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## NATURAL STIMULANT FOR LONG-TERM CONIFEROUS SEEDS

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### Abstract

The paper considers the possibility of using aqueous extracts of conifers to increase the germination of seeds of Scots pine for long-term storage. It was found out that in the woody greens of conifers there are substances with provitamin and growth-stimulating activity, namely carotenoids (0.14 g/l) and phenolic compounds (2.50 g/l). Pine tree greens selected in the Mansky district of the Krasnoyarsk Territory were used as raw materials. Extracts were prepared from pine tree greens as follows: dried and crushed tree greens were extracted with tert-butylmethyl ether. The extract was treated with an aqueous solution of an alkaline agent, then an aqueous-alkaline solution was separated. Distilled water was used as a control. For the experiment, seeds of ordinary pine harvested in 2017, 2018, 2019, 2020 and 2021, provided by the Krasnoyarsk forest seed station, were used. The characteristics of the seeds comply with GOST 14161-86. The seeds were germinated in Petri dishes, filter paper was used as a litter. The seedlings were counted on the 15th day. Per 100 pcs. the following concentrations of the extract of an aqueous solution were taken from the seeds, ml of extract per 1 liter: 0.25:1; 0.5:1; 0,10:1; 0,05:1; 0,025:1. With increasing shelf life, seed germination decreases.

The highest seed germination is observed when using an extract with a concentration of extractives (0.05:1) and seeds of the 2021 harvest year. The results of the research can be recommended to increase the germination of long-term storage pine seeds.

**Keywords:** growth-stimulating activity, aqueous extracts of conifers, *Pinus sylvestris*L.

## **Introduction**

In cutting areas, wood waste accounts for up to 35% of the total biomass of wood, of which 17% is wood greens [1]. These wastes can be used in various industries [2-4]. Currently, new innovative technologies are being developed for the integrated processing of logging waste [5]. One of them is the isolation of biologically active substances from woody greens - plant growth stimulants. Experiments on their use have shown that these preparations provide an increase in the germination of coniferous seeds and plant resistance to diseases and the yield of planting material per unit area [6].

The main task of research in the cultivation of forest crops is to study existing and search for new biostimulants of plant growth. Coniferous wood greens can serve as a promising raw material for the preparation of aqueous extracts [7].

One of the urgent tasks of forestry at the present time is to preserve and improve the sowing qualities of seeds of economically valuable coniferous species. These include Scots pine, Siberian spruce and Siberian cedar pine. With long-term storage of seeds, the sowing qualities and germination are reduced. There are various ways to activate it to increase it. One of them is soaking in solutions of various stimulants. A review of plant growth stimulants, the active ingredient of which are triterpene acids obtained from the needles of Siberian fir, is given in the work of V.A. Raldugin [8]. They activate biological and immune processes in plants, including stimulating germination and growth-stimulating activity of seeds. Such preparations as Novosil, a growth stimulant based on fir triterpene acids [9], as well as Gibbor-M concentrate, a plant growth stimulant containing a group of natural plant growth hormones (gibberellins) that regulate plant growth and development processes, allow to increase crop yield and germination by 10-40%. The effectiveness of Gibberellin A3 can be observed in accelerating seed germination [10].

The aim of the work was to study aqueous solutions of coniferous woody greens to increase the germination of long-term pine seeds. The task of the research included:

- establishment of factors affecting the germination of coniferous seeds;
- selection of growth-stimulating agents based on water-alkaline extracts of coniferous woody greens.

The influence of extracts of coniferous woody greens of various concentrations on the sowing qualities of long-stored seeds is considered. A positive effect has been shown with the shelf life of scots pine seeds up to seven years [11].

## **THE EXPERIMENTAL PART**

Woody greens (pine, spruce and fir in equal proportions) selected in the Mansky district were used as raw materials for the preparation of coniferous aquatic extracts. Extracts were prepared from the woody greens of conifers in the following way: air-dry raw materials were taken, crushed on a screw granulator to a particle size of 0.5 - 1 mm. It was extracted with a mixture (ethyl acetate + diethyl ether) under stirring and at room temperature. Then the extract was treated with alkali, the aqueous solution was separated from the organic phase and dried to a powder[12]. The product was infused in water with stirring at a temperature of 25 ° C, a hydromodule of 1:10. The solid residue was separated by centrifugation followed by filtration. The resulting aqueous infusion contained sodium salts of triterpene acids with a concentration of 0.6 to 0.7 wt.%.

For the experiment, seeds of ordinary pine harvested in 2017, 2018, 2019, 2020 and 2021, provided by the Krasnoyarsk forest seed station, were used. The characteristics of the seeds comply with GOST 14161-86 [13]. Seed preparation and methods of their germination were carried out according to GOST 13056.6-97[14]. The preparation was carried out in the following sequence: the seeds were placed for a day in aqueous solutions of coniferous plants of various concentrations; the prepared seeds of 100 pcs. were laid out in Petri dishes, on wet filter paper, cut according to the size of the cups and covered with a glass lid. The filter paper was kept moist by wetting it with distilled water; the temperature was maintained within 22-24 ° C. The seedlings were counted according to GOST 13056.6-97 on the 7th and 15th days. Per 100 pcs. The following concentrations of aqueous solution extract, ml, were taken from the seeds: 0,25; 0,5; 0,10; 0,05; 0,025. Distilled water served as the control. All experiments

were performed four times. The chemical composition of extracts of woody greens was determined by methods generally accepted in wood chemistry [15]. Experimental data and research results were processed using mathematical statistics methods in Excel and Mathcad computer systems with a confidence of  $P \leq 0.05$  [16-18].

The composition of the aqueous-alkaline extract of coniferous woody greens is presented in Table 1. The content of growth-stimulating substances of the aqueous-alkaline extract of coniferous woody greens is determined: phenolic compounds (2.50 g/l), sodium salts of triterpenic acids (0.67 g/l).

**Table 1** The composition of the extract from the woody greens of coniferous, g / l per absolutely dry mass

The name of the indicator	Content, g/l
Carbohydrates	0,20
Carotenoids	0,14
Fatty acids and their derivatives	0,67
Diterpenoids	0,38
Phenolic compounds	2,50
Lignin-like substances	10,99
Mineral substances	2,11
Total	16,99

The study of the growth-stimulating activity of scots pine seeds was carried out on coniferous extracts of different concentrations prepared by dilution:

Extract 1-0.025ml per 1 liter of distilled water (0.025:1);

Extract 2 -0.05 ml per 1 liter of distilled water (0.05:1);

Extract 3 -0.1 ml per 1 liter of distilled water (0.1:1);

Extract 4 -0.25 ml per 1 liter of distilled water (0.25:1);

Extract 5 -0.5 ml per 1 liter of distilled water (0.5:1).

Seed germination was determined according to GOST 13056.6-97. Distilled water was used as a control. The results of the experiments are shown in the table. 2.

The seed germination energy was determined on day 7 according to the formula [14]:

$$\mathcal{E} = \frac{A \cdot 100}{n}, \%$$

where A is the number of germinated seeds, pieces;

n is the number of seeds in the experiment.

## RESULTS AND DISCUSSION

An analysis of the experimental results (Table 2) showed the presence of two opposite trends in the energy of seed germination: with an increase in the concentration of coniferous extracts to (0.05:1) there is an increase in seed germination energy, a further increase in concentration leads to a decrease in germination energy.

**Table 2** The energy of germination of ordinary pine seeds (*Pinus sylvestris* L) for 7 days on extracts of coniferous woody greens of various concentrations, % per 100 pieces.

The year of the seed harvest	Control	Concentration of solutions, ml/l				
		Extract № 1 (0.5:1)	Extract № 2 (0.25:1)	Extract № 3 (0.1:1)	Extract № 4 (0.05:1)	Extract № 5 (0.025:1)
2017	9,0 ± 0,4	5,0 ± 0,2	13,3 ± 0,5	14,3 ± 0,6	17,0 ± 0,8	16,8 ± 0,7
2018	16,0 ± 0,7	11,3 ± 0,5	27,3 ± 1,3	33,3 ± 1,6	44,8 ± 0,2	36,8 ± 1,8
2019	32,3 ± 1,6	18,3 ± 0,7	37,5 ± 1,8	44,8 ± 2,2	68,5 ± 3,3	54,0 ± 2,3
2020	54,5 ± 2,6	24,3 ± 1,1	43,0 ± 0,9	53,5 ± 2,7	78,3 ± 3,3	65,8 ± 3,2
2021	76,3 ± 3,7	30,8 ± 1,4	49,3 ± 2,2	59,3 ± 2,2	83,8 ± 4,2	76,0 ± 1,8

An increase in the concentration of coniferous extract leads to inhibition of seed growth and a decrease in germination energy. The minimum germination energy of scots pine seeds is observed when using coniferous extracts (0.5:1). The results of germination of seeds of Scots pine (*Pinus sylvestris* L) on the 15th day are shown in Table 3.

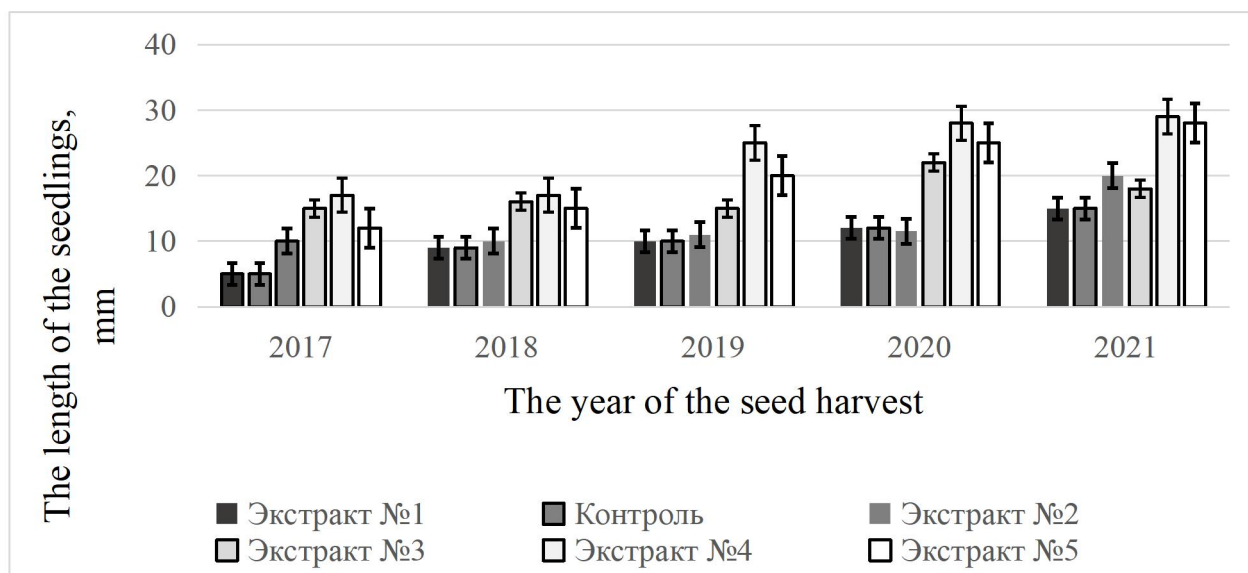
**Table 3** Germination of seeds of Scots pine (*Pinus sylvestris* L) on the 15th day, taking into account non-germinating and rotten seeds, % per 100 pieces.

The year of the seed harvest	Control	Concentration of solutions, ml/l				
		Extract № 1 (0.5:1)	Extract № 2 (0.25:1)	Extract № 3 (0.1:1)	Extract № 4 (0.05:1)	Extract № 5 (0.025:1)
2017	10,0±0,6	6,0±0,4	10,0±0,8	13,0±0,7	14,0±0,5	12,0±0,6
2018	16,3±0,9	9,3±0,9	10,1±0,5	16,6±0,6	19,6±0,9	16,5±0,8
2019	44,3±0,9	39,4±0,6	40,5±0,7	43,1±0,5	55,6±0,4	45,5±1,3
2020	52,5±0,4	42,1±0,5	45,3±0,6	52,5±0,9	65,5±0,5	54,4±1,2
2021	78,5±0,5	58,2±1,2	65,5±0,9	79,4±0,8	88,5±0,6	79,2±1,0

The maximum germination of pine seeds is observed when using coniferous extract № 4 (0.05:1). It is higher than the control by 10% for seeds harvested in 2021 (1 year of storage); by 13% - for seeds in 2020 (2 years of storage); by 11.3% - for seeds in 2019 (3 years of storage); by 3-4% - for seeds in 2018 and 2017 (4-5 years of storage). All other things being

equal, the lowest percentage of germination is observed in the seeds of the 2017 harvest – from 6 to 14%.

The length of the seedlings was determined on the 15th day. The results are shown in Figure 1.



**Figure 1** Dependence of the length of seedlings of scots pine on the 15th day, mm of different harvest years on the concentration of the growth stimulant

The conducted studies have shown that the general dynamics of the growth of the length of seedlings in relation to the control is observed when they are germinated using weakly concentrated solutions of coniferous woody greens up to (0.05:1) and is from 8 to 17% relative to the control. A further increase in concentration leads to a decrease in the length of the seedlings. Apparently, this is due to a decrease in the concentration of biologically active substances, in particular sodium salts of triterpenic acids (0.67g/l) and phenolic compounds (2.61 g/l), which have growth-stimulating activity. When using more concentrated solutions (0.5:1) and (0.25:1) there is no increase in growth-stimulating activity compared to the control.

## Conclusions

1. It was found that concentrated extracts of woody greens have an inhibitory effect on the germination of scots pine seeds (extracts № 1-3) in relation to the control. With an increase in the shelf life, the germination of seeds decreases both in the control and in the experimental sample.

2. It was found out that the greatest germination of seeds is observed when using an extract with a concentration of extractives (0.05:1) Extract № 4.

3. It is proposed to use diluted aqueous extracts of coniferous woody greens (0.05:1) to increase the germination of seeds of the common pine for long-term storage.

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