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## Miscanthus sacchariflorus IN SIBERIA – BIOLOGICAL YIELD PARAMETERS AND DYNAMICS OF BIOFILIC ELEMENTS

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## Abstract

Throughout the world, bioenergy crops are grown to replace fossil resources and reduce greenhouse gas emission. Miscanthus is one of the bioenergetic plants that is widely cultivated in countries with optimal hydrothermal conditions. There is little research on the specifics of Miscanthus spp. cultivation in the continental climate. The aim of the study was a preliminary assessment of the possibility of Miscanthus cultivation in Siberia. The production process of Miscanthus sacchariflorus cv. Soranovsky (The State Register of Breeding Achievements, patent No. 6931 dated 06/06/2013) was assessed under conditions of the Central Forest-Steppe of Novosibirsk Ob (Novosibirsk Province, 54°53'13,5"N, 82°59'36,7"E, agro-gray soil, the experimental base of the Siberian Research Institute of Plant Cultivation and Breeding). During the formation of Miscanthus long-term stands (1-4 years), the aboveground biomass averaged 13 t/ha, the belowground biomass 17 t/ha. These figures are close to the average for this species in the world. The total N, P and K removal with biomass of stems and leaves was 51, 6, and 49 kg/ha, and with stems it was only 23, 3, and 26 kg/ha, respectively. Accumulation of N, P and K in the belowground biomass was 130, 10, and 126 kg/ha, respectively. Therefore, our experiment did not show any depletion of biophilic elements in soil during a four-year growth of miscanthus. Rough estimates have shown the presence of objective prerequisites for the atmospheric carbon sequestration in the fractions of the soil organic matter. During the first year of Miscanthus vegetation, at least 300 kg/ha of carbon accumulated in the mobile fractions of the soil organic matter; an increase in the C/N ratio in the belowground biomass of *Miscanthus* up to 74 vs. 20 in the initial soil was accompanied by a significant decrease in the mineralization rate of newly incoming plant residues. The parameters of the production process of Miscanthus sacchariflorus cv. Soranovsky on agro-gray soil in the Central Forest-Steppe of Novosibirsk Ob region correspond to the growth characteristics of *M. sacchariflorus* cultivated in other regions of the world on different soil types. We conclude that cultivation of *M. sacchariflorus* in Siberia is ecologically and commercially reasonable.

Keywords: bioenergy crops, Miscanthus sacchariflorus, cv. Soranovsky, aboveground biomass, belowground biomass, soil, nutrient removal, carbon sequestration

The production of renewable sources of fuels and raw materials is becoming one of the conditions for the survival of humankind. The cultivation of bioenergetic plants is recognized as one of the most important ways to solve this problem. Among such plants, representatives of the genus *Miscanthus* spp. of the family Poaceae are considered to be among the most promising in the world. This is due to both the valuable chemical properties of biomass and the characteristic high

growth rates and colossal biological productivity of this plant, as well as the conditions of a moderate climate. Miscanthus biomass is used as a renewable source of fuel for the production of lignocellulosic biomass, construction and composite materials (substitutes for wood and plastics) [1, 2]. Chlorophyll is also obtained and paper is produced from biomass [3].

The genus *Miscanthus* includes more than 20 species, distributed from tropical and South Africa to East and Southeast Asia. In Russia, in the Far East, there are 3 species: sugar-flowered miscanthus (*Miscanthus sacchariflorus*), reddening miscanthus (*M. purpurascens*), and Chinese miscanthus (*M. sinensis*) [4].

In the world (in Norway, Germany, Denmark, Great Britain, USA, Canada, and other regions), a fairly large experience has been accumulated in cultivating energy crops, including several species of the genus *Miscanthus*. At present, the main areas of their planting are located mainly in countries with an average annual positive air temperature above  $5-10 \,^{\circ}\text{C}$  and total annual precipitation of at least 600 mm (North America, Europe). The most widely cultivated species is *M. giganteus*, which stands out for its particularly high productivity in the above-mentioned climatic zones. Low frost resistance of this species [5-7] reduces its prospects for growing in the conditions of the Russian Federation.

In Siberia, miscanthus is quite famous as an ornamental plant. The problem of growing crops for biomass production in Siberia remains practically unexplored. It became necessary to select local varieties of miscanthus capable of maintaining high productivity under the limiting factors of the continental climate. Based on a collection of wild miscanthus species selected in the Far East of Russia, the cultivar Soranovsky was bred [8], which, by phenotyping and DNA analysis, was assigned to the species *M. sacchariflorus* [9]. The plants of this species are long-rhizome, have a height of up to 2.0-2.5 m, the stem is straight, rigid, with a pubescent leafy sheath around it. The leaves are narrow, linear-lanceolate, up to 60 cm long. The abaxial surface of the leaf plate is glabrous and non-pubescent, the adaxial surface is pubescent with a whitish, noticeable middle and solid edge. The inflorescence is in the form of a fan-shaped panicle of pale purple color at the beginning of flowering and white-gray at its completion, up to 25 cm long. The axis of the inflorescence is short, with long racemes.

*M. sacchariflorus* of the cultivar Soranovsky, in comparison with *M. giganteus*, is characterized by a higher frost and drought resistance, which makes it possible to recognize its potential for cultivation in the conditions of Siberia [10]. A distinctive feature of the variety is the formation of very long rhizomes that quickly colonize the free space. As a result, in a short period of time, a continuous flat plantation is formed, which successfully competes with weed vegetation. As it is known, the plantation of *M. giganteus*, due to its short rhizomes, slowly colonizes the space and represents overgrown hummocks with empty areas occupied by weed vegetation [10].

A large-scale experiment aimed at comparing the productivity of 15 genotypes of *M. giganteus*, *M. sinensis* and *M. sacchariflorus* in different climatic zones (Great Britain, Germany, the Netherlands, Ukraine, Turkey, Russia — Moscow Province) showed that *M. sacchariflorus*, in comparison with *M. giganteus* and *M. sinensis*, had more pronounced dependence of the biomass value on climatic conditions [11]. In this regard, the substantiation of the possibility of cultivating *M. sacchariflorus* in continental conditions requires special studies.

In addition, to regulate the production process of miscanthus, in particular, to answer the question regarding the doses and types of fertilizers, the

data on the turnover of biophilic elements in the soil-plant system are required. As it is known, the relatively low alienation of biophilic elements with the yield of miscanthus is due to the fact that when the biomass dries up, a significant part of the elements returns to the belowground part of plants (reutilization). As it is noted [12-14], the amount of reutilization depends on climatic conditions, the type of miscanthus, as well as the timing of harvesting.

The value of energetic plants is not limited to producing a renewable source of fuel and raw materials. In recent years, the interest in these crops, especially miscanthus, has been increasing due to the problem of binding the greenhouse gas  $CO_2$ . It is assumed that the colossal productivity of miscanthus biomass will contribute to the sequestration of atmospheric  $CO_2$  in stable fractions of soil organic matter (SOM) [15-19].

In this work, the authors have shown for the first time that a new culture for Russia *Miscanthus sacchariflorus* of the cultivar Soranovsky can be grown in the conditions of the arid and cold climate of Western Siberia. Miscanthus with an average yield of  $10\pm1.5$  t/ha of absolutely dry matter for the first four years of plantation formation did not deplete the soil with elements of mineral nutrition of plants, contributed to the atmospheric carbon sequestration in the fractions of SOM ( $1\pm0.15$  t C/ha per year in mortmass,  $150\pm30$  kg C/ha per year in mobile fractions).

The aim of the study was to assess the possibility of growing *M. sacchariflorus* of the cultivar Soranovsky in the continental conditions of Siberia based on the analysis of plant productivity, dynamics of carbon in the agroecosystem, and determination of the need for mineral fertilizers.

*Materials and methods.* The experiments were conducted in the conditions of the Central Forest-Steppe of Novosibirsk Ob (Novosibirsk region,  $54^{\circ}53'13.5''N$ ,  $82^{\circ}59'36.7''E$ ). A miscanthus plantation with an area of 0.3 hectares was laid in the spring of 2015 at the scientific and experimental base of the Siberian Research Institute of Plant Cultivation and Breeding – a branch of the Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences. The rhizomes of *M. sacchariflorus* cv. Soranovsky (originator – Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences) were used as planting material [9]. The observations continued during 2015-2018.

According to the data by AMS Ogurtsovo, during the growing seasons in 2015, 2017, and 2018, the precipitations were 322, 316, and 297 mm, respectively, the sum of temperatures above 0 °C was 2396, 2340, and 2199 °C, in 2016 231 mm and 2478 °C with vs. long-term values of 263 mm and 2248 °C. Consequently, 2015, 2017, and 2018 were characterized as moderately humid, and 2016 was relatively dry; in all years the hydrothermal coefficient was the same (HTC = 1).

Before the establishment of the miscanthus plantation, a soil-agrochemical survey of the experimental site was performed. The soil was agro-gray typical. The depth of the humus horizon was 30-35 cm, the content of  $C_{org.}$  was 1.57-1.76% (according to Tyurin), N<sub>tot</sub>. 0.13% (according to Kjeldahl), P<sub>2</sub>O<sub>5</sub> 30-40 mg/100 g (according to Chirikov), K<sub>2</sub>O in the extract 10-13 mg/100 g (according to Chirikov), hydrolytic acidity 0.5-1.5 mg-eq/100 g of soil (according to Kappen in the modification of CINAO), and the content of exchangeable Ca and Mg was about 70% of the cation exchange capacity. The granulometric composition was medium loamy.

To study the accumulation of plant biomass, plots of  $50 \times 50$  cm were used. The aboveground plant biomass (AB) was sampled in four-fold repetition annually during the growing season according to the phases of miscanthus development. The biomass was counted separately for plant organs – stems, leaves, and inflorescences (panicles). The belowground biomass (BB) was taken into account by the method of monoliths (height 25 cm, cross-sectional area  $10 \times 10$  cm<sup>2</sup>) annually in 5-fold repetition at the onset of the phase of decline of AB. Mortmass and rhizomes were separated by decantation of the soil with water on a sieve with a mesh diameter of 0.25 mm. The biomass was dried to an absolutely dry state. The NPK content in the aboveground plant biomass was determined annually in the phases of germination, flowering, and decline. After the desiccation of the biomass to an absolutely dry state, its wet ashing was conducted according to the Ginzburg method, the total nitrogen content was determined according to Kjeldahl, phosphorus according to Truog, potassium photometrically (atomic absorption spectrometer with flame atomization AAS Kvant 2A MT, OOO KORTEK, Russia) [20]. The analytical replication was 3-fold.

Based on the actual content of elements in the biomass and the total value of the latter, the authors calculated the removal of nitrogen, phosphorus, and potassium during the period of maximum development of the AB by the end of August—early September (flowering phase) and when it dried up by the end of September—early October. The difference between these values was used to assess the amounts of elements returned to the soil with dry plant residues.

To determine the content of NPK and organic carbon in BB on plots of  $50 \times 50$  cm, the rhizomes were taken in 5-fold biological repetition and each sample was analyzed in 2-fold repetition. The NPK content in the rhizomes was measured in the autumn of 2016. The analysis procedure was the same as for determining NPK in AB. The accumulation of elements in the biomass and their removal with the harvest were calculated from the value of the biomass and the accumulation of the corresponding element in it. The content of organic carbon (Corg.) and total nitrogen (Ntot.) in BB was determined on an elemental CHNS analyzer Vario EL III (Elementar Analysensysteme GmbH, Germany) according to the attached manufacturer's instructions.

Soil sampling to assess the NPK content in 0-20 and 20-40 cm layers was performed in 3-fold repetition on the  $50 \times 50$  cm plots annually at the beginning (germination phase) and at the end (senescence phase) of miscanthus vegetation. The analytical repetition was 3-fold. The agrochemical analysis of the soil was performed by the standard methods: phosphorus was determined in two extracts according to Chirikov and to Karpinsky-Zamyatina (GOST 26204-91, Moscow, 1992; DSTU 4727: 2007, http://www.chemicalnow.ru/chemie-6387.html), nitrate nitrogen ionometrically (GOST 26951-86, Moscow, 1986), potassium in Maslova's extract (AAS Kvant 2A MT, OOO KORTEK, Russia) (GOST 26210-91, M., 1992), the content of organic carbon ( $C_{org.}$ ) according to Tyurin [21].

The indicators characterizing the carbon metabolism in the soil were assessed by the absorption method according to the production of C-CO<sub>2</sub> under laboratory conditions [22]. A sample of soil (200 g of air-dry mass) was placed in plastic vessels with a volume of 1 L with a sealed lid. The incubation was conducted at 25 °C and humidity 60% of the field water capacity. The emitted CO<sub>2</sub> was captured with 0.1 n. NaOH. The remainder of the alkali was determined by titration using a 916 Ti-Touch titrator (Metrohm AG, Switzerland).

To determine the amount of carbon in the mobile fraction of SOM (laboratory experiment 1) after the first 2 years of vegetation, the authors simplified a bit the known technique [23]. The experiment involved soil samples from under miscanthus and from under pure fallow. The amount of carbon in the latter was taken as the initial one, since a miscanthus plantation was made on this soil. The experiment was repeated 3 times. The incubation was conducted until the significant differences in the rate of soil respiration between the variants of the experiment disappeared, which was considered to indicate the complete decomposition of the stock of mobile SOM accumulated in the soil under the miscanthus. Further, the value of mobile SOM was calculated from the difference in the total release of C-CO<sub>2</sub> from the soil from under the miscanthus and fallow.

To study the direction of carbon metabolism in the soil under miscanthus (laboratory experiment 2), ground wheat straw was added to soil samples (from under fallow and miscanthus) at a dose of 600 mg C/kg of soil. The response of the respiratory activity of the soil was assessed as an increase in the production of C-CO<sub>2</sub> in the variant with the introduction of wheat straw (%) compared to the control (soil samples in which no straw was added). The experiment was repeated 3 times.

The experimental data were statistically processed using the MS Excel program. The results are presented as means (M) with standard errors of means ( $\pm$ SEM) or confidence intervals at a significance level of p = 0.05 ( $t_{0.05} \times$  SEM). The reliability of the differences was assessed by the results of calculating the paired Student's *t*-test. With the actual value of the criterion below the level accepted in the experiment (0.05), the differences between the options were considered significant.

*Results.* As a result of the research, it was required to answer the following questions: how does the productivity of *M. sacchariflorus* of the cultivar Soranovsky compare with the development parameters of this and other miscanthus species in different regions of the world; is the soil depleted in terms of the main biophilic elements during the cultivation of miscanthus and is there a need for fertilization; what is the direction of the dynamics of carbon in the agroecosystem of *M. sacchariflorus* of the cultivar Soranovsky in the first years of plantation formation.

The phases of miscanthus development, in which the plant biomass was taken into account, are presented in Table 1.

1. Dates of the onset of phenological phases in *Miscanthus sacchariflorus* plants of cv. Soranovsky by the years of observation (Novosibirsk Province, 54°53'13,5"N, 82°59'36,7"E)

Phase	2015	2016	2017	2018
Planting	05/14	-	_	-
Germination	06/08	05/24	05/24	05/29
Internode growth	06/21	06/02	06/06	06/07
Belowground tillering by means of rhizomes	07/14	_	_	_
Stem elongation	08/25	07/14	07/15	07/17
Flowering	09/18	08/23	08/31	09/04
Decline	10/14	09/30	10/06	10/04
Duration of the vegetation period, days	129	130	136	129
N o t e. Dashes in the table indicate the absence	of data.			

**2.** Biomass dynamics (absolutely dry matter, t/ha) in *Miscanthus sacchariflorus* plants of cv. Soranovsky during the formation of the plantation (n = 12,  $M \pm SEM$ ; Novosibirsk Province, 54°53′13,5″N, 82°59′36,7″E)

	Aboveground								
Year (plan-	seedlings	flowering				d	Belowground		
ting age)	leaves and	leaves	stem	panicle	leaves	stem	panicle	total	Delowground
	stem			F			F		
2015 (1 year)	$0.009 \pm 0.001$	$0.30 \pm 0.04$	010 - 011				$0.05 \pm 0.01$	$0.8 \pm 0.1$	$4.5 \pm 0.7$
2016 (2 years)	$1.0 \pm 0.1$	$6.30 \pm 2.00$	7.1±2.2	$0.40 \pm 0.10$	$6.0\pm0.9$	$6.3 \pm 0.7$	$0.30 \pm 0.10$	12.6±1.5	9.6±1.8
2017 (3 years)	$2.7 \pm 0.7$	$7.80 \pm 1.00$	$8.1 \pm 0.8$	$0.10 \pm 0.09$	7.4±1.2	8.1±1.2	$0.30 {\pm} 0.07$	$15.9 \pm 0.6$	$13.7 \pm 0.8$
2018 (4 years)	2.3±0.6	$5.60{\pm}1.30$	$6.6 \pm 1.5$	$0.05{\pm}0.02$	$4.5 \pm 1.2$	7.4±1.5	$0.20{\pm}0.04$	$12.1 \pm 1.2$	$17.0 \pm 1.1$

Table 2 shows the dynamics of accumulating the AB and BB of miscanthus over the years of the observation.

In the year of planting (2015), miscanthus formed a well-developed bush.

The stock of BB in the first year of the growing season exceeded the stock of AB by more than 5 times. In the second year, the stock of AB exceeded the indicator of the first year by almost 16 times; the stock of BB increased by about 2 times and did not reliably differ from the stock of AB. In the third and fourth years of the growing season, the value of AB stabilized, while BB continued to increase. For 4 years, the yield of AB averaged 10 t/ha, BB 11 t/ha.

Under the conditions of the authors' experiment, during the vegetation period, the content of nitrogen and potassium in the biomass changed the most significantly (the indicator decreased from seedlings to desiccation by 9 and 6 times, respectively) (Table 3).

**3.** Content of biophilic elements in plant biomass of *Miscanthus sacchariflorus* cv. Soranovsky (*n* = 12, *M*±SEM; Novosibirsk Province, 54°53′13,5″N, 82°59′36,7″E, 2015-2018)

Phase N		Р			K				
Phase	leaves	stem	panicle	leaves	stem	panicle	leaves	stem	panicle
Seedlings		$2.99 \pm 0.31$			$0.34{\pm}0.03$			2.43±0.35	_
Flowering	$1.08 \pm 0.20$	$0.26 \pm 0.04$	$1.37 \pm 0.25$	0.19±0.01	$0.11 {\pm} 0.02$	$0.28 \pm 0.05$	$1.38 {\pm} 0.11$	$0.61 \pm 0.17$	$1.21 \pm 0.25$
Drying	$0.46 \pm 0.06$	0.24±0.09	$0.93 \pm 0.29$	0.12±0.04	$0.08 {\pm} 0.02$	$0.14 \pm 0.05$	$0.48{\pm}0.03$	$0.26 \pm 0.02$	0.51±0.19

One of the advantages of miscanthus over other energetic plants is the low removal of biophilic elements from the soil, since when the plants desiccate, an intensive outflow of substances from AB to BB occurs (reutilization). In the year of planting (2015), the biomass was not removed from the field due to its small amount (0.8 t/ha), therefore, for comparison, the following vegetation years of the plant (2016-2018) were taken. In the authors' experiment, the final value of the removal of elements depended on the harvesting technology (Table 4). If only the stems were harvested, the nitrogen removal was 23.3, phosphorus 2.8, and potassium 25.9 kg/ha. The amount of reutilization was, respectively, 74, 69, and 76% of the content of the element in the flowering phase. When harvesting with leaves, the alienation of biophilic elements increased by about 2 times, and the amount of reutilization decreased to 43, 34, and 54%, respectively.

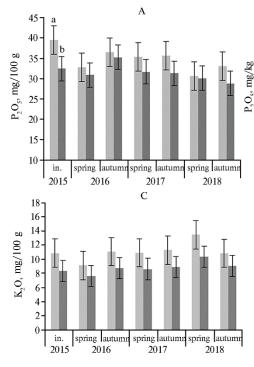
4. Removal and return (kg/ha) of biophilic elements into the soil by *Miscanthus sac*chariflorus cv. Soranovsky plants (n = 12,  $M\pm$ SEM; Novosibirsk Province, 54°53′13,5″N, 82°59′36,7″E, 2016-2018)

Plant organ	Ν	Р	K
F1	owering		
Leaves	65.1±7.3	$5.2 \pm 0.8$	67.4±8.9
Stem	21.1±0.6	$3.6 \pm 0.2$	37.5±2.8
Panicle	$2.4 \pm 0.6$	$0.2 \pm 0.1$	1.8±0.6
Sum (removal)	88.7	8.9	106.7
	Drying		
Leaves	27.3±4.8	$3.1 \pm 0.3$	23.3±3.3
Stem	22.1±1.8	$2.7 \pm 0.1$	25.5±1.5
Panicle	$1.2 \pm 0.6$	$0.1 \pm 0.1$	$0.4 \pm 0.1$
Sum (total removal)	50.6	5.9	49.2
Removal without leaves	23.3	2.8	25.9
Return to soil including leaves and stems	38.1	3.0	57.5
Return to soil excluding leaves (stem harvesting)	65.4	6.1	80.8

To assess the balance of biophilic elements in the soil-plant system, the stock of elements in BB was estimated as the closest source of nutrients. The stock of N, P, K in BB was, respectively, 131, 10, 126 kg/ha, that is, 1.6-2.6 times higher than the amount alienated with the harvest.

Thus, for 3 years of miscanthus vegetation, the maximum removal of N, P, and K with the aboveground plant biomass was, respectively, 150, 18, and 150 kg/ha. It was interesting to observe how this affected the agrochemical properties

of the soil. According to the generally accepted scale [24], the content of phosphorus and potassium in the studied soil is assessed as high. After 4 years of vegetation of miscanthus, the content of phosphorus in the Chirikov extract did not change in both the upper and lower soil horizons (Fig. 1, A). The content of water-soluble forms of the element (Karpinsky extract) at the end of 2018 in the upper soil layer was significantly lower in comparison with the initial value of the indicator, which reflects a decrease in the reserves of readily available phosphorus in the soil during the growth of miscanthus (see Fig. 1, B). It should also be noted that the phosphorus content in the soil remained at the "high" gradation level. The supply of potassium to the soil after 4 years of miscanthus growth did not change (see Fig. 1, C).



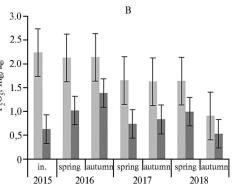


Fig. 1. Dynamics of biophilic elements in the soil during the cultivation of *Miscanthus sacchariflorus* cv. Soranovsky over in the years of observation: A – P according to Chirikov, pobserved = 0.060; B – P according to Karpinsky, pobserved = 0.0019; C – K according to Chirikov, pobserved = 0.9827; a – layer 0-20 cm, b – layer 20-40 cm, in. – initial values; pobserved are calculated for values at the beginning of observations (2015) and at the end of observations (autumn 2018) (n = 9; Novosibirsk Province, 54°53′13,5″N, 82°59′36,7″E). Dara are shown as  $M\pm(t_{0,05} \times SEM)$ .

It is rather difficult to obtain such information on nitrogen in the soil

due to the specificity of the cycle of this element in the agrocenosis. Determining nitrate nitrogen showed quite an expected result: during the vegetation period of miscanthus in 2015-2017, only trace amounts of this compound were present in the soil; in 2018, the authors did not find nitrate nitrogen. A low content of nitrate nitrogen is typical for perennial crops of any cereal perennial grasses [20]; therefore, an insignificant content of nitrate nitrogen in the experiment is not evidence of a deficiency of this element for plants. Thus, for 4 years of the vegetation of miscanthus in the studied agro-gray soil, no significant decrease in the content of the main biophilic elements was found.

The interest in the data on the dynamics of carbon in crops (plantings) of energy crops is associated with the problem of greenhouse gas CO<sub>2</sub> sequestration in SOM fractions. After 4 years of vegetation of miscanthus, the content of C<sub>org.</sub> in the 0-20 cm soil layer was  $1.97\pm0.16\%$  ( $M\pm$ SEM), and in the initial soil  $1.76\pm0.16\%$ , that is, a reliable accumulation of C<sub>org.</sub> in the soil was not established. As it is known, the content of total carbon in soil (C<sub>org.</sub>) is a very stable indicator and it is rather difficult to note its change in short-term experiments

[23]. Nevertheless, rough estimates are possible using the available standard methods, in particular, based on the dynamics of carbon accumulation in BB. Table 5 presents the data on the content and reserves of carbon and nitrogen in mortmass and rhizomes of miscanthus in autumn 2016. As the initial values of the studied parameters, we took the values for one-year fallow, since the miscanthus plantation was formed after the one-year fallowing of a wheat field. The sample in the experiment did not differ significantly in terms of the carbon content in BB (see Table 5). The decrease in the nitrogen content in the mortmass during the cultivation of miscanthus in comparison with the initial soil indicates a less deep processing of organic material in the soil under the miscanthus. The nitrogen content in the rhizomes of miscanthus was significantly lower than in mortmass, so the C/N ratio increased sharply.

5. Content and accumulation of nitrogen and carbon in belowground biomass and in the soil under the fallow and under *Miscanthus sacchariflorus* cv. Soranovsky plants  $(n = 10, M \pm \text{SEM}; \text{Novosibirsk Province}, 54°53'13,5"N, 82°59'36,7"E, autumn 2016)$ 

Sample	Conte	ent, %	C/N	Accumulation, kg/ha		
	Ν	С	C/N	N	С	
Fallow (mortmass)	2.12±0.24	43.39±1.01	25±2.3	40.3±3.9	$760 \pm 80$	
Miscanthus (mortmass)	$0.79 \pm 0.04$	44.61±0.21	66±3.3	41±5.3	$2750 \pm 300$	
Miscanthus (rhizomes)	$0.48 {\pm} 0.05$	44.61±0.65	96±13.3	72±7.3	$4760 \pm 670$	

For two years of miscanthus growth, the carbon stock in mortmass increased 3.6 times compared to the initial value (fallow). As a result, the total carbon stock in the BB of miscanthus was 7.5 t/ha, that is, it increased by about 10 times in comparison with the indicator established before the planting. The increase in the total carbon stock in the BB occurred mainly due to the rhizomes where the C/N ratio reached 100.

Thus, the carbon sequestration in the BB in the first 2 years of the vegetation period of miscanthus was 1 t C/ha per year in mortmass and 2 t C/ha per year in rhizomes. These data give a general idea of the dynamics of SOM without taking into account the changes occurring in the fractions of organic matter of the soil itself. A fairly simple method is known for assessing the amount of the mobile fraction of SOM, which is most accessible for microbial decomposition. The soil is incubated until the release of C-CO<sub>2</sub> ceases to increase [23]. However, it is clear that this procedure is quite long-term. In a laboratory experiment, the authors tried to estimate the amount of the most accessible part of SOM accumulated by miscanthus during the first 2 years of vegetation. For this, the soil from under fallow and from under miscanthus was incubated under optimal conditions of heat and humidity. After 6 weeks, the rate of CO<sub>2</sub> production in the samples of both soils reached the same value (Fig. 2).

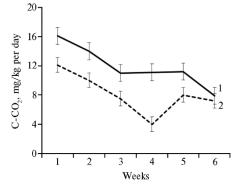


Fig. 2. Comparative rate of C-CO<sub>2</sub> production by soil under *Miscanthus sacchariflorus* cv. Soranovsky (1) and one-year fallow land on which the plantation was formed (a basal level) [2]; pobserved = 0.004 for weeks 1-5, pobserved = 0.980 for week 6 (lab test, n = 3; Novosibirsk Province, 54°53′13,5″N, 82°59′36,7″E, 2016). Data are shown as  $M\pm(n_{.05} \times \text{SEM})$ .

For 6 weeks, the total release of C-CO<sub>2</sub> from the soil under miscanthus was  $1039\pm25$  kg/ha, under the fallow  $750\pm15$  kg/ha, that is, the C accumulation of the mobile part of organic matter

under miscanthus was almost 300 kg/ha more than under the fallow. It follows

from this that under the plantings of *M. sacchariflorus* of the cultivar Soranovsky for two vegetation periods (2015-2016), about 300 kg C/ha of mobile SOM was accumulated in the soil in comparison with its initial stock in the soil of the annual fallow land, on which the plantation was formed.

The content of SOM is due to the resulting rate between the rate of entry of organic residues into the soil and their mineralization to the final products. The changes in the components of the balance of these events are small and, as a consequence, the intensity of SOM accumulation is difficult to estimate. However, with the help of laboratory measurements, it is possible to determine the tendencies of this process by assessing the respiratory response of the soil to the introduction of plant residues and thus to measure the ratio of removed and retained OM in the soil. The response of the respiratory activity of the soil to the input of new organic matter serves as one of the criteria for the intensity of carbon metabolism in the soil: the lower the response, the lower the rate of mineralization, that is, the dynamics are directed towards the accumulation of carbon in the SOM.

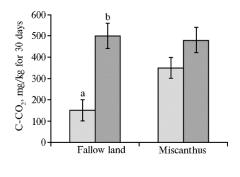


Fig. 3. C-CO2 production by the soil under one-year fallow on which the plantation was formed (a basal level) and under *Miscanthus sacchariflorus* cv. Soranovsky in control (a) and with wheat straw incorporation (b); pobserved < 0.001 for the fallow, pobserved = 0.038 for miscanthus (lab test, n = 3; Novosibirsk Province,  $54^{\circ}53'13,5''N$ ,  $82^{\circ}59'36,7''E$ , 2016). Data are shown as  $M \pm (r_{0.05} \times \text{SEM})$ .

Fig. 3 shows the results of such an experiment. Of incorporated C (600 mg/kg as straw) from the fallow soil, approximately 350-400 mg C-CO<sub>2</sub>/kg were released, while

for the soil under miscanthus, the differences between the variants (control and experiment) were minimal. Respiratory response for fallow soil was 250%, for the soil under miscanthus 40%. From the obtained data, it follows that the microbial decomposition of the new organic matter in the soil during the cultivation of miscanthus was much weaker than against the background of fallow. The inhibition of mineralization processes in the variant with miscanthus is obviously associated with the accumulation of plant residues with a high C/N ratio.

Harvesting period	Value	M. sinensis	M. sacchariflorus	M. giganteus
Autumn	Average	12	15	26
	Min-max	4.6-22	1.4-35.0	13.8-37.8
Winter	Average	9	10	19
	Min-max	3.1-17.6	0.4-22.4	9.2-26.4

6. Yield (absolutely dry matter, t/ha) of different species of *Miscanthus* for the first 3 years of vegetation calculated according to literature data [25, 26]

Thus, the presented work summarizes the results of the first stage of studying the possibility of growing miscanthus for biomass in Siberia. Here, the data were obtained in the harshest climatic conditions (arid cold climate) compared to the published results worldwide. Nevertheless, the average yield of *M. sacchariflorus* of the cultivar Soranovsky, established in the work (10-13 t/ha) (see Table 2), is within the values typical for this species (Table 6). When analyzing Table 6, attention is drawn to the very wide range of fluctuations in the yield of miscanthus in the world. This is due to the fact that the authors of the reviews [25, 26] tried to collect the maximum amount of factual data obtained under a variety of conditions (namely, climatic, soil, and agrotechnical ones). At the same time, the range of yield fluctuations in the M. sacchariflorus species was wider than in the other two species. This once again confirms the conclusion of other authors [11] regarding a more pronounced dependence of the yield of M. sacchariflorus on soil and climatic conditions.

Country region	Aver	age annual values	Smaaina	Yield,	Removal, kg/ha			References
Country, region	T, ℃	precipitation, mm	Species	t/ha	Ν	Р	K	References
USA:								[13, 29]
State of New Jersey	11.2	1211	M. giganteus	9.5	27.1	4.3	32.6	
State of Kentucky	12.8	1166	M. giganteus	19.0	112.9	14.4	74.5	
State of Illinois	11.1	1043	M. giganteus	15.6	54.2	3.4	39.6	
State of Nebraska	9.7	704	M. giganteus	27.7	116	7.9	165	
Spain	13.9	100.3	M. giganteus	17.6	46.4	3.9	23.0	[14]
France	11.5	557	M. giganteus	16.9				[16]
France (autumn/winter)	17.8	390	M. giganteus	22/15.5	101/29	19/7	139/45	[27]
Russia:								[11]
Moscow	4.5	620	M. sinensis	7.8				
			M. giganteus	5.7				
			M. sacchariflorus	4.2				
Western Siberia	1.7	459	M. sacchariflorus	13/7	50/23	6/3	49/26	Current research
N o t e. Slashes indicate absence of data.	the val	ues for harvesting th	ne entire biomass	and only	the ste	ms. 7	The gap	s indicate the

7. Removal of biophilic elements with biomass of *Miscanthus* in different regions of the world (according to literature data)

The comparison of the data we obtained on the removal of biophilic elements is rather difficult to perform both geographically and by the species of miscanthus. For Russia, we did not find such information; moreover, in other countries, this information concerns mainly the species *M. giganteus*. As it can be seen from Table 7, nitrogen and potassium are most intensively removed with the biomass of miscanthus. The total removal depends on the yield of the crop and the timing of harvesting. As the literature data show [27, 28], during winter harvesting, in comparison with autumn harvesting, the removal of elements can decrease three times. The calculation of the removal of elements per unit of yield is presented in Table 8. The indicator varies greatly from different sources. In our experiment, the specific removal was rather low and approached the indicators [28] obtained during winter harvesting.

**8.** Removal of biophilic elements with biomass of *Miscanthus* (kg/t) in different regions (calculations per unit of the yield mass are based on the literature data of Table 7)

Country, region	Timing/harvesting technology	Ν	Р	Κ
USA, State of New Jersey	Winter	2.9	0.5	3.4
USA, State of Kentucky	Autumn	5.9	0.8	3.9
USA, State of Illinois	Autumn	3.5	0.2	2.5
USA, State of Nebraska	Winter	4.2	0.3	6.0
Spain	Winter	2.6	0.2	1.3
France	Autumn	4.6	0.9	6.3
	Winter	1.9	0.5	2.9
Russia, Western Siberia (current research)	Harvesting with leaves	3.0	0.4	2.9
	Harvesting the stems	1.4	0.2	1.5

Our data on the yield and removal of elements with biomass are, on the whole, close to those obtained in the world for other soils and climatic conditions.

As to the dynamics of biophilic elements in the soil-plant system, according to the literature data [13, 30], the most deficient element in perennial plantings of miscanthus is phosphorus, which, despite its relatively low removal, is poorly reutilized. The same tendency is observed in the authors' experiment, although after 4 years of cultivation of miscanthus, a decrease was noted in the content of only the most mobile fraction of this element in the soil. The analysis of the ratio of the reserves of elements in BB and their removal shows that already in the first years of the vegetation period, a sufficient supply of nitrogen, phosphorus, and potassium was formed to successfully replace the removal of elements with the biomass of miscanthus. The same pattern was revealed by the studies in other regions of the world. In particular, Dohleman et al. [12] in the USA (the State of Illinois, 40°03'21.3"N, 88°12'3.4"W) showed that for 5-7 years of vegetation, the yield of *M. giganteus* (about 40 ft/ha) did not decrease even without fertilization. The authors attribute this to the dynamic release of elements, in particular nitrogen, from rhizomes and, in general, from BB. Thus, the nitrogen reserve in rhizomes, which in April was 264 kg/ha, decreased to 145 kg/ha by June, and then, due to reutilization from AB, increased by December to 373 kg/ha. From the presented data, it follows that 45% of nitrogen contained in the rhizomes was actively absorbed by the growing biomass. In our experiment, with nitrogen removal equal to 50 kg/ha (including leaves) or 23 kg/ha (when harvesting only stems), the reserve of the element in the rhizomes was 130 kg/ha, that is, the removal compensation was sufficient.

According to our findings, over 4 years of development of *M. sacchariflorus* plants of the cultivar Soranovsky, no signs of soil depletion by biophilic elements were found. Moreover, during this period, a significant supply of nitrogen and carbon was formed in BB.

The possibility of the carbon sequestration of greenhouse gas CO<sub>2</sub> in the SOM fractions during the growth of miscanthus as a whole is beyond doubt. In particular, a quantitative forecast has been developed for the UK, according to which, upon obtaining stable yields of miscanthus of 12 t/ha per year, by 2090 it is possible to stabilize the positive dynamics of SOM [18]. Such expectations are mainly based on the colossal productivity of the AB and BB of miscanthus. The value of carbon sequestration in BB in the authors' experiment (approximately 1 t C/ha in mortmass, 2 t C/ha in rhizomes per year) is guite close to the literature data. Thus, according to the generalized data by Robertson et al. [17], under 3-4year-old *M. giganteus* plantings, the average annual carbon accumulation in the roots was about 860 kg C/ha, in rhizomes 2660 kg C/ha. The transfer of this carbon to SOM is a dynamic process that depends on the rate of biomass mineralization to end products, which is determined by specific soil-climatic and agrotechnical conditions. We have experimentally shown that, under miscanthus, the intensity of this process decreased sharply already in the first 2 years of the vegetation due to a significant increase in the C/N ratio, which may be a prerequisite for the accumulation of organic matter in the soil. As it is known, the imbalance between biophilic elements is the most effective natural mechanism that prevents the rapid decomposition of biomass on the planet to end products [31]. In our experiment, we did not find a significant increase in the total carbon content in the soil. According to the publications, even after 20 years of *M. giganteus* vegetation, the expected increase in the content of SOM may not be observed [17]. One of the probable reasons is the gradual replacement of the already existing SOM with a new one. Thus, it was reported [15] that for 6 years in 10-year-old M. giganteus plantings, the annual replenishment of SOM with C4 plant carbon was 780 kg/ha, and 68% of detritus carbon in the 0-10 cm soil layer belonged to miscanthus. Moreover, the share of miscanthus carbon in the fraction resistant to NaOCl oxidation was 15%. The authors emphasize that the replenishment of stable fractions of SOM can occur in a relatively short period of time. Researchers continue to study the reasons why the accumulation of SOM under miscanthus often does not match the amount of incoming plant residues. In particular, with the help of the  ${}^{13}C$  isotope, it was shown that the residues of C4 plants in the soil were mineralized to final products faster in comparison with those of C3

plants [19]. Thus, the problem of air carbon sequestration in SOM fractions is rather complicated and requires additional study under specific soil and climatic conditions.

Thus, for Miscanthus sacchariflorus of the cultivar Soranovsky, the possibility of its cultivation in the arid and cold climate of Western Siberia was assessed. It was found that on the agro-gray soil of the central forest-steppe of the Ob region during the formation of perennial plantings (1-4 years), the AB value of miscanthus reached 13 t/ha, and the average yield for 4 years was 10 t/ha. These indicators are generally comparable to the productivity observed in *M. sacchariflorus* in the world under more favorable climatic conditions. The ratio of the removal of the main biophilic elements with the yield of AB, as well as a sufficient reserve of the latter in BB, allows concluding that there is no depletion of the soil with elements of mineral nutrition of plants during the cultivation of miscanthus. This conclusion is confirmed by the stability of the agrochemical properties of the studied soil over the observation period. Rough estimates of the components of the carbon balance (C accumulation in mortmass of 1 t/ha, in the mobile SOM fraction of 150 kg/ha per year) in the agroecosystem showed the objective prerequisites for atmospheric carbon sequestration in fractions of SOM. Thus, the cultivation of *M. sacchariflorus* for biomass in Siberia is guite reasonable ecologically, agrotechnically, and economically.

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