The effect of a Trichodermin biopreparation based on poplar litter on the germination of tomatoes and wheat

Olga O. Mamaeva* and *Elena* V. Isaeva

Reshetnev Siberian State University of Science and Technology, Krasnoyarsk, Russian Federation

Abstract. The paper presents the results of the study of a biological product of the type "Trichodermin", obtained as a result of biodegradation of plant waste - poplar litter - by Siberian strains K6-15 *Trichoderma spp.* Two methods were used: seed dusting and the introduction of a biological product into the soil with different concentrations. Germination and number of infected seeds of tomato (Irina F1 variety, Geolia) and Novosibirskaya 31 soft spring wheat were evaluated in the work.

1 Introduction

Trichoderma is one of the most studied mushrooms currently. *Trichoderma Pers.: Fr*. Kind was described first more than 200 years ago [1].

On the basis of toxins, enzymes, antibiotics of fungi of the genus *Trichoderma*, preparations are obtained for the biological control of diseases and stimulation of plant growth [2–5]. An example is the *Trichodermin* type biological product, designed to protect agricultural plants from pathogens that spread through the soil and plant residues. Trichoderma is able to suppress more than 60 types of pathogens - *Pythium, Botrytis, Phoma, Sclerotinia, Fusarium, Ascochyta, Alternaria,* etc. [6].

It should be noted that modern intensive agriculture is unthinkable without pesticides, which cause irreparable harm to the environment and human health. Therefore, the rejection of pesticides and the transition to biological products such as "Trichodermin" and others is a very important and relevant area of research.

Various plant wastes can serve as substrates for obtaining Trichodermin [7–11]. Bran, wheat and sorghum straw, peat, grain, beet pulp, corn stalks, grape pomace and sunflower husks are used for mass production of the drug by a surface method in different countries of the world [6].

In a number of works, the possibility of obtaining Trichodermin by cultivating fungi of the *Trichoderma* genus on cellulose granules [7], the vegetative part of Jerusalem artichoke [8], tree greens and post-extraction residues of fir [9], fir bark [10], and larch bark pod [11] has been proved. .

Obtaining a biological product of the type "Trichodermin" is also possible on the vegetative part of the poplar, namely the post-extraction residue of buds and shoots. A

* Corresponding author: olga07_95@mail.ru

biological preparation based on the Magansky strain ("MG-97" *T. aspirellum*) contains 0.22·109, the Krasnoyarsky strain ("10-99" *T. harzianum*) contains 0.52·109 spores/g) [12].

In addition, the substrate for the cultivation of *Trichoderma* fungi can be balsam poplar leaves, including fallen leaves, which are processed to a limited extent, mainly burned or disposed of in a landfill. It has been established that fungi of the genus *Trichoderma* (K6-15 strain) can form 0.4-3.6·109 spores/g on deciduous substrates [13].

Thus, *Trichoderma* is one of the most studied fungi at present. The reason for this interest lies in the great practical and ecological significance of the genus. On the basis of the fungus, it is possible to obtain a biological product that helps to increase the size of the root system, growth and vitality of plants by controlling the rhizosphere microflora, the percentage of seed germination, increasing the accumulation of nutrients and influencing plant metabolism. It should be noted that fungi of the *Trichoderma* genus grow at a faster rate on the vegetative part of plants than on other wastes, such as wood.

The purpose of this study was to study the effect of a microbiological preparation obtained in the process of biodestruction of fallen poplar leaves by a microscopic fungus of the *Trichoderma* genus on the germination of some agricultural crops.

2 Research methods

Fallen leaves of balsam poplar (*Populus balzamifera L.*) were used to obtain a biological product. Leaf samples were taken in September 2021 on the territory of Krasnoyarsk. The raw material was dried, ground (particle size 5-6 mm) and stored in closed vessels at constant humidity.

The Siberian strain of microscopic fungi of the K6-15 *Trichoderma spp.* genus isolated from cedar wood on the territory of the arboretum of the Forest Institute named after A. V.N. Sukachev of SB RAS (Krasnoyarsk, Akademgorodok) in 2015. This strain was kindly provided by Yu. A. Litovka, Professor of the Department of Chemical Technology of Wood and Biotechnology of Siberian State University named after M. F. Reshentev.

The solid-phase method of cultivation was used in the work. The preparation of the plant substrate was carried out as follows: the crushed substrate was brought to 70% humidity with tap water, placed in Petri dishes, and sterilized for 30 min at a pressure of $1.01 \cdot 10^5$ Pa several times in a VK-75 autoclave. The substrate was cooled to (25 ±1) °C and a spore suspension of the museum strain K6-15 *Trichoderma spp.* was added with a titer of $1 \cdot 10^6$ spores per 1 g of substrate. The cultures were incubated in a thermostat at (28±2)°C for 14 days. The titer of spores was counted in a Goryaev chamber [10].

The resulting biodegradation product was dried at a temperature of (25 ± 1) °C to a residual moisture content of not more than 10%, crushed to a powder and used as a biological product.

Various methods of introducing biological preparations are practiced, such as seed treatment, soil application, spraying [14, 15]. Experiments to evaluate the effectiveness of the drug were carried out by two methods, using tomato seeds (variety Irina F1, Geolia) and Novosibirskaya 31 wheat grain (*Triticum aestivum L.*) infected with fungi of the *Penicillium* genus. The first method consisted in dusting the seeds with a biological product, the second method was the introduction of the drug into the soil (0.05 g and 0.01 g per 20 cm^3 of land). Germination and the number of infected seeds were evaluated in the experiment.

3 Research results

To study the biological product obtained in the process of destruction of fallen leaves of balsamic poplar using K6-15 *Trichoderma spp.* strain (titer 7.1 108 CFU/g), tomato and wheat seeds were used.

Tomato is one of the most common vegetable crops in Russia. According to the data for 2020, Russia ranks 12th (2.9 million tons), tomatoes were grown on 80,767 hectares with an average yield of 36.8 kg/m² [16].

Figure 1 shows the results of the germination of a tomato seed using the obtained preparation of the Trichodermin type.

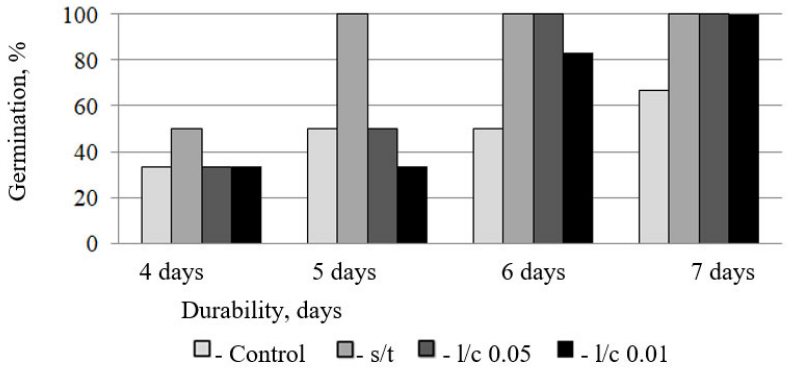

Fig. 1. Tomato seed germination (s/t - seed treatment, l/c - land cultivation)

It was found that using the pollination method, the germination of tomato seeds on the 5th day was 100%, which is two times higher compared to the control. In the process of soil treatment with a biological preparation, 100% germination was observed on the 6th day of germination when the preparation was applied at a concentration of 0.05 g/20 cm3.

Novosibirskaya 31 wheat (Triticum aestivum L.) is a soft spring wheat. In the middle of the 20th century, spring wheat was replaced by more productive winter wheat, but now there is a steady demand for insurance varieties. According to the monitoring data of the Federal State Budgetary Institution Rosselkhoztsentr, in 2022 Novosibirsk wheat 31 of the selection of the Federal State Budgetary Institution Institute of Cytology and Genetics of the Siberian Branch of the Russian Academy of Sciences took the second place in terms of sowing volumes in the Russian Federation (134.9 thousand tons) [17].

In this work, we used the already infected wheat grain Novosibirskaya 31, which was provided by the Krasnoyarsk Agrarian University. It is known that at high grain moisture content (more than 15–16%) during storage, mold fungi can develop, in particular, species of the genera Penicillium, Aspergillus, Mucor, etc., causing a decrease in seed germination and quality of commercial products [18].

The results of germinating the grain of infected wheat are shown in Figure 2.

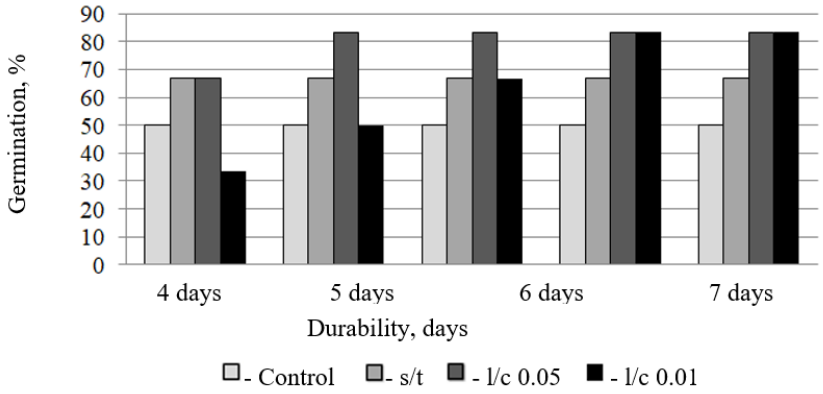

Fig**. 2.** Germination of wheat grain (s/t - seed treatment, l/c - land cultivation)

Figure 2 shows that when the infected grain of wheat was treated by pollination, the germination capacity increased already on the 3rd day by 1.3 times and amounted to + 136% of the control and then does not change. When the biopreparation is applied to the ground with a concentration of 0.05 g/20 cm^3, the maximum germination is observed on the 4th day (+166% to the control), the same germination when the preparation is applied with a concentration of 0.01 g/20 cm3 is recorded on the 6th day.

In the process of germination, traces of rot were found in 33% of the grain in the control; no traces of rot were found when using the biological product.

4 Conclusion

Thus, the results of the study showed that the use of the drug increases the germination of seeds of both tomato and wheat. In addition, during the germination of wheat grain, the drug completely suppresses the development of mold fungi. The data obtained are of interest for further research in order to use this environmentally friendly biological product to protect plants from phytopathogens.

The work was carried out within the framework of the state task of the Ministry of Education and Science of the Russian Federation for the implementation by the team of the scientific laboratory "Deep processing of plant raw materials" of the project "Technology and equipment for the chemical processing of biomass of plant raw materials" (Theme number FEFE-2020-0016).
The work was performed using the equipment of the Krasnoyarsk Regional Center for Collective Use of the FRC KSC SB RAS. We express our gratitude to the staff of the Center for Collective Use for their assistance in conducting research.

References

1. D. V. Baurin, Advances in Chemistry and Chemical Technology **23, 10 (103),** 77-79 (2009).
2. G. E. Harman, C. R. Howell, A. Viterbo, I. Ch &, Nat. Rev. Microbiol **2(1)**, 43-56 (2004)
3. N. N. Grinko, Ecological aspects of regulation of populations of phytopathogenic micromycetes of vegetable crops in greenhouses **37** (2001).
4. F. K. Alimova, N. G. Zakharova, G. A. Mako, A. N. Fattakhova, Pakistan J. of scientific and industrial reseach **2002** 38-45 (2002)

5. D. G. Georgakopoulos, P. Fiddaman, C. Leifert, N. E. Malathrakis, J. Appl Microbiol **92(6)**, 1078-1086 (2002)
6. F. K. Alimova, Industrial use of fungi of the genus Trichoderma **209** (2006)
7. I. T. Geller, I. B. Berkutova, and A. L. Pereverzeva, Biotechnology of Microorganisms in Agriculture, **1989** 115-122 (1989)
8. T. V. Ryazanova, A. N. Chuprova, and Yu. A. Litovka, Sistemy. Methods. Technologies **1(29),** 147-151 (2016)
9. Yu. A. Litovka, A. G. Savitskaya, T. V. Vasilievna, and N. A. Neshumaeva, Chemistry of Plant Raw Materials **3,** 167-172 (2011)
10. P. N. Bondar, Fungal strains of the genus Trichoderma Pers (Fr.) as a basis for creating plant protection preparations and obtaining feed additives **220** (2011)
11. E. G. Makhova, Cultivation of fungi of the genus Trichoderma on carbohydrate substrates and obtaining a biological product **22** (2003)
12. G. A. Lozhkina, E. V. Isaeva, Patent No. 2322501 RF Method for complex processing of the vegetative part of balsam poplar **6** (2008)
13. E. V. Isaeva, T. V. Ryazanova, O. O. Mamaeva, RF Patent No. 2763403 C1 Method for the complex processing of the vegetative part of the balsam poplar by the bioconversion method **9** (2021)
14. R. N. Okigbo, Phytopathology **148(6)**, 351-355 (2000)
15. E. K. Yuzefovich and D. V. Voitka, Plant Protection **40,** 219-229 (2016)
16. FreshPlaza, EC (2021), https://www.tomatonews.com/en/worldwide-total-fresh-tomato-production-exceeds-187-million-tonnes-in-2020_2_1565.html
17. Top varieties of spring wheat for 2022, https://direct.farm/post/top-sortov-yarovoy-pshenitsy-za-2022-god-12977
18. M. Koishybaev, Wheat Diseases **394** (2018)