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Growth of Schrenk's Spruce (*Picea schrenkiana*) Seedlings Related to the Pre-Sowing Stimulating Seed Treatment

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Abstract. The uniqueness of the biological properties and the occurrence in nature of the Schrenk's Spruce (Picea schrenkiana Fisch. & C.A. Mey.), its great forestry significance, and important eco-stabilizing functions of its stands in the Northern Tian Shan are of great interest to a wide range of researchers in many countries. However, many characteristics of this endemic species have been poorly studied so far, and information on the effectiveness of its introduction is extremely limited. We investigated the characteristics of growth and development of spruce seedlings in the conditions of the Nizhny Novgorod region. The seeds were harvested in natural plantations on the territory of the Almaty State Nature Reserve, which is located in the Central part of the Trans-Ili Alatau in South-Eastern Kazakhstan. They were sown in an open ground nursery under stimulating treatment with the following bioactive stimulants: siliplant, ferovit, ecofus, kornevin, heteroauxin, zircon, extrasol, epin, and albit. The soaking of all the seeds in water at the same exposure for 24 h served as control. The aftereffect of bioactive substances in the pre-sowing stimulating treatment of Shrenk's spruce seeds was found to activate the growth processes of the aboveground part of its seedlings during the introduction into the Nizhny Novgorod region. It was found that the biennial part of seedlings axial shoot had unequal length in different experimental options. The highest values were achieved in the variants with the use of the following stimulants: ferovit $(5.95\pm$ ± 0.02 cm), heteroauxin (5.62 ± 0.16 cm), ecofus (5.47 ± 0.32 cm), and albit (5.49 ± 0.015 cm). The generalized value was 5.08 ± 0.07 cm. The growth characteristics of seedlings also respond to the stimulating effect. The best results in this case were shown by ferovit (2.25±0.023 cm) and heteroauxin (1.96±0.17 cm). The differences in the number of side shoots are less contrasting. However, we can also see the previously noted trends: ferovit $(1.42\pm0.25 \text{ pcs})$; heteroauxin (1.34±0.19 pcs); and albit (1.31±0.19 pcs). We obtained the certificate of sufficient resource of natural conditions of the region for seed breeding of Schrenk's spruce in the open ground and the significant potential of its introduction to the Nizhny Novgorod region.

Keywords: Schrenk's spruce, introduction, seed propagation, growth stimulants, seedlings, morphometrics, analysis of variance, Nizhny Novgorod region

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Научная статья

Рост сеянцев ели Шренка (*Picea schrenkiana*) при предпосевной стимулирующей обработке семян

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Аннотация. Уникальность биологии и распространения ели Шренка (*Picea schrenkiana* Fisch. & C.A. Mey.), большое лесохозяйственное значение и важные эколого-стабилизирующие функции ее насаждений на Северном Тянь-Шане обуславливают неослабевающий интерес исследователей разных стран к этому эндемичному виду. Однако на сегодняшний день многие его характеристики изучены слабо, крайне ограничены сведения об эффективности интродукции. Нами исследованы особенности роста и развития сеянцев ели Шренка в условиях Нижегородской области. Семена были заготовлены в естественных насаждениях на территории Алматинского государственного природного заповедника, который расположен в центральной части Заилийского Алатау на юго-востоке Казахстана. Высевание семян производили в питомнике открытого грунта при стимулирующей обработке биоактивными препаратами: силиплантом, феровитом, экофусом, корневином, гетероауксином, цирконом, экстрасолом, эпином и альбитом. Контролем выступало намачивание семян в воде в единой для всех вариантов экспозиции 24 ч. Эффект применения биоактивных веществ при предпосевной стимулирующей обработке семян ели Шренка выразился

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в активизации ростовых процессов надземной части ее сеянцев при интродукции в Нижегородскую область. Установлена неодинаковая длина 2-летней части осевого побега сеянцев по вариантам опыта. Наибольшие значения достигнуты в вариантах применения феровита (5,95±0,02 см), гетероауксина (5,62±0,16 см), экофуса (5,47±0,32 см) и альбита (5,49±0,015 см). Среднее значение составило 5,08±0,07 см. Ростовые характеристики сеянцев также реагируют на стимулирующее воздействие. Лучшие результаты и в этом случае показали препараты феровит (2,25±0,023 см) и гетероауксин (1,96±0,17 см). Менее контрастны различия по количеству боковых побегов. Однако и здесь прослеживаются ранее отмеченные тенденции: феровит – 1,42±0,25 шт., гетероауксин – 1,34±0,19 шт., альбит – 1,31±0,19 шт. Получено свидетельство о достаточном ресурсе природных условий региона для семенного размножения ели Шренка в открытом грунте и о существенном потенциале ее интродукции в Нижегородскую область.

Ключевые слова: ель Шренка, интродукция, семенное размножение, стимуляторы роста, сеянцы, морфометрия, дисперсионный анализ, Нижегородская область

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Introduction

Schrenk's spruce (Picea schrenkiana Fisch. & C.A. Mey.) is recognized as the main forest-forming species of the Northern Tian Shan [16, 47, 50] and, being an endemic species of the natural flora of this region, belongs to the dominant species of the forest cover [17, 25, 27, 48]. Having a large economic value [20, 21, 22, 29] and performing important eco-stabilizing functions [2, 18, 50], Schrenk's spruce is under constant scrutiny by researchers [1, 15, 19, 32, 33, 44]. Its occurrence in nature is described in detail [13, 25, 27, 51], the spatial and population structure of natural stands of this breed is well studied [4-6, 13], ecological reactions and biological features are described [16-18, 45, 46], data on karyology is accumulated [6, 15, 30, 43, 55]. The problems of Schrenk's spruce natural regeneration are considered in Kazakhstan and Kyrgyzstan [20, 32, 40]. The features of the Schrenk's spruce seedlings development [49] are studied and the relevance of creating its forest crops is shown [28, 34, 54] in the context of these problems. However, the taxonomy and systematic position, intraspecific variability and form diversity of this species remain debatable [4-6, 47, 48]. In the Russian Federation, the list of economically used spruce species is constantly updated, including non-regional species [10-12, 36, 37]. The Schrenk's spruce is the least studied among them. We introduced it into the Nizhny Novgorod region for the first time taking into account the formed theoretical concepts [3, 7, 8, 38, 39, 45].

The research aims at determining the prospects for the introduction of Schrenk's spruce into the Nizhny Novgorod region via evaluation of the growth processes of its seedlings against the background of stimulating treatment with bioactive compounds.

Research objects and methods

We studied two-year-old seedlings of Schrenk's spruce grown out of seeds harvested in the Almaty State Nature Reserve natural plantations. The reserve is located in the Central part of the Trans-Ili Alatau in the South-East of Kazakhstan (coordinates: 43° 06' 00" N 77^{\circ} 19' 00" E). The high-altitude borders of the spruce forest belt

are 1,600–2,800 m above sea level. Seedlings were grown in the open ground with artificial irrigation at the experimental site of the Faculty of Forestry of the Nizhny Novgorod State Agricultural Academy. Its geographical coordinates are 56° 19' 43" N 44° 00' 07" E, and the height above sea level is 136 m.

The methodological framework for developing operating procedures consisted of theoretical ideas on the basic requirements for the organization of an experiment (its typicality, suitability, expediency and reliability), the principles of the only logical difference and the randomized distribution of objects when they are included in the sample [23, 31, 41, 42, 52, 56]. That is why the differentiating influence of chronographic and environmental factors was levelled by simultaneous recording of all plants, including into the experiment scheme only single-aged seedlings growing in the same soil and climatic conditions, in the same agricultural background and in the same phenological state. The organizational and methodological scheme provided the construction of a one-factor variance complex and the performance of statistical analysis [14, 23, 26, 31, 41, 42, 52, 56].

Based on the fact that one of the key assessments of the plants introduction results is a set of their seed propagation indicators, we studied the morphometric parameters of the aboveground part and root systems of Schrenk's spruce seedlings in the conditions of their transfer to the Nizhny Novgorod region. At the same time, the stimulating effect of biologically active substances in pre-sowing seed treatment, which is described relative to an extensive list of tree species [24, 53], including spruce [9], was also considered. We tested the following preparations formed the test options: siliplant -0.05 % (option 1); ferovit 0.15 % (option 2); ecofus -0.3 % (option 3); kornevin – 0.01 % (option 4); heteroauxin – 0.02 % (option 5); zircon – 0.025 % (option 6); extrasol - 1.5 % (option 7); epin - 0.05 % (option 8); albite -0.1 % (option 9). The soaking of all the seeds in water (Water option) at the same exposure for 24 h served as Control. The average value generalized for the entire data array is called Total. The length of the aerial part and roots were measured with a ruler with accuracy of 0.5 mm; diameter – with an electronic caliper (Digital Caliper SH20) to accuracy of 0.01 mm. The repetition of the experience is threefold. A total of 346 seedlings were biometrized.

Results and discussion

The effectiveness of Schrenk's spruce seed propagation in the conditions of introduction was evaluated by the parameters of growth of seedlings sowed in the open ground and treated with stimulators (fig. 1–3). The length of the two-year part of the axial shoot of Schrenk's spruce seedlings was not the same according to the experimental options (fig. 1*a*). The highest values were achieved when the following stimulators were used: ferovit (option 2; 5.95 ± 0.024 cm) and heteroauxin (option 5; 5.62 ± 0.16 cm).

The estimates for ecofus (option 3) and albit (option 9) were slightly lower: 5.47 ± 0.32 cm and 5.49 ± 0.015 cm, respectively. Even weaker is the effect of siliplant (option 1) and epin (option 8) use: 4.29 ± 0.17 and 4.31 ± 17.30 cm, respectively. A similar situation is in the group of seeds soaked in water before the sowing (Control option): 4.27 ± 0.14 cm. At the same time, the difference in estimates is clearly noticeable, and the largest of them exceeded the smallest by 1.68 cm or 1.39 times. Generalized value for the entire data array (Total option) was 5.08 ± 0.07 cm. For the other parameters of the axial shoot of seedlings, the marked trends were generally preserved (fig. 1*b*, 1*c*).



Fig. 1. Parameters of the axial shoot of Schrenk's spruce seedlings: a – length of the two-year part of the axial shoot; b – total height of the aboveground part; c – diameter of the root collar

The increment characteristics of seedlings also showed heterogeneity in different options of the stimulating effect (fig. 2).

Like in the previous option, the highest current increment values were registered in the application with ferovit (option 2) and heteroauxin (option 5): 2.25 ± 0.023 cm and 1.96 ± 0.17 cm, respectively. They significantly exceeded the control value (1.36 ± 0.09 cm) and the generalized average value (1.70 ± 0.05 cm).

The use of albit (option 9) provoked a comparable increase $(1.90\pm0.17 \text{ cm})$. Other stimulants showed less effectiveness in this regard, which can be attributed to the above-mentioned increment characteristics in general (fig. 2b, 2c). The general estimates of linear growth of the axial and lateral shoots at the time of measuring, as well as their average values (fig. 3a, 3b), retained the main trends established for the characteristics of the current increment (fig. 2). The differences in the number of lateral shoots in the aboveground part of the seedlings are less contrasting (fig. 3c), however, the previously noted ratios are also observed here. In particular, the use of ferovite (option 2), heteroauxin (option 5) and albite (option 9) was accompanied by high values, while siliplant (option 1) and epin (option 8) had the lowest values. They were close to the control values.



Fig. 2. Linear increment of Schrenk's spruce seedlings: a – current increment of the axial shoot; b – total current increment of shoots; c – average current increment of shoots

So it was found that the stimulating seed treatment of Schrenk's spruce seedlings by different stimulators provokes different changes in their growth and development features. The following features of growth and development were studied: feature 1 – height of two-year aboveground part of the seedlings without the last increment, cm; feature 2 – diameter of the root collar of seedlings, mm; feature 3 – current linear growth of the axial shoot in height, cm; feature 4 – total height of the aboveground part of seedlings including the last growth, cm; feature 5 – total current linear growth of shoots including axial shoots, cm; feature 6 – average current linear growth of shoots including axial shoots, cm; feature 7 – total linear shoot growth on all the whorls, cm; feature 8 – overall average linear growth of shoots including axial shoots, cm; feature 9 – total number of side shoots in all the seedling whorls, pcs. Their relative estimates expressed in terms of the coefficient of variation (C_{yp} %) are represented in table 1.

It is noticeable that the variability of linear parameters of the Schrenk's spruce seedlings axial shoot within the boundaries of the generalized data array (Total option) refers to the average (feature 1) and increased (features 2 and 4) level on the Mamaev's scale. The coefficients of variation were 23.94, 26.30 and 27.28 %, respectively. Other characteristics of seedlings are more variable, and their corresponding ratings are high, such as feature 6, feature 8, feature 9 (42.95, 39.71 and 44.03 %), and very high, such as feature 3, feature 5, feature 7 (58.81, 57.01 and 66.47 %) on the same scale.



Fig. 3. Total growth of Schrenk's spruce seedlings: a – total growth of shoots; b – total average growth of shoots; c – number of side shoots in the aboveground part

Table 1

Variability of characteristics of the aboveground part of Schrenk's spruce seedlings

Option	Feature									
	1	2	3	4	5	6	7	8	9	
1	17.51	19.70	45.50	18.17	54.49	35.78	51.09	36.01	42.71	
2	22.93	23.26	58.12	26.31	49.96	41.32	59.23	39.18	43.14	
3	23.24	32.64	51.51	22.56	47.57	37.88	56.30	33.99	42.36	
4	16.38	18.63	36.42	16.23	49.37	33.48	50.24	33.03	42.83	
5	20.34	24.35	62.18	25.72	54.53	44.30	64.90	42.25	43.27	
6	23.22	24.55	52.99	23.21	49.39	39.35	51.07	36.77	41.94	
7	28.11	29.34	59.74	30.58	60.99	44.12	69.34	39.32	44.39	
8	17.30	17.48	47.55	17.12	49.72	31.90	46.13	31.72	37.74	
9	18.75	24.01	61.68	25.59	53.83	41.58	64.45	39.51	42.36	
Water	17.22	16.64	35.93	16.78	53.06	29.97	55.48	30.32	48.92	
Total	23.94	26.30	58.81	27.28	57.01	42.95	66.47	39.71	44.03	

Absolute estimates of the variability of the analyzed indicators were characterized by ranges of limits, which were established at the stage of statistical analysis. In particular, the absolute maximum of the total height of the aboveground part (12.5 cm) exceeded the absolute minimum of the same indicator (3.7 cm) by 3.38 times. The range of root collar diameter limits was 2.21 mm (from 0.64 mm to 2.85 mm). The linear current growth of the axial shoot had a range of values from 0 to 4.5 cm, since it was not registered for individual seedlings. The total increment of shoots in the records for all seedling whorls had a spread of values from 0.3 to 16.8 cm, which formed a range of 16.5 cm and exceeded the limits by 56 times.

The effectiveness of the use of biologically active substances to stimulate the germination of Schrenk's spruce seeds (at the background of aftereffect) was evaluated by the one-way ANOVA (table 2).

Table 2

	F _{act}	Share of factor influence $(h^2 \pm s_h^2)$							Criteria for	
Feature		by Plokhinsky's algorithm			by Snedecor's algorithm			differences		
		h ²	$\pm s_h^2$	F_h^2	h ²	$\pm s_h^2$	F_h^2	LSD ₀₅	D ₀₅	
1	10.44	0.2185	0.0209	10.436	0.2161	0.0210	10.293	0.517	0.820	
2	8.30	0.1819	0.0219	8.302	0.1758	0.0221	7.965	0.165	0.262	
3	3.64	0.0887	0.0244	3.635	0.0715	0.0249	2.874	0.458	0.727	
4	10.26	0.2156	0.0210	10.264	0.2130	0.0211	10.105	0.786	1.248	
5	5.20	0.1223	0.0235	5.200	0.1093	0.0239	4.582	0.869	1.379	
6	5.46	0.1276	0.0234	5.462	0.1153	0.0237	4.867	0.312	0.495	
7	6.04	0.1393	0.0231	6.041	0.1284	0.0233	5.499	1.282	2.034	
8	4.25	0.1022	0.0240	4.251	0.0868	0.0245	3.546	0.265	0.421	
9	2.53	0.0635	0.0251	2.533	0.0429	0.0256	1.673	0.572	0.908	

The effect of stimulants on the growth of Schrenk's spruce seedlings

Here we used the following statistical indicators: F_{act} – experimental value of Fisher test; F_{05} – table value of Fisher criterion for 5 % significance level (F_{05} = 1.91); F_{01} – table value of Fisher test at 1 % significance level (F_{01} = 2.48); h^2 – the proportion of influence of the organized factors; $\pm s_h^2$ – error share of influence of the organized factors; F_h^2 – the Fisher criterion in the assessment of the validity of the share of influence of the organized factors; LSD_{05} – least significance level.

The one-way ANOVA confidently refutes the null hypothesis that there are no differences if different growth stimulants are used. The calculated values of the F-criterion were almost always greater than their acceptable table limits at the significance levels accepted in the article. This is true relative for almost all considered parameters of Schrenk's spruce seedlings. The only exception is the total number of side shoots in all seedling whorls (feature 9), where the significance of differences is estimated at 5 % significance and not at 1 % significance.

Estimates of growth stimulators impact on the development of spruce seedlings in calculations according the the Plokhinsky's algorithm are different for various parameters. The highest values of this indicator concerned the length of a two-year part of axial shoot of seedlings (feature 1) and the total height of the aerial part of seedlings from the last growth (feature 4). The estimates are quite reliable and were $21.85\pm2.09 \%$ ($F_h^2 = 10.44$) and $21.56\pm2.10 \%$ ($F_h^2 = 10.26$), respectively. The minimum impact was registered by the current linear growth of the axial shoot in height (feature 3) and by the total number of lateral shoots in all the seedling whorls (feature 9). In the first case, the estimates were reliable at the 5 and 1 % significance levels and amounted to $8.87\pm2.44 \%$ ($F_h^2 = 3.64$). In the second case, they were reliable at 5 % and unreliable at 1 % significance level and reached the level of $6.35\pm2.51 \%$ ($F_h^2 = 2.53$). Calculations using the Snedekor's algorithm showed mostly comparable results (table 2).

Significant difference estimates (LSD₀₅ and D₀₅) allowed us to determine between which options of stimulating treatment the difference in average values corresponds to the level of significance. In particular, it was possible to determine which options of stimulating treatment significantly differed from soaking seeds in water for a particular feature of seedlings. Thus, the control value in the length of the two-year part of the axial shoot (feature 1) did not significantly differ with that received after the use of biologically active substances such as siliplant (option 1), kornevin (option 4), and epin (option 8). This is confirmed by the least significant difference (LSD_{05}) and the criterion of Tukey (D_{05}) . This conclusion is also true for a range of other features: diameter of the seedling root collar (feature 2); current linear growth of the axial shoot in height (feature 3); total height of the aboveground part of seedlings with the last growth (feature 4). As for other features of the aboveground part, the situation is different. For example, for the total current linear growth of shoots, including axial shoots (feature 5), the control group did not show significant differences with 6 options of stimulating treatment, and only in three cases the aftereffect was confirmed by the criteria of significant differences: option 2 – ferovit; option 5 – heteroauxin; and option 9 – albit.

Conclusion

The aftereffect of bioactive substances in the pre-sowing stimulating treatment of Shrenk's spruce seeds is preserved. It is expressed in the activation of growth processes of the aboveground part of its seedlings during the introduction in the Nizhny Novgorod region.

The effectiveness of different bioactive substances is not the same for different growth indicators of the seedling aboveground part and has a specific level of manifestation. For some of them, a significant excess of tested indicator values in relation to the control was achieved, confirmed by the analysis of variance. The best results for most of the seedling characteristics were obtained by the use of ferovit, heteroauxin, and albit stimulators. The last circumstance necessitates making a differentiated approach to the selection of biostimulants used to expand the possibilities of the Schrenk's spruce introduction by seed material.

The registered level of growth and development of the aboveground part of Schrenk's spruce seedlings in the ground indicates a sufficient resource for seed reproduction in the region's natural conditions and a significant potential for its introduction in the Nizhny Novgorod region.

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