Efficacy of *Pseudomonas* spp. in control of apple powdery mildew

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Abstract. The results of Pseudomonas spp.`s effect on Podosphoera leucotricha (Ell. et Ev.) (Salm.) in commercial plantings of apple trees of Renet Simirenko variety are presented in the article. It reveals that use of the bio-fungicide in three consistent treatments during fruit ripening period at the rate of 4.0 l/ha reduces the disease` spread and development intensity, ensuring biological efficacy of 94.4-99.6%.

1 Introduction

In horticulture of the Russian Federation apples are the main fruit crop, with its fruits being of a high taste quality, and also being a highly valuable sources of vitamins and microelements` complex. The demand for apples predetermines the necessity of ensuring their high yield and quality. Mycopathogens pose a serious threat in this regard, among which powdery mildew, caused by *Podosphoera leucotricha* (Ell. et Ev.) (Salm.) [1, 2, 3, 4] ascomycete is economically significant in many regions of apple cultivation. At severe affection level the disease causes a decrease in shoots growth, death of inflorescences, leaves, shoots, leads to decrease in fruit quality and yield, and that results in the increase of the market fruits price [4, 5]. In the south of Russia, on highly susceptible apple varieties, the disease epiphytotics affects up to 80% of leaves and shoots, which leads to decrease of frost resistance of trees and the yield`s decrease by almost 50%. Fruits are strongly affected during the ripening period, a rusty mesh is formed on them, they are smaller and deformed [6].

The climate of the Krasnodar Territory usually accounts for mild winters, warm and sunny spring, relative humidity of 70-100%, long growing season, and all that is favorable for accumulation of pathogen inoculum. These factors account for the need of regular spraying for powdery mildew control: from the infection start in the budding phase of the apple tree to the harvest. However, long-term use of chemical pesticides in intensive horticulture causes irreparable damage both to the environment and human health in the result of poor-quality food consumption.

In order to reduce the chemicals` treatments volume and improve the yield quality, increase the yield rate, it is necessary to search for and apply new, non-traditional fungicides and microbiological agents [7, 8]. In addition, when using chemicals, the risk of developing resistance to them is quite high. There are scientific data that the most harmful species of *Erysiphales* are resistant to several classes of major chemical compounds used for powdery

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mildew control: sterol demethylation inhibitors, external quinone inhibitors, succinate dehydrogenase inhibitors; resistance can develop in the course of several years [9]. The main recommendations for reducing resistance development possibility include use of fungicides with alternative action mechanisms: new chemical classes [1] or microbiological products. The search for microbial agents for apple powdery mildew control, according to many researchers, is a powerful alternative strategy [3] and is carried out all over the world. For this purpose, strains of *Bacillus spp. (B. subtilis, B., amyloliquefaciens, B. velezensis*) [3] and *Pseudomonas spp*. are used. In industrial apple orchards of Russia, only four biological products are currently allowed for powdery mildew control: based on B. subtilis - Baktofit, SP strain IPM 215, Baktofit, SK strain IPM 215, Fitosporin-M, Zh strain 26D, and also Rizoplan, Zh based on *Pseudomonas fluorescens* strain AP-33.

The relevance of these studies is associated with the observed trend of susceptibility decrease of fungicides of chemical synthesis to apple tree powdery mildew pathogen, that of increase of its harmfulness during fruit ripening, and also of the limited range of microbiological products for its control.

2 Materials and methods

As it known, it is possible to increase the biological products` effectiveness in the course of field tests by optimizing application timing and consumption rates. Our studies in biological efficacy evaluation of experimental biofungicide based on bacteria of *Pseudomonas* genus in apple powdery mildew control were carried out in the field. The production experiment was carried out according to generally accepted methods in 2021 in the garden located in the western part of the Krasnodar Territory, on the Renet Simirenko variety of late ripening. Planting scheme 5 x 2 m, tree height 2.0-2.5 m, 2010 planting year. The area of the test variant is 1.5 ha. Control variant was at a plot of four rows with 300 m2 total area. The biological product was used three times in successive treatments: on August 20 - at stage 79, about 90% of the cultivar-typical fruit size was achieved; August 27 - at stage 81, the beginning of fruit ripeness (variety-specific lightening of the main color); 09.09 – at stage 85, advanced fruit ripeness, increasing varietal intensity of color. There were tested two application rates of microbio-fungicide: 2.0 and 4.0 l/ha. As a standard, we used Fitosporin-M, Zh, the active principle of *Bacillus subtilis* strain 26D (titer of at least 1 billion living cells and spores/ml) with application rate of 2.0 l/ha. In this time ranges in control variant there were no treatment either with biological formulation or chemical fungicides. The tank mixture rate was 1000 l/ha. Recording was made: 20.08 – before the use of biological formulations; August 26 - on the 6th day after the first treatment; 09/02 and 09/08 - on the 6th and 12th days after the second treatment; September 15 - on the 6th day after the third treatment; September 24 - on the 15th day after the third treatment, during harvesting. After each treatment the diseases spread and development intensity were determined. The biological efficacy of the products was calculated using the Abbott formula.

3 Results and Discussion

The first signs of powdery mildew in 2021 were noted on April 9, at “a pink bud” stage, 0.7% of leaf rosettes were affected. The air temperature for spring conidia in April was below the optimum: it did not exceed +10.0°C…+12.4°C. The most favorable conditions for powdery mildew happened in August and September: periodic change of significant precipitation with hot dry weather.

In season 2021, the fruits were not affected by powdery mildew. On the leaves before testing, the disease spread was 3.50%, the development intensity was 1.25%.

After the first treatment with biological formulation on the 6th day (August, 26), when using a biofungicide with application rate of 2.0 l/ha, the number of affected leaves decreased by 4.7 times, their damage intensity decreased by 1.8 times; when applied at application rate of 4.0 l/ha these indicators decreased by 5.8 and 6.3 times respectively, while in standard variant indicators were by 8.8 and 12.5 times respectively (table 1). The difference in biological efficacy of 2.0 and 4.0 L/ha biofungicide application rates was significant. In control variant in the period of 20-26.08, the spread and development intensity of the disease increased by 2.9 times.

Table 1. Biological efficacy of the bio-preparation based on *Pseudomonas* in apple powdery mildew control, %.

Variant	Recording dates									
	26.08		02.09		08.09		15.09		24.09	
	P, %	BE, %	P, %	BE, %	P, %	BE, %	P, %	BE, %	P, %	BE, %
Biofungicide 2.0 l/ha	0.75	92.5	3.00	72.7	2.00	88.2	0.50	97.4	3.00	87.5
Biofungicide 4.0 l/ha	0.60	94.0	0.20	98.2	1.83	89.2	0.20	98.9	3.00	87.5
Fitosporin-M, Zh 2 l/ha	0.40	96.0	0.20	98.2	1.33	92.2	0.20	98.9	2.00	91.7
Control	10.00	-	11.00	-	17.00	-	19.00	-	24.00	-
HCP		2.1		6.1		2.3		1.5		2.5

Symbols: P – desease spread; BE - biological efficacy.

After the second treatment with biological product on the 6th (02.09) day, in the variant of applying the biofungicide with a consumption rate of 2.0 l/ha, the development of powdery mildew was not restrained: the number of affected leaves increased 4 times, the intensity of their damage increased 2 times, as in control. In the variant of application of the biofungicide with a consumption rate of 4.0 l/ha, the spread and intensity of the development of the disease, on the contrary, decreased: by 3 and 2 times, respectively; the effectiveness of protection in the variant corresponded to the effectiveness of the standard. In the control variant, for 7 days (August 26-September 26), the intensity of the development of the disease increased from 3.6 to 8.5%, or 2.4 times.

On the 12th day after the second treatment (September 08), in both variants of the application of the biofungicide, similar values of biological efficiency were obtained. In the control from 02.09 to 08.09, the spread and intensity of development of powdery mildew increased by 1.5 and 1.4 times, respectively.

After the third treatment on the 6th day (September 15), in all variants of biological products use the spread and development intensity of the disease decreased: in the variant of the biofungicide with application rate of 2.0 l/ha, by 4 and 2.3 times, respectively, in the variant with a consumption rate of 4 .0 l/ha - by 9.2 and 16.6 times. The biological efficacy of both vatiants corresponded to the efficacy of the standard. Over the past 7 days (08-15.09) in control the spread and development intensity of powdery mildew increased very slightly.

Data recording during harvesting showed that in control variant, the disease`s spread and the development intensity amounted to 24.0 and 18.0%, respectively. The variant of using the bio-fungicide at application rate of 4.0 l/ha was not inferior to the standard variant in terms of efficiency and significantly exceeded the variant of using a biofungicide with an application rate of 2.0 l/ha.

4 Conclusion

As the result of the tests it was found that under production conditions, an experimental microbiological fungicide based on bacteria of *Pseudomonas* genus can be used for apple powdery mildew control in three successive treatments in phases: when about 90% of the cultivar-typical fruit size is achieved; at the beginning of fruit ripeness; at an advanced fruit ripeness. A higher protection efficiency, not inferior to the effectiveness of Fitosporin-M, Zh product, registered for use on apple trees, is achieved when using a biofungicide with a consumption rate of 4.0 l/ha.

The use of the biofungicide based on *Pseudomonas spp.* during fruit ripening period will allow for yielding environmentally friendly products, reduce pesticide pressure, as well as the risk of *P. Leucotricha* resistance to chemical fungicides.

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