side of the bulls of the 2nd group - 57.8%.

CONCLUSION

The results of two scientific and economic experiments showed that crossbred gobies were characterized by the best meat production. Their superiority over control animals in live weight reached 15.8%, in slaughter yield

Табл. 2. Результаты контрольного убоя 15-месячных бычков ($M \pm m$)
Table. 2. Results of the control slaughter of 15-month-old bulls ($M \pm m$)

Indicator	Group		
	1-st	2-nd	3-rd
Live weight, kg: removable	338,0 ± 3,22	378,2 ± 2,49^{***1}	389,0 ± 3,57^{***1*2}
pre-slaughter	327,3 ± 3,15	364,6 ± 2,76^{***1}	379,1 ± 2,61^{***1**2}
Weight, kg: slaughter	186,0 ± 2,31	207,1 ± 2,50^{**1}	222,2 ± 2,23^{***1***2}
Hot carcass	172,2 ± 3,18	197,0 ± 2,15^{**1}	209,3 ± 2,20^{***1**2}
Slaughter yield, %	56,7	56,8	58,6
Carcass yield, %:	52,6	54,0	55,2

Табл. 3. Динамика живой массы симментальских, герефорд × симментальских бычков, кг
Table. 3. Dynamics of live weight of Simmental, Hereford × Simmental bulls, kg

Age, months	Group (n = 10)	
	1-st	2-nd
8	212,2 ± 3,38	235,1 ± 3,78***
12	282,4 ± 5,81	311,8 ± 3,43***
15	328,0 ± 4,36	343,2 ± 5,13*
18	368,3 ± 5,20	397,7 ± 4,52***

Табл. 4. Результаты контрольного убоя подопытных бычков ($M \pm m$)

Table. 4. Results of control slaughter of experimental bulls ($M \pm m$)

Indicator	Group	
	1-st	2-nd
Weight, kg:		
removable	350,6 ± 3,28	382,0 ± 3,45***
pre-slaughter	335,1 ± 4,20	382,3 ± 14,4***
slaughter	184,3 ± 4,26	212,3 ± 3,77***
Hot carcass	170,0 ± 3,19	197,0 ± 4,15***
Yield, %:		
slaughter	55,3	57,8
carcass	50,6	53,6

- up to 2.5%. In our opinion, it is necessary to make wider use of industrial crossbreeding of selected (culled) dairy cows with bulls of beef breeds, which will increase meat productivity and increase the number of beef cattle, especially in private subsidiary farms and farming households.

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ВЛИЯНИЕ НАНОЧАСТИЦ СЕРЕБРА НА БАКТЕРИЦИДНУЮ АКТИВНОСТЬ И АНТИБИОТИКОЧУВСТВИТЕЛЬНОСТЬ *SALMONELLA ENTERITIDIS* 182

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Проведены исследования по определению синергетического эффекта применения комбинаций антибактериальных веществ, включающих антибиотики, дезинфектант септабик и AgNPs. Выявлен значительный рост бактерицидной активности в комбинации септабик + AgNPs + нитокс и септабик + AgNPs + цефтиофур. Определение чувствительности *Salmonella enteritidis* 182 к антибактериальным препаратам показало наличие устойчивости к 8 (38,1%) препаратам, малой чувствительности – к 7 (33,3%), чувствительности – к 6 (28,6%) и отсутствие препаратов с высокой чувствительностью. После культивирования *S. enteritidis* 182 с антибактериальными препаратами и их комбинациями установлено увеличение количества лекарственных средств, к которым изучаемый штамм был чувствителен. Наличие чувствительности выявлено к 7–10 препаратам, что на 4,7–19,6% больше, чем в контрольных показателях. Установлена ранее отсутствовавшая высокая чувствительность к 2–8 (9,5–38,0%) антибактериальным препаратам. Культивирование *S. enteritidis* 182 с AgNPs показало наивысший рост антибиотикочувствительности из всех изучаемых комбинаций антибактериальных средств в виде увеличения размера задержки роста. Это дает основание предположить о ведущей роли AgNPs в преодолении антибиотикорезистентности. Инкубирование *S. enteritidis* 182 после контакта с септабиком и арговитом вызвало максимальное увеличение диаметра задержки роста микроорганизма без снижения показателя к отдельным видам препаратов (за исключением септабика, где установлена утрата чувствительности к тилозину). При добавлении различных антибиотиков к комбинации септабик + арговит отмечены факты снижения зоны задержки роста или его исчезновения. Описан комбинированный эффект сочетанного применения антибактериальных препаратов и наночастиц серебра в отношении бактерий с множественной лекарственной устойчивостью.

Ключевые слова: наночастицы серебра, *Salmonella enteritidis*, антибиотики, антибиотикорезистентность, AgNPs, септабик

EFFECT OF SILVER NANOPARTICLES ON BACTERICIDAL ACTIVITY AND ANTIBIOTIC SENSITIVITY OF *SALMONELLA ENTERITIDIS 182*

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Studies have been carried out to determine the synergetic effect of the use of combinations of antibacterial substances, including antibiotics, septabic disinfectant and AgNPs. A significant increase in bactericidal activity was revealed in the combination of septabic + AgNPs + nitox and septabic + AgNPs + ceftiofur. Determination of the sensitivity of *Salmonella enteritidis* 182 to antibacterial drugs showed the presence of resistance to 8 drugs (38.1%), low sensitivity to 7 (33.3%), sensitivity to 6 (28.6%) and the absence of preparations with high sensitivity indicators. After cultivation of *S. enteritidis* 182 with antibacterial drugs and their combinations, an increase in the number of preparations to which the studied strain was sensitive was found. The presence of sensitivity to 7–10 drugs was revealed, which is 4.7–19.6% higher than in the control indicators. A previously absent high sensitivity to 2–8 antibacterial drugs (9.5–38.0%) was established. The cultivation of *S. enteritidis* 182 with AgNPs showed the highest increase in antibiotic sensitivity of all the studied combinations of antibacterial agents in the form of the growth inhibition size increase. This suggests

a leading role of AgNPs in overcoming antibiotic resistance. Incubation of *S. enteritidis 182* after contact with septabic and argovite caused a maximum increase in the diameter of the growth inhibition of the microorganism without decreasing sensitivity to certain types of drugs (with the exception of septabic, where the loss of sensitivity to tylosin was established). When adding various antibiotics to the combination of septabic + argovit, a decrease in the growth inhibition zone or its disappearance were noted. The combined effect of the use of antibacterial drugs together with silver nanoparticles against bacteria with multidrug resistance is described.

Keywords: silver nanoparticles, *Salmonella enteritidis*, antibiotics, antibiotic resistance, AgNPs, septabik

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INTRODUCTION

The widespread use of antibacterial drugs in animal production has led to an increase in the isolation of antibiotic-resistant strains of microorganisms. This has significantly reduced the effectiveness of chemotherapy and has created conditions for the transmission of antibiotic-resistant genes through animal products to humans. In 2001 The UN WHO recognized the problem of antibiotic resistance as a global problem and developed a series of interventions to limit the occurrence and spread of this phenomenon, which equally affects both medical and veterinary service delivery. Carbapenem-resistant micro-organisms of the family Enterobacteriaceae are seen as a threat to public health. Bacterial strains isolated from cattle are a major channel for this resistance. Damages in the USA from infections caused by antibiotic-resistant microorganisms are estimated at $55-70 billion; similar losses in Europe exceed €1.5 billion per year[1, 2] [1].

A group of intestinal pathogens of the *Salmonella* genus, which can cause pathologies in animals and poultry and food poisoning in humans, pose a particular threat. Given the additional risks posed by persistent pathogens associated with the presence of antibiotic-resistant genes, the problem of infectious pathologies is becoming alarming and requires urgent solutions.

Research is underway to develop veterinary medicines that do not contain antibiotics but have antibacterial properties. One well-known method of treating and preventing infectious diseases is the use of silver preparations. These drugs are widely used in the treatment of gastrointestinal diseases of humans and animals, obstetric and gynecological (mastitis, endometritis) pathologies of cattle, as well as the treatment of wounds, ulcers, bedsores in surgical practice[3] [2-5]. In addition to the bactericidal properties of silver preparations, studies have revealed the presence of silver nanoparticles (AgNPs) to overcome and reduce the antibiotic

[1]World Health Organization. WHO Global Strategy for Containment of Antimicrobial Resistance. Geneva, 2001. WHO/CDS/CSR/DRS/2001.2.

[2]Strategy for preventing the spread of antimicrobial resistance: order of the Government of the Russian Federation No. 2045-r dated September 25, 2017.

[3]Blagitko E.M., Bugaychenko N.V., Shorina G.N. and others. Results of local application of argolite and hydropenta – silver-containing preparations on a natural mineral basis. Nanotechnologies and nanomaterials for biology and medicine: materials of scientific and practical research. conf. with int. participation: 2 hours. Novosibirsk, 2007. Part 2. P. 39–49.

resistance of opportunistic and pathogenic microflora[4]. Positive experience has been gained in the use of disinfectants as antibacterial medicines (ecocidal in oral administration), which opens up prospects for the use of these agents as therapeutic agents [6]. The need therefore arises to investigate the presence of possible synergistic properties in the combined use of antibacterial agents of different pharmacological groups.

A study of the effect of colloidal silver on the morphology and development of *Salmonella enteritidis* cell populations using scanning electron microscopy showed that AgNPs in low concentrations promotes partial destruction of the bacterial cell wall, prevents normal cell division, initiates the processes of heteromorphism. However, the S. enteritidis population remains viable. Once in a favorable environment, it fully restores its morphological properties and the possibility of growth and development (bacteriostatic effect). At high concentrations, AgNPs causes rapid cell death in the population. Thus, the bactericidal effect of colloidal silver solutions depends directly on the concentration of nanoparticles in them. The higher their concentration, the deeper the lesion of cellular structures, the more pronounced is the disinfecting effect of the applied preparation [7].

It was found that nanosized systems can not only improve the therapeutic activity of antibacterial agents, but also inhibit the stimulation of resistance by overcoming resistance strategies developed by bacteria, including the degradation of the drug under the action of β-lactamase, thickening the walls of bacterial cells [8]. The synergistic effect of AgNPs in combination with gentamicin against *Escherichia coli, Proteus mirabilis, Pseudomonas aeruginosa, Salmonella typhi* and *Staphylococcus aureus* showed a significant increase in the diameter (up to 26-34 mm) of bacterial growth retention zones [9].

The aim of the study was to investigate the bactericidal properties of various combinations of antibacterial agents and their effect on the possibility of overcoming antibiotic resistance in *S. enteritidis* 182.

MATERIAL AND METHODS

A preparation argovit containing AgNPs (12-14 μg/ml) was used for the study; ceftimag, containing ceftiofur hydrochloride 100 mg as an active substance in 1 ml; containing methyl ether of paraoxybenzoic acid 1.8 mg, propyl ether of paraoxybenzoic acid 0.2 mg and propylene glycol dicaprylate/dicaprates up to 1 ml; oxytetracycline as a 10% aqueous solution; enrofloxacin containing 50 mg of enrofloxacin in 1 ml; gentamicin, containing 40 mg of gentamicin sulfate in 1 ml; nitox, containing 200 mg of oxytetracycline dihydrate as an active ingredient in 1 ml of the formulation; excipients - magnesium oxide, N,N-dimethylacetamide, rongalit, monoethanolamine and water for injection; azithromycin in the form of azithromycin citrate derived from azithromycin dihydrate, 100 mg in 1 ml; excipients - citric acid monohydrate, sodium hydroxide, water for injection.

The disinfectant Septabic contains the active ingredient urea clathrate didecyldimethylammonium bromide, which is active against Gram-positive and Gram-negative bacteria (including Mycobacterium tuberculosis), dermatophytes, yeast-like fungi, candida, hepatitis B virus, HIV, influenza viruses, parainfluenza[5].

Microbial sensitivity of the *S. enteritidis* ATCC 182 reference strain to antibacterial agents and combinations was determined from a dilution with the lowest bacteriostatic concentration, 0.2 ml of which was added to an AMP and the antibiotic sensitivity of the microorganisms was determined by discodiffusion.

Determination of sensitivity was carried out to 21 antibacterial drugs with subsequent incubation for 24 hours at a temperature of 37.5 ± 0.5 0 C.The sensitivity of microorganisms to antibiotics was determined by the degree of delay in the growth diameter around the disc:

[4]Mamonova I.A. The influence of nanoparticles of the transition group of metals on antibiotic-resistant strains of microorganisms: author. dis. PhD in biology. M., 2013. 21 p.

[5]https://infodez.ru/product/1452_septabik.html

up to 10 mm - resistant, up to 15 - insensitive, up to 20 - sensitive, more than 20 mm - highly sensitive[6].

RESULTS AND DISCUSSION

Determination of the bactericidal activity of antibacterial agents in combination with AgNPs and septabic disinfectant is justified by its possible use in the treatment of infectious pathologies due to acceptable parameters of acute toxicity, which refers to class 3 (GOST 12.1.007-76) of moderately hazardous compound when introduced in the stomach and to class 4 of low-hazardous compound when applied to skin and when inhaled at saturation concentration. AgNPs has a mild sensitising effect[7].

Studies carried out to determine the synergistic effect of combinations of antibacterial agents including antibiotics, disinfectant preparation and AgNPs showed a significant increase in bactericidal activity in the combination septabic + AgNPs + nitox and septabic + AgNPs + ceftiofur (see Table 1).

Determination of the sensitivity of *S. enteritidis* 182 to antibacterial agents showed resistance to 8 (38.1%), low sensitivity to 7 (33.3%), sensitivity to 6 (28.6%) and no drugs with high sensitivity. After culturing *S. enteritidis* 182 with antibacterial agents and their combinations an increase in the number of drugs to which the studied strain was sensitive was found. The presence of sensitivity was detected to 7-10 drugs, which is 4.7-19.6% more than in the target scores. A previously absent high sensitivity to 2–8 (9.5–38.0%) antibacterial drugs was established (see Table 2).

The cultivation of *S. enteritidis* 182 with AgNPs showed the highest increase in antibiotic sensitivity among all the studied combinations of antibacterial agents in the form of the largest growth retardation of the microorganism, which suggests the leading role of AgNPs in overcoming antibiotic resistance. Incubation of *S. enteritidis* 182 after contact with septabic and argovit caused a maximum increase in the diameter of the growth retardation of the microorganism without a decrease in the indicator for certain types of drugs (with the exception of septabic, where the loss of sensitivity to tylosin was established). With the addition of various antibiotics to the combination of septabic + argovit, facts of a decrease in the growth retardation zone or its disappearance were noted. Thus, enrofloxacin reduced the diameter of the growth retardation of a microorganism or led to its disappearance by 6 (28.6%) drugs, oxytetracycline, gentamicin - by 4 (19.0%), nitox, ceftiofur, azithromycin - by 2 (9.5%) ... In these cases, in all combinations, a loss of sensitivity to tylosin and neomycin was noted when azithromycin, enrofloxacin, nitox and ceftiofur were added to the combination of septabic + argovit (see Tables 3, 4).

The results of the conducted studies indicate the presence of synergistic properties of the combined use of antibacterial drugs and confirm previous studies by other authors. The

Табл. 1. Чувствительность *S. enteritidis* 182 к антибактериальным препаратам и их комбинациям

Table 1. Sensitivity of *S. enteritidis* 182 to antibacterial drugs and their combinations

Preparation	Concentration, μg/ml		
	AgNPs	Septabic	Antibiotic
AgNPs	115	–	–
Septabic	–	312	–
Septabic + AgNPs	0,2	156	–
Septabic + AgNPs + oxytetracycline	0,1	0,78	0,78
Septabic + AgNPs + azithromycin	0,05	0,39	0,39
Septabic +AgNPs + gentamicin	0,05	0,39	0,39
Septabic + AgNPs + enrofloxacin	0,05	0,39	0,39
Septabic + AgNPs + nitox	0,00125	0,0975	0,0975
Septabic + AgNPs + ceftiofur	0,00125	0,0975	0,0975

[6]Determination of the sensitivity of microorganisms to antibacterial drugs: guidelines MUK 4.2. 1890-04, CRIE. M., 2004.101 p.
[7]https://infodez.ru/product/1452_septabik.html

combined effect of the combined use of antibacterial drugs and silver nanoparticles against bacteria with multidrug resistance is described. It was found that the synergistic effect of the antibiotics such as ciprofloxacin, imipenem, gentamicin, vancomycin, trimethoprim and nanoparticles led to an increase in antibacterial activity 0.2-7.0 times (on average 2.8). This shows that nanoparticles can be effectively used in combination with antibiotics to increase their effectiveness against various pathogenic microorganisms [10]. The antibacterial effect of the combined use of AgNPs and vancomycin against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Streptococcus pneumonia* has been established [11].

Synergistic antibacterial activity in combination of AgNPs with tetracycline, neomycin and penicillin was tested against antibiotic-resistant *S. typhimurium* DT104. A dose-dependent inhibition of the growth of bacteria *S. typhimurium* DT104 was noted for the complexes tetracycline - AgNPs and neomycin - AgNPs (0.07 and 0.43 μg / ml, respectively). At the same time, the combination of penicillin - AgNPs did not inhibit the growth of the microorganism [12]. The antimicrobial activity of the synthesized bioconjugates AgNPs and endogenous antibiotics was studied. Conjugates of AgNPs with peptide C-Bac3.4 have antimicrobial activity, including against the antibiotic-resistant *P. aeruginosa* strain, as well as the methicillin-resistant *St. aureus* ATCC 33591. Studies have shown that the complexes of AgNPs with antimicrobial peptides do not exhibit the pronounced membranolytic effect inherent in peptides [13].

AgNPs, synthesized by *A. calcoaceticus* LRVP54 for 24 h, in the form of monodisperse spherical nanoparticles with a size of 8–12 nm, were obtained from metal salts of silver nitrate 0.7 mm in size with subsequent heating at a temperature of 70 °C. The disc diffusion method established a higher antibacterial activity against gram-negative microorganisms. A high synergistic antibacterial effect (3.8 times) of the use of AgNPs and vancomycin for *Enterobacter aerogenes* was established, while a decrease in the minimum bactericidal concentration was observed. It is noted that multidrug-resistant *Acinetobacter baumannii* is highly sensitive in the presence of AgNPs and to antibiotics, with the exception of cephalosporins. A similar effect was observed when AgNPs were added to the culture broth with vancomycin-resistant *Streptococcus mutans*. Thus, biogenically synthesized AgNPs showed a significant synergistic antibacterial effect on β-lactam antibiotics [14].

It has been found that the combination of silver and sulfadimethoxine makes it possible to increase the antibacterial activity and create a drug to which bacteria are not addictive at certain concentrations of silver in solution. Preparations with AgNPs and sulfadimethox-

Табл. 2. Изменение чувствительности *S. enteritidis* 182 после контакта с антибактериальными средствами и их комбинациями

Table 2. Changes in the sensitivity of *S. enteritidis* 182 after contact with antibacterial agents and their combinations

Preparation	Number of preparations							
	У	%	М	%	Ч	%	В	%
Argovit	1	4,8	3	14,3	12	57,1	5	23,8
Septabic	4	19,0	2	9,5	10	47,6	5	23,8
Septabic + AgNPs	3	14,3	2	9,5	10	47,6	6	28,6
Septabic + AgNPs + oxytetracycline	8	38,0	3	14,3	7	33,3	3	14,3
Septabic + AgNPs + azithromycin	3	14,3	2	9,5	8	38,0	8	38,0
Septabic + AgNPs + gentamicin	7	33,3	2	9,5	9	42,74	3	14,3
Septabic + AgNPs + enrofloxacin	12	57,0	–	–	7	33,3	2	9,5
Septabic + AgNPs + nitox	4	19,0	1	4,75	10	47,6	6	28,6
Septabic + AgNPs + ceftiofur	6	28,6	3	14,3	8	38,0	4	19,0

Note. Y - stable, M - insensitive, H - sensitive, V - highly sensitive.

Табл. 3. Изменение зоны задержки роста *S. enteritidis* 182 после контакта с антибактериальными препаратами и их комбинациями

Table 3. Changes in the growth inhibition zone of *S. enteritidis* 182 after contact with antibacterial drugs and their combinations

Preparation	Control group	Septabic	%	Septabic + AgNPs	%	Septabic + AgNPs + oxytetracycline	%	Septabic + AgNPs + azithromycin	%	Septabic + AgNPs + gentamicin	%
Ampicillin / sulfabactam	–	–	–	21	100	–	–	11	100	–	–
Ampicillin	15	20	33,3	22	46,7	14	–6,7	21	40	–	–100
Amikacin	–	21	100	20	100	20	100	22	100	20	100
Benzylpenicillin	–	–	–	19	100	–	–	–	–	–	–
Gentamicin	15	18	20	16	6,7	20	33,3	20	33,3	20	33,3
Doxycycline	15	13	–13,3	16	6,7	–	–100	16	6,7	13	–13,3
Polymyxin	-	15	100	19	100	–	–	15	100	15	100
Carbecillin	20	21	5	21	5	21	5	25	25	23	15
Norfloxacin	–	24	100	–	–	23	100	21	100	22	100
Neomycin	16	18	12,5	18	12,5	18	12,5	–	–100	20	25
Enrofloxacin	17	18	5,9	18	5,9	20	17,6	22	29,4	20	17,6
Ciprofloxacin	20	25	25	23	100	21	5	21	5	25	25
Tetracycline	–	20	100	15	100	14	100	18	100	17	100
Oxytetracycline	15	18	20	13	13,3	13	–13,3	17	13,3	18	20
Lincomycin	–	16	100	–	–	–	–	20	100	–	–
Tylosin	15	–	–100	21	27,0	–	–100	–	-100	–	–100
Levomycetin	15	20	33,3	16	6,7	16	6,7	22	46,7	20	33,3
Streptomycin	15	20	33,3	18	20	20	33,3	21	40	16	6,7
Ticarcillin / clavulanic acid	19	20	5,3	24	26,3	16	–15,8	20	5,3	–	–100
Ofloxacin	16	23	43,7	17	6,2	–	–100	20	25	17	6,2
Rifampicin	–	–	–	–	–	–	–	20	100	–	–

ine have less antibacterial activity compared to complexes of silver in the ionic form, but the antimicrobial activity of preparations with AgNPs becomes less dependent on the concentration and nature of the substrate[8].

The synergistic and antagonistic effects of the combined use of metal nanoparticles and antibiotics against pathogenic strains of microorganisms are noted. AgNPs and ZnONPs exhibited increased antibacterial activity with increasing concentration against *S. aureus*, *E. coli*. The synergistic effect of the antibiotics such as azithromycin, cefotaxime, cefuroxime, fosfomycin and chloramphenicol against *E. coli* and *S. aureus* was significantly higher in the presence of AgNPs compared with antibiotic use alone (see footnote 8).

The synergistic effect of the antibiotics such as azithromycin, oxacillin, cefotaxime, cefuroxime, fosfomycin, and oxytetracycline against *E. coli* was significantly higher in the presence of ZnONPs compared with the use of the antibiotic alone. The synergistic effect of the antibiotics azithromycin, cefotoxime, cefofoxime, cefosfimoxime, cefimpoxim, cefofoxime, cefosfimoxime, cefoximol, and oxytetracycline against *S. aureus* was also significantly higher in the presence of ZnONPs compared to monouse of antibiotics [15].

[8] Lopanov A.N., Lysykh V.A. Antibacterial activity of sulfadimethoxine with silver. Perspective scientific research: experience, problems and development prospects: materials of the international scientific conference. Ufa, 2020, pp. 93–100.

Табл. 4. Изменение зоны задержки роста *S. enteritidis* 182 после контакта с антибактериальными препаратами и их комбинациями

Table 4. Changes in the growth inhibition zone of *S. enteritidis* 182 after contact with antibacterial drugs and their combinations

Preparation	Control group	Argo-vit	%	Septabic + Ag + enrofloxacin	%	Septabic + AgNPs+ nitox	%	Septabic + AgNPs + ceftiofur	%
Ampicillin / sulfabactam	–	12	100	–	–	13	100	13	100
Ampicillin	15	21		–	–100	22	46,7	20	33,3
Amikacin	–	20	100	–	–	20	100	21	100
Benzylpenicillin	–	11	100	–	–	16	100	–	–
Gentamicin	15	20	33,3	22	46,7	24	60	19	26,7
Doxycycline	15	16	6,7	16	6,7	16	6,7	13	–13,3
Polymyxin	–	16	100	–	–	20	100	16	100
Carbecillin	20	23	15	–	–100	23	15	–	–20
Norfloxacin	–	21	100	22	100	25	100	20	100
Neomycin	16	18	12,5	–	–100	–	–100	13	–18,7
Enrofloxacin	17	21	23,5	20	–17,6	20	17,6	22	29,4
Ciprofloxacin	20	21	5	–	–100	23	15	23	15
Tetracycline	–	20	100	–	–	20	100	–	–
Oxytetracycline	15	18	20	20	33,3	20	33,3	16	6,7
Lincomycin	–	–	–	–	–	–	–	–	–
Tylosin	15	16	6,7	–	–100	–	–100	–	–100
Levomycetin	15	16	6,7	20	33,3	20	33,3	16	6,7
Streptomycin	15	20	33,3	20	33,3	20	33,3	20	33,3
Ticarcillin / clavulanic acid	19	20	5,3	20	5,3	22	15,8	25	31,6
Ofloxacin	16	18	12,5	19	18,7	18	12,5	18	12,5
Rifampicin	–	10	100	–	–	10	100	–	–

Literature data confirm the different antibacterial effect of using AgNPs and antibiotics against bacteria isolated from animals that exhibit resistance to antibiotics. The inhibitory concentration was calculated to classify the observed collective antibacterial activity as synergistic, additive (only the sum of individual drug effects), indifferent (no effect), or antagonistic. Of the 40 tests performed, 7 were synergistic, 17 were additive and 16 were indifferent. None of the combinations tested showed an antagonistic effect. Most synergistic effects were observed for combinations with AgNPs co-administered with gentamicin. The greatest increase in antibacterial activity was found in combination therapy with penicillin G against *Actinobacillus pleuropneumoniae*, *Actinobacillus pleuropneumoniae* and *Pasteurella multocida*. Initially resistant to amoxicillin, gentamicin and colistin, they are susceptible to these antibiotics in combination with AgNPs. Research shows that AgNPs have potential as adjuvants for the treatment of bacterial diseases in animals [16].

Synergistic action of AgNPs in combination with erythromycin and levofloxacin against *St. aureus* was observed. Antimicrobial activity with antibiotics compared to pure silver nanoparticles was increased by a factor of 1.16-1.32. This synergism may be relevant for the treatment of infections caused by multidrug-resistant bacteria [17].

Analysis of the research results of many authors shows both a marked increase in the bactericidal activity of antibiotics when combined with AgNPs and its decrease. Given the variety of antibacterial drugs used in medicine and agriculture, this mixed result requires further research in the search for new combinations with AgNPs.

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ЦИТОГЕНЕТИЧЕСКИЕ НАРУШЕНИЯ У МОЛОДНЯКА КРУПНОГО РОГАТОГО СКОТА ПРИ ВАКЦИНАЦИИ ПРОТИВ САЛЬМОНЕЛЛЕЗА

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Изучен спектр и определены частоты цитогенетических нарушений у молодняка крупного рогатого скота, иммунизированного вакциной против сальмонеллеза телят. Исследования проведены в хозяйстве Новосибирской области на 10 клинически здоровых голштинизированных черно-пестрых телятах 10–17-дневного возраста. Использована концентрированная формолквасцовая вакцина против сальмонеллеза (паратифа) телят в дозе 2 мл (реиммунизация в дозе 2 мл) с интервалом 10 сут между инъекциями. Вакцина изготовлена из культуры бактерий штамма *Salmonella dublin* № 373, инактивированного формалином с добавлением алюмокалиевых квасцов и хлорида кальция. Цитогенетический анализ периферической крови у телят проведен до вакцинации (контроль), через 2 и 9 сут после вакцинации, через 2 и 9 сут после ревакцинации. Установлено, что спектр соматической хромосомной нестабильности в лимфоцитах периферической крови телят после вакцинации и ревакцинации представлен полиплоидией, гипоплоидией и гиперплоидией, хроматидными и хромосомными разрывами, одиночными и парными фрагментами хромосом. Выявлено, что спектр соматической хромосомной нестабильности после двукратных иммунизаций инактивированной вакциной против сальмонеллеза не отличался от спектра спонтанно возникающих мутаций у данного вида. Вакцинация и последующая ревакцинация телят в сравнении с довакцинационным периодом не приводили к достоверному увеличению частот анеуплоидных и полиплоидных клеток. При двукратной иммунизации телят в лимфоцитарных клетках крови животных отмечен волновой характер в вариации частот геномных мутаций от максимальных до минимальных значений аналогично продленному мутагенезу. Обнаружена тенденция увеличения частоты структурных нарушений хромосом через 2, 9 сут после вакцинации и 2 сут после ревакцинации. Отмечено достоверное возрастание в 2,9 раза частоты клеток с хромосомными аберрациями в лимфоцитах крови животных через 9 сут после их повторной иммунизации за счет разрывов и парных фрагментов хромосом. После вакцинации и ревакцинации хроматидные разрывы наиболее часто регистрировались в медиальных районах одной из хроматид, хромосомные разрывы в медиальных и теломерных районах обеих хроматид.

Ключевые слова: геномные мутации, хромосомные мутации, лимфоциты периферической крови, телята, вакцинация, ревакцинация, сальмонеллез

CYTOGENETIC ABNORMALITIES IN YOUNG CATTLE DURING VACCINATION AGAINST SALMONELLOSES

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The spectrum and the frequencies of cytogenetic abnormalities in young cattle immunized with a vaccine against salmonellosis of calves were investigated. The study was carried out on the farm of Novosibirsk region on 10 clinically healthy Holstein black-and-white calves of 10–17 days of age. A concentrated formol-alum vaccine against salmonellosis (paratyphoid) of calves was used at a dose of 2 ml (reimmunization at a dose of 2 ml) with an interval of 10 days between injections. The vaccine was made from the culture of bacteria of the *Salmonella dublin* strain № 373, inactivated with formalin with the addition of potassium alum and calcium chloride. Cytogenetic analysis of peripheral blood in calves was carried out before vaccination (control), 2 and 9 days after vaccination,

2 and 9 days after revaccination. It was found that the spectrum of somatic chromosomal instability in peripheral blood lymphocytes of calves after vaccination and revaccination is represented by polyploidy, hypoploidy and hyperploidy, chromatid and chromosomal breaks, single and paired fragments of chromosomes. It was revealed that the spectrum of somatic chromosomal instability after double immunizations with an inactivated vaccine against salmonellosis did not differ from the spectrum of spontaneously occurring mutations in this species. Vaccination and subsequent revaccination of calves in comparison with the pre-vaccination period did not lead to a significant increase in the frequency of aneuploid and polyploid cells. During double immunization of calves, a wave pattern in the variation of genomic mutation frequencies from maximum to minimum values in the lymphocytic blood cells of animals was noted, similar to prolonged mutagenesis. A tendency was found for the frequency of structural chromosome abnormalities to increase 2 and 9 days after vaccination and 2 days after revaccination. There was a credible 2.9-fold increase in the frequency of cells with chromosomal aberrations in the blood lymphocytes of animals 9 days after their repeated immunization due to breaks and paired fragments of chromosomes. After vaccination and revaccination, chromatid breaks were most often recorded in the medial regions of one of the chromatids, and chromosomal breaks in the medial and telomeric regions of both chromatids.

Keywords: genome mutations, chromosomal mutations, peripheral blood lymphocytes, calves, vaccination, revaccination, salmonellosis

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INTRODUCTION

An increase in livestock production in modern conditions is based on improving the genetic potential of animals and creating highly productive herds resistant to diseases[1] [1–4].

It has now been established that a number of infectious diseases are caused by pathogenic microorganisms. Their impact contributes to the appearance of abnormalities in the chromosomal apparatus of humans and laboratory animals. In medicine, numerous studies have been carried out to study the cytogenetic consequences of infectious diseases and vaccinations [5–8]. It has been suggested that antigens and toxins of some infectious agents and vaccines may be mutagenic. Reactive metabolites of oxygen and nitrogen, formed in the foci of inflammation and in the course of the immune response, can cause an increase in the frequency of DNA damage and cytogenetic disorders [5, 6].

The influence of infectious agents, including vaccines, on the instability of karyotypes in hu-

[1]Patent RU No. 2 083 102 C1 IPC A 01 K 67/02. The method of complex selection of bulls-producers for the resistance of offspring to diseases / V.L. Petukhov, L.K. Ernst, A.G. Nezavitin, A.I. Zheltikov, O.S. Korotkevich, S.G. Kulikova, V.G. Marenkov, N.N. Kochnev, V.P. Shishkov, V.T. Khristenko; applicant and patentee Scientific Research Institute of Veterinary Genetics and Breeding. No. 95113836/13; declared 08/01/1995; publ. 10.07.1997, Bul. No. 19. 10 p.

mans and laboratory animals is often ambiguous. According to N.N. Ilyinskikh [5], this is explained not only by the strain characteristics of infectious agents, but also by the specificity of the genotype of the organism, its age, the functional state of protection and resistance to the mutagenic influence of infectious factors.

In veterinary medicine and farm animal genetics, there are few publications on the subject, despite the need for research due to the widespread use of vaccination in agriculture[2,3] [9].

Salmonellosis (paratyphoid fever) is one of the most common infectious diseases of young cattle in Siberia [10]. It is caused by bacteria of the genus *Salmonella* (*S. dublin*, less often *S. typhimurium* and bacteria of other serological types), which primarily affect young animals aged from 10 days to 5 months. In young animals, the acute course of the disease is characterized by fever, septicemia, toxicosis and diarrhea, subacute and chronic - pneumonia and arthritis; in adult females, abortion. Vaccination is still the most effective way to prevent the spread of infectious diseases in farm animal populations. To date, the cytogenetic consequences of vaccinations against salmonellosis in cattle have not been investigated.

The aim of the work is to study cytogenetic disturbances in young cattle when vaccinated against Salmonellosis.

The research objectives are:

– determination of the spectrum of cytogenetic disorders in peripheral blood lymphocytes in calves at different periods of immunization against salmonellosis,

– investigation of the effect of double immunizations with an inactivated vaccine against salmonellosis on the frequency of cytogenetic disorders in immunocompetent cells in young cattle.

MATERIAL AND METHODS

The studies were conducted on 10 clinically healthy 10-17-day old Holsteinized black-motley calves in a farm in Novosibirsk district, Novosibirsk region. Blood for cytogenetic analysis was taken from each animal five times: before vaccination (control), 2 and 9 days after vaccination, 2 and 9 days after revaccination with the salmonellosis vaccine.

A concentrated formolvac vaccine against salmonellosis (paratyphoid) of calves was used in a dose of 2 ml (reimmunization in a dose of 2 ml) with an interval of 10 days between injections. The vaccine is made from bacterial culture of *Salmonella dublin* strain No. 373 inactivated with formalin (0.4% by volume) with the addition of potassium alum and calcium chloride (0.1% by volume) as an adjuvant.

The material for the cytogenetic study was the peripheral blood lymphocytes of animals stimulated by phytohemagglutinin. Preparations of metaphase chromosomes were prepared according to the method of P.S. Moorchead et al. [11] with modifications. The preparations were stained with Giemsa stain. The classification of somatic mutations was carried out according to the criteria proposed by N.P. Bochkov and A.N. Chebotarev [12]. As a result of a cytogenetic study of calves in different periods, 9472 metaphase plates were analyzed in detail. The frequencies of numerical and structural chromosome abnormalities were calculated per 100 cells. Frequencies of cytogenetic abnormalities are given in tables with percentage errors.

The research results were processed by standard methods of variation statistics using Microsoft Office Excel 2010. The significance of differences between the frequencies of cytogenetic indicators in the groups was evaluated by Fisher's method with φ-transformation of fre-

[2]Nazarenko Yu.S., Kulikova S.G., Loginov S.I. The influence of vaccinations on somatic chromosomal instability in animals and humans // The role of agricultural science in the sustainable development of rural areas: collection of articles. V All-Russian (national) scientific. conf. (Novosibirsk, December 18, 2020). Novosibirsk: RC NSAU "Zolotoy Kolos", 2020. P. 516–519.

[3]Kulikova S.G., Loginov S.I., Nazarenko Yu.S. The influence of vaccination against salmonellosis on the associative ability of acrocentric chromosomes in cattle // Theory and practice of modern agricultural science: collection of articles. IV national (All-Russian) scientific. conf. from international participation (Novosibirsk, February 26, 2021). Novosibirsk: RC NSAU " Zolotoy Kolos ", 2021. P. 908-912.

quencies. For the zero-frequency value the error was calculated by the method of B.L. Van der Waerden [13].

RESULTS AND DISCUSSION

Studies of peripheral blood lymphocyte cultures of clinically healthy Holsteinized black-motley calves found that the spectrum of cytogenetic abnormalities in animals after vaccination and revaccination against salmonellosis is represented by polyploidy, hypoploidy and hyperploidy cells, chromatid and chromosome breaks, single and paired chromosome fragments regardless of the time since the vaccine administration. Polyploid and aneuploid cells were detected among the numerical abnormalities in individuals both before and after vaccination and revaccination against salmonellosis (see Table 1.).

It was found that vaccination and revaccination of young cattle against salmonellosis did not lead to a significant increase in the frequency of genomic mutations in the somatic cells of animals. On the contrary, a tendency was found for a gradual decrease in the frequency of cells with an altered chromosome number 2 and 9 days after vaccination of calves against salmonellosis. 9 days after revaccination of animals, a significant decrease in the frequency of cells with genomic mutations to 7.18% was noted, control - 9.62% ($p < 0.05$).

The maximum frequency of cells with a changed number of chromosomes (14.0%) was found 2 days after revaccination of calves against salmonellosis, which exceeded the same indicator in animals 9 days after their vaccination and revaccination by 1.7 and 2.0 times, respectively ($p < 0.05$-0.01).

Among the cells with an altered chromosome number, aneuploid cells with hypoploid sets predominated (35.6-51.8%). The average frequency of hypoploid cells in calves before vaccination was 4.98%. The frequency of hypoploid cells was 1,9 times lower in the animals 9 days after revaccination against Salmonellosis

Табл. 1. Частота клеток с измененным числом хромосом у молодняка крупного рогатого скота до и после вакцинации, ревакцинации против сальмонеллеза, %

Table 1. Frequency of cells with changed number of chromosomes in young cattle before and after vaccination, and revaccination against salmonellosis, %

Indicator	Period of study				
	before vaccination (control)	after vaccination		after revaccination	
		after 2 days	after 9 days	after 2 days	after 9 days
Polyploid cells	3,40 ± 0,57	4,40 ± 0,75	3,16 ± 0,63	6,00 ± 1,68	2,52 ± 0,47
Aneuploid cells, including:	6,22 ± 1,10	4,47 ± 1,06	4,94 ± 1,10	8,00 ± 2,71	4,66 ± 0,91
hypoploid cells, including types:	4,98 ± 0,99	3,16 ± 0,90	3,38 ± 0,92	6,00 ± 2,37	2,61 ± 0,69*
2*n*-1	1,04 ± 0,46	2,11 ± 0,74	1,30 ± 0,58	1,00 ± 0,99	1,12 ± 0,45
2*n*-2	2,49 ± 0,71	0,00 ± 0,26***	1,30 ± 0,58	4,00 ± 1,96	0,56 ± 0,32**
2*n*-3 and more	1,45 ± 0,54	1,05 ± 0,52	0,78 ± 0,45	1,00 ± 0,99	0,93 ± 0,41
hyperploid cells, including types:	1,24 ± 0,51	1,32 ± 0,58	1,56 ± 0,63	2,00 ± 1,40	2,05 ± 0,61
2*n*+1	1,04 ± 0,46	0,53 ± 0,37	1,04 ± 0,52	2,00 ± 1,40	1,68 ± 0,55
2*n*+2	0,00 ± 0,21	0,53 ± 0,37*	0,52 ± 0,37*	0,00 ± 0,97	0,19 ± 0,19
2*n*+3 and more	0,21 ± 0,21	0,26 ± 0,26	0,00 ± 0,26	0,00 ± 0,97	0,19 ± 0,19
Cells with an altered number of chromosomes	9,62 ± 0,93	8,87 ± 1,04	8,10 ± 0,99	14,00 ± 2,45	7,18 ± 0,77*

Hereinafter *$p < 0,05$.
**$p < 0,01$.
***$p < 0,001$.

Significant differences are indicated in comparison with the period before vaccination (control).

compared with the control (p <0,05). There was no significant increase in the frequency of hyperploid cells in animals after their vaccination and revaccination after 2 and 9 days compared with the period before drug administration (see Table 1), but there was a clear tendency for a gradual increase in the frequency of this cell type after primary and repeated calf immunization (p > 0.05).

Different types of hypoploid and hyperploid cells were detected, the frequencies of which varied in different periods of study in young cattle (see Table 1). The frequency of cells with a loss of two chromosomes was found to decrease in 2 days after vaccination and in 9 days after revaccination of calves against Salmonellosis, to 0,0 and 0,56%, respectively, in the prevaccination period - 2,49% (p < 0,01-0,001). The highest frequency (4.0%) of cells with a deficiency of two chromosomes was observed 2 days after revaccination of animals, which was 7.14 times higher than 9 days after revaccination of calves against Salmonellosis (p < 0.05).

No significant differences were found in the frequency of cells with an excess of one or three or more chromosomes in calves before vaccination compared with animals vaccinated and revaccinated against Salmonellosis. Cells with an excess of two chromosomes (2n+2) were detected 2 and 9 days after vaccination of calves against Salmonellosis, with the frequency of 0.53 and 0.52%, respectively, while they were absent in controls (p < 0.05).

Another type of genomic mutation reported in the present studies is polyploidy (see Table 1). No significant differences were found in the frequency of polyploidy cells in calves at all periods after vaccination and revaccination against salmonellosis compared to controls (p > 0.05). At the same time, a 2.38-fold reduction in the frequency of polyploid cells was found in calves 9 days after revaccination, compared with animals examined 2 days after revaccination against Salmonellosis (p < 0.05).

Analysis of the polyploid cell spectrum (see Table 2) showed that tri- and tetraploid cells were detected in the animals without exception in all periods of the study (before vaccination, 2 and 9 days after vaccination and revaccination against salmonellosis). Cells with higher ploidy (5n and/or 6n) were recorded only in calves before vaccination and 2 days after their vaccination and revaccination against salmonellosis. The proportion of tetraploid cells was 75.7-95.9% of the total number of polyploid cells.

Along with genomic abnormalities in young cattle before vaccination, after vaccination and revaccination with inactivated vaccine against salmonellosis, chromosomal mutations were revealed. In the present studies, the spectrum of chromosomal aberrations is represented by single and paired fragments, chromatid and chromosomal breaks (see Table 3).

The frequencies of cells with aberrations and the total number of chromosome aberrations in the examined young calves had a tendency to

Табл. 2. Спектр и частота полиплоидных клеток у молодняка крупного рогатого скота до и после вакцинации, ревакцинации против сальмонеллеза, %

Table 2. Spectrum and frequency of polyploid cells in young cattle before and after vaccination, and revaccination against salmonellosis, %

Period of study	Cell ploidy			
	3*n*	4*n*	5*n*	6*n* and more
Before vaccination (control)	0,40 ± 0,20	2,80 ± 0,52	0,10 ± 0,10	0,10 ± 0,10
After vaccination:				
after 2 days	0,40 ± 0,23	3,33 ± 0,66	0,27 ± 0,19	0,40 ± 0,23
after 9 days	0,13 ± 0,13	3,03 ± 0,62	0,00 ± 0,13	0,00 ± 0,13
After revaccination:				
after 2 days	0,50 ± 0,50	5,00 ± 1,54	0,00 ± 0,50	0,50 ± 0,50
after 9 days	0,36 ± 0,18	2,16 ± 0,44	0,00 ± 0,09	0,00 ± 0,09

a gradual increase in 2, 9 days after vaccination and in 2 days after calf revaccination, in 9 days after revaccination the animals significantly exceeded the control in 2,9 and 2,5 times ($p < 0,01$). This increase was due to an increase in the frequency of cells with chromosome breaks and their total number in vaccinated and revaccinated calves against salmonellosis. Before vaccination, the frequency of cells with chromosome breaks was 1.04 ± 0.46%, but 9 days after vaccination it was 1.82 ± 0.68% ($p > 0.05$), and 9 days after revaccination it was 2.98 ± 0.73% ($p < 0.05$).

Chromatid and chromosomal aberrations were recorded in calf lymphocytes during the whole period of observation, both before vaccination and after vaccination and revaccination against Salmonellosis. There was no significant increase in the frequencies of each of these types of chromosome aberrations separately in the postvaccination and revaccination periods compared to the pre-vaccination period. On the whole, a significant increase in the frequency of these aberrations in 9 days after revaccination of animals against Salmonellosis in comparison with control was found. It should be noted that 2 days after calf vaccination against Salmonellosis no chromatid chromosome breaks were detected in comparison with the pre-vaccination period, where their incidence was 0.62 ± 0.36%.

Analysis of the chromosome breaks localization showed that they were recorded in large and medium chromosomes in the pericentromeric, medial and telomeric regions of the long arms of one or both chromatids. Chromosome breaks of the chromatid type are found mainly in the medial regions of one of the chromatids (almost 80% of all chromatid breaks). Breaks in telomeric regions were found in calves only before vaccination with a frequency of 0.41 ± 0.29%, in near-centromeric regions of chromatids - only 9 days after revaccination of young animals against salmonellosis with a frequency of 0.56 ± 0.32%. Breaks of the chromosomal type were most often localized in the medial (43% of cases) and telomeric (46%) regions of both chromatids, much less frequently (11%) in the pericentromeric regions. In the studied animals, both before and after vaccination and revaccination, cells with one and two chromosome breaks were recorded, and lymphocytes with one break prevailed, the proportion of which varied from 80 to 100%. No cells with

Табл. 3. Частота аберраций хромосом у молодняка крупного рогатого скота до и после вакцинации, ревакцинации против сальмонеллеза, %

Table 3. Frequency of chromosome aberrations in young cattle before and after vaccination, and revaccination against salmonellosis, %

Indicator	Period of study				
	before vaccination (control)	after vaccination		after revaccination	
		after 2 days	after 9 days	after 2 days	after 9 days
Cells with chromosome aberrations	1,66 ± 0,58	2,11 ± 0,74	2,86 ± 0,85	3,00 ± 1,71	4,84 ± 0,93**
Chromosome aberrations	2,07 ± 0,65	2,37 ± 0,78	2,86 ± 0,85	4,00 ± 1,96	5,21 ± 0,96**
Cells with fragments of chromosomes	0,83 ± 0,41	1,05 ± 0,52	1,04 ± 0,52	1,00 ± 0,99	1,86 ± 0,58
Fragments of chromosomes Including:	0,83 ± 0,41	1,05 ± 0,52	1,04 ± 0,52	1,00 ± 0,99	1,86 ± 0,58
single fragments	0,62 ± 0,36	0,53 ± 0,37	0,52 ± 0,37	0,00 ± 0,97	0,56 ± 0,32
paired fragments	0,21 ± 0,21	0,53 ± 0,37	0,52 ± 0,37	1,00 ± 0,99	1,30 ± 0,49*
Cells with chromosome breaks	1,04 ± 0,46	1,32 ± 0,58	1,82 ± 0,68	3,00 ± 1,71	2,98 ± 0,73*
Chromosome breaks Including:	1,24 ± 0,51	1,32 ± 0,58	1,82 ± 0,68	3,00 ± 1,71	3,36 ± 0,78*
chromatid breaks	0,62 ± 0,36	0,00 ± 0,26*	0,52 ± 0,37	2,00 ± 1,40	1,68 ± 0,55
chromosomal breaks	0,62 ± 0,36	1,32 ± 0,58	1,30 ± 0,58	1,00 ± 0,99	1,68 ± 0,55

three or more chromosome breaks were detected. There was a 3.1-fold higher frequency of cells with one chromosome break 9 days after calf revaccination against Salmonellosis, compared with the period before vaccination ($p < 0.05$).

There were no significant differences in the frequencies of single and paired chromosome fragments in calves in all periods of the study compared to controls (see Table 3). No differences were found after vaccination and revaccination when comparing these parameters between them, except for the frequency of cells with paired chromosome fragments, which was 6.2 times higher 9 days after revaccination than before vaccination ($p < 0,05$). It was found that cells with only one fragment were found in calves before vaccination and then at all periods after their vaccination and revaccination against salmonellosis.

As a result of the conducted studies of cattle, the mutagenic effect of routine vaccinations against salmonellosis of calves was assessed.

It was found that in peripheral blood lymphocytes of young cattle the spectrum of cytogenetic disorders induced by the inactivated vaccine against salmonellosis of calves is represented by polyploid, hyperploid and hypoploid cells, single and paired fragments of chromosomes and chromatid and chromosomal breaks.

It was revealed that the spectrum of somatic chromosomal instability after double immunizations with an inactivated vaccine against salmonellosis did not differ from the spectrum of spontaneously occurring mutations in this species [14–17].

To date, the mutagenic properties of many viral and bacterial vaccines have been studied in humans, in particular against measles, smallpox, poliomyelitis, influenza, rabies, mumps, yellow fever, herpes simplex, tick-borne encephalitis, brucellosis, typhoid fever and others [5–8, 18]. These studies showed that in most cases live vaccines had a pronounced mutagenic effect, while inactivated vaccines did not cause an increase in the frequency of cytogenetic disorders in the lymphocytes of vaccinated people. But according to J. Chun et al. [19], no increase in the number of cytogenetic disorders was found in 7 children vaccinated with a live measles vaccine.

The results of these studies indicate an ambiguous effect of vaccination and repeated revaccination after 10 days with an inactivated vaccine against salmonellosis on the frequency of genomic and structural mutations in clinically healthy Holsteinized black-and-white calves. There was no increase in the frequency of aneuploid and polyploid cells after vaccination and revaccination of calves with an inactivated salmonellosis vaccine in comparison with the pre-vaccination period. A tendency towards a gradual decrease in the frequency of cells with altered chromosome number in calves in all periods following vaccination against salmonellosis has been detected. A sharp increase in the frequency of these cells (both aneuploid and polyploid cells) was noted 2 days after revaccination, followed by a significant decrease in the frequency of cells with genomic mutations 9 days after revaccination in comparison with the period before vaccination. A significant change in the frequency of polyploid cells in different periods after revaccination of animals against salmonellosis was established. The significant variations in the frequencies of genomic mutations in young cattle revealed in studies after repeated immunization are consistent with the statement that with prolonged mutagenesis, which is possible during revaccinations, its intensity is characterized by a wave character with fluctuations in aberration levels from maximum to minimum [20].

The fact that the frequency of cells with an altered number of chromosomes in young cattle decreased significantly 9 days after revaccination against salmonellosis in comparison with intact animals, established in the study, may be caused by age features and/or different functional state of the immune system of the studied calves, responsible for the elimination of cells with cytogenetic abnormalities. An enhancement of the genome-protective system and other mechanisms cannot be excluded [18].

The results of these and earlier studies [15] and data from other scientists [17, 21] indicate that spontaneous and agent-induced mutagenesis of various nature in cattle most often revealed cells with a tetraploid set of chromosomes among polyploid cells.

At the same time, with regard to chromosome aberrations, it was found that immunization of calves with inactivated vaccine against Salmonellosis tended to gradually increase the frequency of chromosome structural disorders in 2, 9 days after vaccination and in 2 days after revaccination. This resulted in a significant increase in the frequency of cells with chromosome aberrations in animal blood lymphocytes 9 days after re-vaccination due to breaks and paired chromosome fragments. A similar result was obtained by N.N. Ilinskikh [5] during cytogenetic examination of healthy donors vaccinated against brucellosis. As early as 2 days after vaccine introduction, a significant increase in chromosome structural disorders was detected: the frequency of cells with chromosome breaks to 0.9 ± 0.2% vs. 0.1 ± 0.06% in the control and 2.3 ± 0.2% with chromatid breaks to 1.0 ± 0.01% in the control.

Studies by a number of scientists have shown that mutagenic factors are capable of specifically affecting individual chromosomes and their zones [5, 18]. When studying the mechanisms of the karyopathogenic effect of a brucellosis vaccine (strain 19 BA) in humans, large chromosomes of group A were affected more frequently, and disorders in small chromosomes of groups F and G were extremely rarely observed [5]. A similar pattern was detected in the present studies. The specificity of lesions of large and medium chromosome groups and their regions in young cattle after vaccination and revaccination against salmonellosis has been established. According to N.N. Ilinskikh [5], the biochemical structure of the infectious agent itself and its immunogenicity are of great importance in chromosome lesions. It was found that in calves after vaccination and re-vaccination against salmonellosis, chromatid breaks were most often registered in the medial regions of one of the chromatids, while chromosomal breaks were found in the medial and telomere regions of both chromatids. Apparently, the specificity of lesion of some chromosome regions by antigenic factors of inactivated vaccines as well as by some infectious agents is associated with peculiarities of structure and functioning of these chromosome regions. Some studies have shown that infectious agents, by altering cell metabolism, can initiate a chain of reactions that lead to visible damage to chromosomes [22].

Consequently, revaccination with inactivated salmonellosis vaccine induces cytogenetic abnormalities in reimmunized calves, causing a significant increase in the frequency of cells with chromosomal aberrations in peripheral blood lymphocytes of the animals. A cumulative effect of double vaccination with inactivated bacterial vaccine in cattle seems to be observed, resulting in an increased frequency of DNA damage due to the mutagenic properties of the vaccine antigens during the formation of the immune response.

CONCLUSION

As a result of studying the spectrum and frequency of cytogenetic disorders in somatic cells in clinically healthy Holsteinized black-and-white calves before vaccination, after vaccination and revaccination with an inactivated vaccine against salmonellosis, the mutagenic effect of routine immunizations against this disease in cattle was evaluated.

The spectrum of cytogenetic disorders induced by the inactivated vaccine against salmonellosis in young cattle in peripheral blood lymphocytes is represented by polyploid, hyperploid and hypoploid cells, single and paired chromosome fragments and chromatid and chromosomal breaks. The spectrum of cytogenetic disorders caused by double immunization with the vaccine against salmonellosis of calves did not differ from the spectrum of spontaneously occurring mutations in this species.

Different effects of vaccination and re-vaccination after 10 days with an inactivated vaccine against salmonellosis on the frequencies

of genomic and structural mutations in immunocompetent cells in young cattle were found. Vaccinations and subsequent revaccinations of calves with an inactivated vaccine against salmonellosis in comparison with the pre-vaccination period did not cause an increase in the frequencies of aneuploid and polyploid cells.

The intensity of prolonged mutagenesis during revaccination of calves against salmonellosis was characterized by a wave character with fluctuations in the frequencies of cells with genomic mutations from maximum to minimum values.

Revaccination with an inactivated salmonellosis vaccine has a mutagenic effect on the chromosomal apparatus of re-immunized calves, causing a significant increase in the frequency of cells with chromosomal aberrations in the peripheral blood lymphocytes of animals due to breaks and paired fragments of chromosomes.

The specificity of damage to certain regions of chromosomes in young cattle after vaccination and revaccination against salmonellosis was revealed. In calves after double immunization, chromatid breaks were most often recorded in the medial regions of one of the chromatids, and chromosomal breaks - in the medial and telomeric regions of both chromatids.

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ДИССОЦИИРОВАННЫЕ ФОРМЫ МОРАКСЕЛЛ, ВЫДЕЛЕННЫЕ ИЗ ПОРАЖЕННЫХ ГЛАЗ КРУПНОГО РОГАТОГО СКОТА

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Проведено изучение диссоциации эпизоотических культур моракселл. Исследования проведены в хозяйствующих субъектах Алматинской области Республики Казахстан на 233 гол. крупного рогатого скота с клиническими признаками кератоконъюнктивита. Изоляцию возбудителя моракселлеза осуществляли бактериологическими смывами из конъюнктивального мешка глаз животных. Лабораторные исследования проводили согласно утвержденным методическим указаниям. Установлено, что бактерии рода *Moraxella* диссоциируют при выращивании на твердой питательной среде в течение более 6 ч в условиях термостата при температуре 37 °С. Бактерии изучены способами: окрашивание по Уайт-Вилсону, термоагглютинация и проба с акрифлавином. При оценке выросших колоний по Уайт-Вилсону установлено для кристаллвиолета оптимальное разведение 1 : 2000, для краски генцианвиолет – 1 : 1000. В этом случае колонии в S-форме имеют темно-фиолетовый цвет с металлическим оттенком, а диссоциированные колонии в R-форме не окрашиваются. При наличии диссоциированных клеток отмечены преципитация (термоагглютинация), образование осадка и просветление надосадочной жидкости при 90 ºС в течение 30 мин. Взвесь не диссоциированных колоний при этом оставалась мутной. При взвешивании микробных клеток изолированных бактериальной петлей из отдельных выросших колоний в растворе акрифлавина, диссоциированные бактерии склеиваются, образуя конгломераты. При изучении антигенной активности S-, R-форм моракселл выявлено, что активность S-антигена значительно превышала таковую из R-форм. Данные о диссоциации культур моракселл могут быть использованы при разработке диагностических и профилактических препаратов при моракселлезе крупного рогатого скота.

Ключевые слова: *Moraxella*, референтные штаммы, диссоциация, эпизоотические культуры, S-R-колонии

DISSOCIATED FORMS OF MORAXELLA ISOLATED FROM THE AFFECTED EYES OF CATTLE

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The dissociation phenomenon of epizootic cultures of Moraxella was studied. The study was conducted in economic entities of Almaty region of the Republic of Kazakhstan for 233 heads of cattle with clinical signs of keratoconjunctivitis. Isolation of the causative agent of Moraxella was performed by bacteriological washes from the conjunctival sacs of the eyes of animals. The laboratory study was carried out according to the approved methodological guidelines. It was found

that bacteria of the genus *Moraxella* dissociate when grown on a solid nutrient medium for more than 6 hours in a thermostat at 37 °C. The bacteria were studied by the following methods: staining according to White-Wilson, thermoagglutination and acriflavine assay. When evaluating the grown colonies according to White-Wilson, the optimal dilution for crystal violet was found to be 1 : 2000, and for gentian violet stain 1 : 1000. In this case, the colonies in the S-form have a dark purple color with a metallic tint, and the dissociated colonies in the R-form do not stain. In the presence of dissociated cells, precipitation (thermoagglutination), sediment formation and clearing of the supernatant fluid at 90 °C for 30 minutes were noted. The suspension of undissociated colonies remained cloudy. When weighing microbial cells isolated by a bacterial loop from individual grown colonies in a solution of acriflavine, dissociated bacteria stick together to form conglomerates. When studying the antigenic activity of the S-, R- forms of Moraxella, it was revealed that the activity of the S-antigen significantly exceeded that of the R-forms. Data on the dissociation of Moraxella cultures can be used for the development of diagnostic and prophylactic drugs against moraxellosis in cattle.

Keywords: *Moraxella*, reference strains, dissociation, epizootic cultures, S-R- colonies

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INTRODUCTION

Many economic entities in the Republic of Kazakhstan have imported cattle from non-CIS countries to improve the genetic potential of their breeds. The import of beef breeding stock into the Republic of Kazakhstan with the causative agent of infectious keratoconjunctivitis (Pink-eye), the movement of infected animals has led to a significant spread and emergence of stationary nidus of this disease. Factors of various kinds that irritate the conjunctival mucosa, such as mechanical trauma, insects, dry and dusty particles, direct ultraviolet radiation from sunlight, etc., contribute to the aggravation of the disease.

Moraxelosis has not previously been registered among cattle in Kazakhstan. Monitoring of infectious keratoconjunctivitis moraxelosis aetiology in the territory of the Republic of Kazakhstan for 2016-2019 showed that the disease was detected in nine regions. Clinical examination of both imported and local livestock of different sex and age groups and different breeds (Aberdeen-Angus, Herefords, Holsteinfries, Kazakh Whitehead, Auliekol breeds and local non-bred animals) was conducted. Bacteriological examination of biomaterial taken from affected eyes and nasal cavity mucosa of animals and its subsequent identification (morphological, cultural, tinctorial, biological, serological

studies)[1,2] were carried out [1-8]. Data on the variability of Moraxella cultures in the specialist literature are currently scarce.

Morphological differences in the β-haemolysis zone of colonies of the reference strain *Moraxella bovis* Epp 63 have been described by some authors. Thus, colonies of the S (spreading) and C (corroding) forms were recorded to be 1-2 mm in diameter, smooth with well-defined edges, and formed corrosive agar. Type N (nonspreading and noncorroding) colonies were recorded with a slightly larger diameter (2-4 mm). These colonies did not have clearly delineated margins, did not corrode the agar, and had a granular texture (9).

Under a light microscope, the colonies present three characteristic concentric growth zones (peripheral, middle and central circular) (10).

The biological significance of dissociation lies in the acquisition by bacteria of certain selective advantages, which ensure their existence in their environment. Cases of greater resistance of S-forms of bacteria to phagocytosis by macrophages and to the bactericidal action of serum have been described.

R-form bacteria, unlike the S-form, are more resistant to the action of environmental factors, but they are less resistant to cellular immunity factors and persist longer in water and milk [11]. Dissociation usually proceeds in the S→R direction, sometimes through the formation of colonies of intermediate forms of bacteria, and is accompanied by changes in the biochemical, morphological, antigenic, and pathogenic properties of the microorganisms.

The reverse (reverse R to S) transition is observed much less frequently. Most pathogenic bacteria form S-colonies, except for the pathogens of tuberculosis, plague, anthrax and some others. Comparative electron microscopy of sections of genetically resistant R- and S-forms of Brucella showed that they have the same basic structural elements (cell wall, cytoplasmic membrane, cytoplasm, nucleoid). The coccoid forms of dissociated R cells of brucells, more pronounced than those of the S-forms of brucells, and C-shaped envelope invaginations - with a bumpy-folded relief - were recorded (bacterial cells of bacilliform shape with a smooth-grained structural surface were detected in the S-form) [12].

The aim of the research is to study the variability of epizootic cultures of Moraxella isolated from diseased eyes of cattle in the territory of the Republic of Kazakhstan.

MATERIAL AND METHODS

Studies were conducted in economic entities "Arkharly Maibuyrek", "Baiserke Agro" and "Farmagro" of Almaty region (southern region of Kazakhstan). After examination of 1,965 head of cattle from May to September 2019, 233 head with clinical signs of keratoconjunctivitis were selected. Isolation of the causative agent of moraxellosis was carried out by bacteriological washes from the conjunctival sac of the eyes. Biomaterial was taken with sterile wands with a plastic handle from a transport tube with individually packaged Amies medium (made in Italy). With rotating movements of the sterile applicator, existing oozes were removed from the affected eye. Obtained samples of clinical pathological material were transported in a thermo-compartment with ice to the bacteriology laboratory within 3-4 hours. Laboratory tests were performed according to approved guidelines[3].

Smears were prepared from each specimen of pathological material, Gram stained and examined under an immersion microscope, noting the presence or absence of morphologically

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similar organisms to *Moraxella bovis*. The material was then inoculated on blood (5% defibrinated ram's blood) Hottinger's agar. The results of the inoculations were recorded after 12-24 h of incubation at 37 o C, transferring typical Moraxella β-haemolysis zones to fresh nutrient media for isolation of pure cultures. Two epizootic cultures of *Moraxella bovis* isolated from sick animals, reference strains Moraxella bovis ATSS 17948TM and *Moraxella bovoculi* BAA 1259TM, obtained from "LGC Standards Sp.z. o.o. (made in Poland). Acryflavin assay, thermoagglutination reaction and White-Wilson colony staining were used to determine dissociation.

The immunological activity of S- and R-form Moraxella antigens was studied by complement binding reaction and long-term complement binding reaction (CFT/CLFT) with homologous sera, which were obtained by immunizing rabbits [7, 9]. Acryflavin solutions were prepared in the ratio of 1: 500, 1: 1000, 1: 1500, 1: 2000, 1: 3000, 1: 5000 in distilled water. A drop of acryflavin solution was applied to a degreased glass and a bacteriological loop of Moraxella culture was thoroughly stirred in it. During the first 4 min in the case of dissociation a granularity appears in the form of conglomerates of glued moraxellae.

For the thermoagglutination reaction, a bacterial suspension of Moraxella in physiological solution equivalent to the McFarland turbidity standard 4.0 was prepared from a daily agar culture, poured into 8.0 cm3 tubes and heated in a water bath at 90 °C for 30 min. The reaction is recorded after 1 and 24 h after heating.

For White-Wilson staining, a suspension of Moraxellae in sterile physiological solution was prepared from a daily agar culture so that a sufficient number (100-150) of isolated colonies would grow in Petri dishes when sown on agar. For this purpose, firstly, a suspension of Moraxella was prepared with a concentration of 1 billion microbial cells in 1.0 cm^3. Then, using a tenfold dilution method, the composition was adjusted to a concentration of 100-1000 CFU by adding 0.5 cm^3 of Moraxella suspension to 4.5 cm^3 of physiological solution in each successive test tube at a concentration of 10-6 and 10-7. From the last dilution of the suspension (-10-6, -10-7) containing 100-1000 microbial cells in 1.0 cm^3, 0.1 cm^3, three Petri dishes for each variant were seeded on nutrient medium.

RESULTS AND DISCUSSION

When stained with aniline dye colonies of Moraxella cultures were examined under a MEIJI TECHNO light microscope (made in Japan) with a digital camera, S-shaped colonies (see Figure 1, a) with three zones of colony growth (see Figure 1, b) and colony periphery with a spreading corrosive agar morphology (see Figure 1, c) were recorded.

S-form colonies on solid media are convex with well-defined edges, smooth 1-2 mm in diameter (× 10) (see Figure 1). At × 40 the colonies possessed three characteristic concentric growth zones. At the periphery was a narrow annular zone (peripheral ring) that surrounded another, wider annular zone (middle ring). The latter surrounded the central ring zone. From the outer ring zone, bacteria formed superficial colonies with a spreading corrosive agar morphology (see Figure 1, c).

The results obtained for the dissociation of the cultures of epizootic and reference strains of *Moraxella bovis* and *Moraxella bovoculi* by the acryflavin assay are shown in Table 1.

According to the results of the Moraxella dissociation test with acryflavin, the 6-hour bacterial cell cultures do not give agglutination, i.e. the result is negative, the microbial suspension is homogenously turbid (see Table 1).

After 12 h of bacterial growth in this experiment, slightly different results were obtained. Placing the grown culture into a solution of acriflavin causes partial agglutination of the bacterial cells and the formation of grains (see Table 1). In the experiment, 24-hour cultures when mixed with acriflavin were completely agglutinated and large grains of agglutinate in the clear surrounding liquid were observed (see Figure 2).

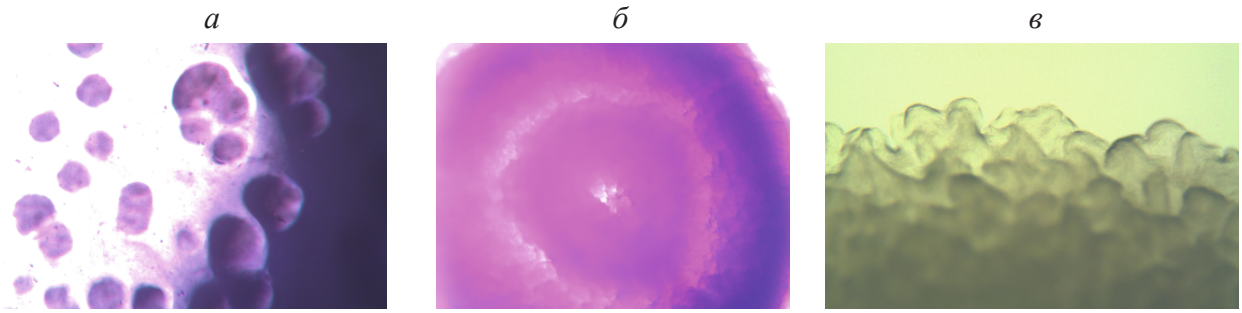

Рис. 1. Колонии S-формы культур мораксел под световым микроскопом:
а – колонии S-формы при × 10, *б* – зоны колонии при × 40, *в* – внешний край колонии

Fig. 1. S-form colonies of Moraxella cultures under a light microscope:
а – colonies of S-form × 10 magnification, *б* – colony zones at × 40 magnification, *в* – outer edge of the colony

The optimum dilution of acriflavine for determining moraxella dissociation varies from 1: 500 to 1: 2000. In this case, 5-6-hour S-form cultures in an acriflavine solution remain homogeneous, while 18-24-hour and daily cultures form a conglomerate with lucidity of the liquid.

The suspension of a 6-hour culture after thermal agglutination remained cloudy, no precipitation was observed after 1 and 24 hours. During thermoagglutination of a daily culture, precipitation to the bottom of the test tube and clarification of the liquid were recorded.

Thus, another manifestation of dissociation of Moraxella cultures is the positive thermoagglutination reaction, which is pronounced in R-forms of Moraxella cultures colonies.

When colonies were stained by White-Wilson in Petri dishes after 5-6 h, colonies belonging to the smooth (S) type grew. They had a convex correctly outlined smooth shape. The diameter of the colonies ranged from 0.3-0.5 to 0.8-1.0 mm. When stained with crystal violet or gentian violet at dilutions of 1: 500 to 1: 4000, colonies had the following appearance, light violet to dark blue, convex, smooth and with well-defined margins. After 18, 24 and 48 hours, the colonies became rugose and wrinkled, and remained white or pale yellowish in colour when stained. Colonies of R-form Moraxella remained unchanged, i.e. did not stain, which distinguishes them fundamentally from some microorganisms (Brucella, Salmonella, etc.).

After incubation at 37 °C for 18-20 h, a working solution of crystal violet at a dilution of 1: 500 to 1: 4000 was poured into agar plates where about 100-150 colonies had grown. After 60 s the dye was removed and the colonies were viewed with a magnifying glass (see Table 2).

Crystall violet and gentian violet staining showed no fundamental differences (see Table 2). The optimal dilution of crystal violet dye, where dissociation was clearly fixed, was found to be 1: 2000, for gentian violet dye it was 1: 1000. Colonies in the S-form were stained dark purple with a metallic hue, while dissociated colonies in the R-form did not change and retained a light yellow or white colour, becoming differently striated and wrinkled (see Fig. 3, a, b, c).

According to observations, 5-6 h moraxella colonies had a smooth shape (see Figure 3, a) and were stained with White-Wilson aniline dye. After 24-48 h, the colonies had a rough surface starting from the centre and were not stained (see Fig. 3, b, c).

The dissociation of colonies of cultures of epizootic and reference *Moraxella bovis* and *Moraxella bovoculi* strains was studied using conventional methods: exposure to temperature and, consequently, thermoprecipitation or thermoagglutination, acryflavin assay and White-Wilson staining of colonies with gentian violet.

The antigenic activity of S-, R-forms of Moraxella was studied by CFT/CLFT with antigens [5] prepared from the specified bacte-

Табл. 1. Результаты постановки проб на диссоциацию мораксел с акрифлавином
Table 1. Test results for dissociation of Moraxella with acriflavine

Dilution ratio of acriflavine with distilled water	Growth period of moraxella on nutrient media, h	The presence of agglutination				
		Epizootic culture		Reference strain		Control
		Moraxella bovis 2017-44	Fa16	*Moraxella bovis* ATCC 17948™	*Moraxella bovoculi* BAA 1259™	Suspension of moraxell in 0,85% NaCl solution
1 : 500	6	–	–	–	–	–
	12	+	++	++	++	–
	24	#	#	#	#	–
	48	#	#	#	#	–
1 : 1000	6	–	–	–	–	–
	12	++	++	+	++	–
	24	#	#	#	#	–
	48	#	#	#	#	–
1 : 1500	6	–	–	–	–	–
	12	+	++	++	++	–
	24	#	#	#	#	–
	48	#	#	#	#	–
1 : 2000	6	–	–	–	–	–
	12	+	+	+	+	–
	24	#	#	#	#	–
	48	#	#	#	#	–
1 : 3000	6	–	–	–	–	–
	12	+	+	+	+	–
	24	+	+	+	+	–
	48	+	+	+	+	–
1 : 5000	6	–	–	–	–	–
	12	–	–	–	–	–
	24	–	–	–	–	–
	48	–	–	–	–	–

Note. + - the severity of the formation of granularity (agglutination).

а *б*

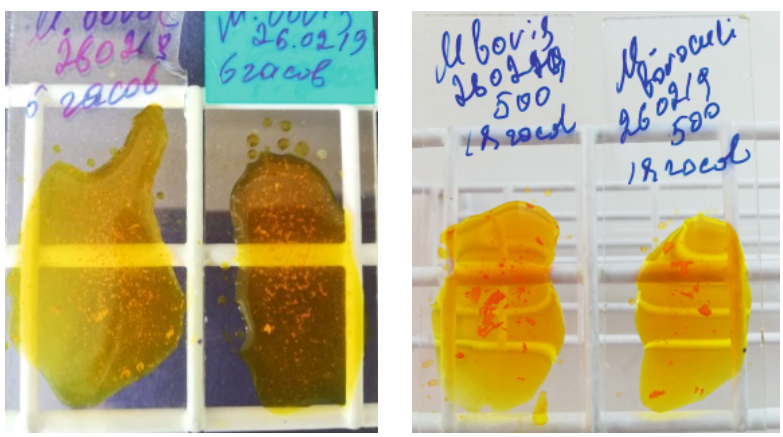

Рис. 2. Агглютинация культур мораксел акрифлавином:
а – 6-часовые культуры мораксел, *б* – 18-часовые культуры мораксел

Fig. 2. Agglutination of Moraxella cultures with acriflavine:
а – 6-hour Moraxella cultures, *б* – 18-hour Moraxella cultures

rial species. Experiments were performed with positive and negative sera. Positive sera were obtained by immunizing rabbits with a suspension of different forms of Moraxella [5, 7]. The results are shown in Table 3.

The antigen from the S-form of the moraxella does not react with R-serum in CFT, in CLFT its titer was shown to be 1: 10 (see Table 3). The R-antigen does not capture complement-binding to moraxella in the S-form in RGC, in the long-term complement binding reaction the titer of the R-antigen was recorded as 1: 10. The activity of the S-antigen is significantly higher than that of the R-form of the moraxella.

Thus, the question arises as to whether there is a possible reversion of R cells to the S-form.

Табл. 2. Результаты окрашивания колонии культур по Уайт-Вилсону
Table 2. Results of colony staining according to White-Wilson

Stain dilution ratios	Duration of culture growth, h	Staining according to White-Wilson							
		Crystal violet				Gentian violet			
		Epizootic culture		Reference strain		Epizootic culture		Reference strain	
		Moraxella bovis 2017-44	Fa16	*Moraxella bovis* ATCC 17948™	*Moraxella bovoculi* \ BAA 1259™	*Moraxella bovis* 2017-44	Fa16	*Moraxellella bovis* ATCC 17948™	*Moraxella bovoculi* BAA 1259™
1 : 500	6	+	+	+	+	+	+	+	+
	12	–	–	–	–	–	–	–	–
	24	–	–	–	–	–	–	–	–
	48	–	–	–	–	–	–	–	–
1 : 1000	6	++	++	++	++	++	++	++	++
	12	–	–	–	–	–	–	–	–
	24	–	–	–	–	–	–	–	–
	48	–	–	–	–	–	–	–	–
1 : 2000	6	+++	+++	+++	+++	+++	+++	+++	+++
	12	–	–	–	–	–	–	–	–
	24	–	–	–	–	–	–	–	–
	48	–	–	–	–	–	–	–	–
1 : 4000	6	#	#	#	#	#	#	#	#
	12	–	–	–	–	–	–	–	–
	24	–	–	–	–	–	–	–	–
	48	–	–	–	–	–	–	–	–

Note. Colony staining with crystal violet solution: dash - no colony staining; + - colonies are colored slightly pale blue; ++ - colonies become pale blue; +++ - colonies turn purple; # - colonies turn dark purple.
Colony staining with gentian violet solution: dash - no colony staining; + - colonies are colored slightly pale blue; ++ - colonies become pale blue; +++ - colonies are colored blue; # - colonies turn dark purple.

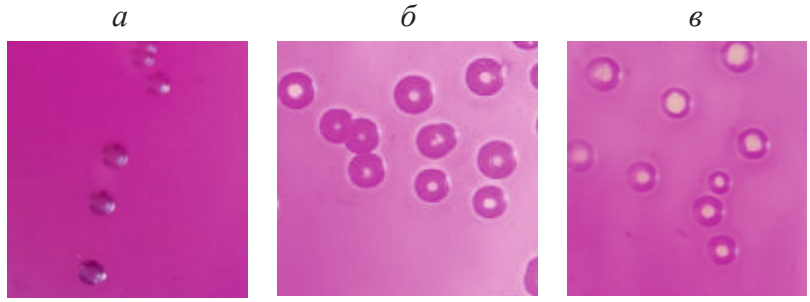

Рис. 3. Диссоциация колонии моракселл:
а – 6-часовые колонии моракселл в S-форме, *б* – 12 часовые колонии в S-, R- форме, *в* – 24-часовые колонии моракселл

Fig. 3. Dissociation of the moraxella colony:
а – 6-hour Moraxella colonies of S-form, *б* – 12-hour colonies of S-, R- form, *в* – 24-hour Moraxella colonies

This needs to be taken into account when making diagnostic and protective antigens from the S-R forms of the moraxellosis pathogen and requires further investigation.

CONCLUSION

As a result of studying the changes in cultures of bacteria of genus Moraxella after more than 6 hours of cultivation on solid nutrient medium, it was found that dissociation of microorganisms can be detected by staining of grown colonies with gentian violet or crystal violet by White-Wilson method, heating bacterial suspension in test tube at 90 o C for 30 min. In cases of dissociated cells, precipitate formation and lucidity of the supernatant were observed.

The presence of dissociated forms of bacteria is also detected by weighing microbial cells isolated by bacterial loop from individual colonies grown in acryflavin solution. The dissociated bacteria stick together to form conglomerates that are clearly detectable visually.

Data on the dissociation of Moraxella cultures can be taken into account in the development of diagnostic and prophylactic preparations for bovine moraxellosis.

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Табл. 3. Результаты РСК/РДСК с S-, R-противомракселлезными гипериммунными сыворотками

Table 3. Results of CFT/CLFT with S-, R-anti-moraxellosis hyperimmune sera

Immunological test	Serum	Antigen titer from	
		S-form	R-form
CFT	S	40	–
	R	–	20
	–	–	–
CLFT	S	80	10
	R	10	40
	–	–	–

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