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Efficiency of *Bacillus thuringiensis* bioinsecticide in grapevine protection

Viktor I. Dolzhenko[1], Olga V. Shapoval[2], Yuri N. Pleskachev[3], Tamara S. Astarkhanova[4] ✉

[1]All-Russian Institute of Plant Protection, *St. Petersburg, Russian Federation*
[2]Pryanishnikov Institute of Agrochemistry, *Moscow, Russian Federation*
[3]Federal Research Center "Nemchinovka", *Moscow region, Russian Federation*
[4]RUDN University, *Moscow, Russian Federation*
✉ astarkhanova-ts@rudn.ru

Abstract. The problem of contamination of agricultural products with toxic substances is an urgent problem in production. One of the most chemically treated crop is vineyards. Multiple treatments during one growing season have led to the fact that grape plantings have become an accumulator of unsafe chemical plant protection products. Moreover, grapes are consumed fresh. Accordingly, the selection of environmentally friendly pesticides and the replacement of chemicals with biological ones has become a priority for producers of this crop. Our research was devoted to evaluation of bioinsecticide based on entomopathogenic bacteria *B. thuringiensis* in the system of protecting grapes from pests. Such studies are of great interest and, according to the literature, 90 % of registered insecticides are based on *Bacillus thuringiensis*. Microorganisms isolated from nature as plant protection agents, when reintroduced into natural conditions, preserve beneficial species in biocenoses. At the same time, they are selective and, while affecting certain harmful objects, do not cause harm to humans and the environment. The purpose of this work was to study the effect of the bioinsecticide Biometch Insecto, WP on grapevine pests: European grape worm (*Lobesia botrana* Den. & Schiff.) and leafy form of phylloxera (*Viteus vitifolii* Fitch.). Biometch Insecto, WP is based on strains of *Bacillus thuringiensis* var. *kurstaki* HG207 and *Beauveria bassiana* HG208. The effect of entomopathogens was compared with the effect of insecticide Bioslip, BW, L, the active ingredient of which is *Beauveria bassiana*. The results of the research revealed high biological effectiveness (90…100 %) of Biometch Insecto, WP in protecting grapevines from pests when applied during the period of mass hatching of European grape worm caterpillars and emergence of phylloxera larvae from galls.

Keywords. European grape worm, leaf form of phylloxera, grapevine, Biometch Insecto, *Beauveria bassiana*

Conflicts of interest. The authors declared no conflicts of interest. Biometch Insecto, WP is not registered and is used for scientific purposes.

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Introduction

Human activity has come into contradiction with the global problem of environmental protection, which has caused an urgent need to use pesticides that do not have a harmful effect on humans and the environment [1].

This problem is especially visible in production of grapes, where 12-fold treatments are used in one growing season, crop rotations due to monoculture are excluded. Therefore, the toxicological load on the agrocenoses of grapevine plantations must be reduced.

Grapes are consumed fresh and the decomposition of harmful toxicants, which could occur during heat treatment, is impossible [2, 3].

High biological efficiency and speed of action of chemical pesticides contributed to production of high crop yields, but the negative consequences of chemical pesticides application were also identified — their accumulation in ecosystem and the development of resistant populations [4, 5]. It has led to a sharp increase in the number of phytophages and epiphytotic diseases, requiring an increasing use of chemicals for their control [6]. These and other negative consequences of using pesticides have led to awareness of the need to improve plant protection, move from individual techniques to their integration in the system, and develop more environmentally friendly methods [7]. These are biopesticides, which are directly natural materials or agents developed on their basis. Production of biopesticides is more economically profitable and environmentally friendly.

In the modern production of plant protection products, bacterial agents belonging to the new generation of insecticides are effective against about 400 species of insects, including vineyard pests, and 90 % of registered insecticides are made of *Bacillus thuringiensis* [8, 9]. Bacteria produce specific crystalline toxins that have great entomocidal activity [10].

Bacillus thuringiensis var. *kurstaki* has an intestinal effect, the fungus is effective only when it enters insect intestines during active feeding. The toxin, activated in intestinal tract of caterpillar, damages the inner lining of intestine, as a result, osmotic balance is disrupted and alkaline contents of the intestine leak into the body of the caterpillar [11, 12].

Beauveria bassiana fungus reproduces only by conidia, which, once on the insect's body, secrete an enzyme at the points of attachment. It dissolves the cuticle and conidia grow into the body cavity. The toxins released by the fungus during development lead to the death of the insect [13]. Further development of the fungus occurs in the dead insect [14]. The effectiveness of biological agents in protecting grapevines from pests and diseases depends on knowledge of real phytosanitary situation in the plantings, on

cultivar resistance, cultivating technology and climatic conditions. After substantiating biologization of protection during grape production, the range of bioagents and application terms should be determined [15].

The purpose of the study was to evaluate the effectiveness of bioinsecticide Biomech Insecto, WP based on strains of *Bacillus thuringiensis* var. *kurstaki* HG207 and *Beauveria bassiana* HG208 on grapevine pests: European grape worm (*Lobesia botrana* Den. & Schiff.) and grape phylloxera (*Viteus vitifolii* Fitch.).

Materials and methods

The research was carried out on Riesling grapevines in the second soil-climatic zone in conditions of the Republic of Dagestan. The research material was the insectoacaricide Biometch Insecto, WP, presented for scientific purposes.

The experiment was small-plot and had 5 bushes. Counts of European grape worms were perfomed in accordance with Methodological Guidelines for registration testing of insecticides, acaricides, pheromones, molluscicides and rodenticides in crop production. Moscow; 2022.

Grape phylloxera (leaf form) — *Viteus vitifolii* Fitch. appeared during the period when larvae hatched from overwintering eggs and colonized opening buds, which coincided with appearance of 4–5 leaves on the shoots. Treatments were carried out during the period of mass emergence of larvae of the first and second generations from galls, which coincided with appearance of 9…12 or 17…20 leaves on the shoots. Counts were carried out three times, before treatment, on the 30th day after treatment and at the end of the growing season.

Biological efficiency was calculated using the Henderson and Tilton formula. If it was not possible to accurately determine the number of dead insects, the effectiveness was determined using the Abbott formula.

Results and discussion

During the experiments on grape plantations, hatching of European grape worm caterpillars of the first generation and mass emergence of second-generation larvae from the galls were noted during appearance of 17…20 leaves on the shoots. Treatments were carried out with Solo backpack sprayer at a rate of 500 L/ha of working solution three times with an interval of 7 days.

The tested bioinsecticide Biomech Insecto, WP showed high efficiency in controlling European grape worm on grapevine. The average number of caterpillars per m^2 in Variant 1 (application rate 0.7 kg/ha) on the 3rd day after the last treatment was 7.5 and on the 21st day decreased to 3.0, biological efficiency increased from 79.5 to 91.1 %. After increasing application rate of Biomech Insecto, WP to 2.0 kg/ha (Variant 2), average number of caterpillars per m^2 decreased to 2.5, effectiveness of Biomech Insecto, WP reached 92.6 %. The effectiveness of the tested bioincecticide was close to the effectiveness of the standard Bioslip, BW, L (active ingredient — *Beauveria*

bassiana), when the average number of caterpillars per m^2 was reduced to 2.3 and the efficiency reached 93.4 % (Table 1).

Table 1

Biological effectiveness of Biomech Insecto, WP in controlling European grape worm (*Lobesia botrana*)

Variant	Application rate	Replication	Average number of caterpillars per plant					Decrease in pest number, %, after application by day			
			Before application	Days after application							
				3	7	14	21	3	7	14	21
1. Biomech Insecto, WP	0.7 kg/ha	1	25.1	9	7	5	3	71.4	77.9	83.4	89.6
		2	31.2	7	6	4	2	82.1	84.7	89.3	94.4
		3	29.9	8	5	2	4	78.6	86.7	94.4	88.4
		4	30.8	6	6	3	3	84.4	84.5	91.9	91.5
		mean	29.3	7.5	6.0	3.5	3.0	79.5	83.7	90.0	91.1
2. Biomech Insecto, WP	2.0 kg/ha	1	27.2	7	5	4	3	79.4	85.4	87.7	90.4
		2	32.8	8	4	2	2	80.5	90.3	94.9	94.7
		3	30.6	6	5	4	3	84.3	87.0	89.1	91.5
		4	27.4	7	7	2	2	79.6	79.7	93.9	93.7
		mean	29.5	7.0	5.3	3.0	2.5	81.0	85.9	91.5	92.6
3. BioSleep, BW, L Standard	3.0 L/ha, two treatment	1	25.7	7	5	3	2	78.2	84.6	90.3	93.2
		2	33.1	5	3	3	3	87.9	92.8	92.4	92.1
		3	30.9	8	5	2	2	79.3	87.2	94.6	94.4
		4	28.8	7	6	3	2	80.6	83.5	91.3	94.0
		mean	29.6	6.8	4.8	2.8	2.3	81.8	87.3	92.3	93.4
Average to control	–	mean	29.4	36.8	46.4	55.7	64.2	–	–	–	–

Treatments carried out during the period of mass emergence of second-generation larvae from galls and 7 and 14 days after the first treatment showed the high effectiveness of the bioagent in controlling the leaf form of phylloxera — *Viteus vitifolii* Fitch.

On the 30th day after treatment, the number of colonized grapevines decreased on average to 1.0 in Variant 1 and to 0.75 in Variant 2, or by 74.2 and 79.2 %, respectively. The intensity of gall formation by the end of the growing season was 0.5 points in Variant 1, before treatment — 4.0 points, with decrease in intensity of gall formation to 86.7 %. With increase in application rate to 2.0 kg (Variant 2), the effectiveness of the bioproduct reached 0.25 points while the intensity of gall formation decreased to 90.0 %. In the standard variant Bioslip, BW, L, the indicators did not have a significant difference with the indicators of the variant with Biomech Insecto, WP; the reduction in the intensity of gall formation was 88.8 % (Table 2).

Table 2

Biological effectiveness of Biomech Insecto, WP in controlling grape phylloxera (*Dactylosphaera vitifoliae*)

Variant	Application rate	Replication	Number of infested vines		Gall formation intensity, grade		Decrease, %		Biological berry yield, c/ha
			Before application	30 days after application	Before application	End of growing season	Vine infestation	Gall formation intensity	
1. Biomech Insecto, WP	0.7 kg/ha	1	5	1	5	1	80.0	80.0	70.2
		2	4	1	4	0	75.0	100.0	70.5
		3	3	1	3	1	66.7	66.7	70.3
		4	4	1	5	0	75.0	100.0	71.2
		mean	4	1.0	4.3	0.5	74.2	86.7	70.6
2. Biomech Insecto, WP	2.0 kg/ha	1	5	1	4	0	75.0	80.0	78.5
		2	5	0	5	1	100.0	80.0	78.9
		3	3	1	5	0	66.7	100.0	82.1
		4	4	1	3.5	0	75.0	100.0	80.8
		mean	3.8	0.75	4.4	0.25	79.2	90.0	80.1
3. BioSleep, BW, L Standard	3.0 L/ha	1	4	1	3	0	75.0	100.0	79.6
		2	5	1	4	1	80.0	75.0	78.5
		3	5	1	5	1	80.0	80.0	74.8
		4	4	1	3	0	75.0	100.0	77.5
		mean	4.5	1.0	3.8	0.25	77.5	88.8	77.6

Conclusion

Data obtained in experiments on the use of Biomech Insecto, WP to control grapevine pests revealed:

1. Biomech Insecto, WP at the dose of 0.75…2.0 kg/ha provides protection of grapevine from European grape worm (*Lobesia botrana* Den. & Schiff.) and the leafy form of phylloxera (*Viteus vitifolii* Fitch.).

2. Depending on the research year and the soil-climatic zone, safety interval for the bioinsecticide vary from 14 to 21 days.

3. Biomech Insecto, WP can be applied three times during the hatching period of first-generation caterpillars of European grape worm with an interval of 7 days.

4. Measures to control leaf form of phylloxera (*Viteus vitifolii* Fitch.) can be performed during mass emergence of first-generation larvae from galls at 0.7…2.0 kg/ha application rate.

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About authors:

Dolzhenko Viktor Ivanovich — Doctor of Agricultural Sciences, Professor, Academician of the Russian Academy of Sciences, Head of the Center for Biological Regulation, All-Russian Institute of Plant Protection, 3 Podbelskogo highway, Pushkin, St. Petersburg, 196608, Russian Federation; e-mail: dolzhenko@iczr.ru
ORCID: 0000-0003-4700-0377

Shapoval Olga Vladimirovna — Doctor of Agricultural Sciences, Professor, Chief Researcher, Laboratory for Testing Elements of Agricultural Technologies, Agrochemicals and Pesticides, Pryanishnikov Institute of Agrochemistry, 31a Pryanishnikova st., Moscow, 127434, Russian Federation; e-mail: elgen@mail.ru
ORCID: 0000-0002-3375-527X

Pleskachev Yuri Nikolaevich — Doctor of Agricultural Sciences, Professor, Chief Researcher, Center for Agriculture, Federal Research Center "Nemchinovka", 6 Agrokhimikov st., Novoivanovskoe vil., Odintsovskiy district, Moscow region, 143026, Russian Federation; e-mail: pleskachiov@yandex.ru
ORCID: 0000-0001-5771-5021

Astarkhanova Tamara Sarzhanovna — Doctor of Agricultural Sciences, Professor, Agrobiotechnological Department, Agrarian and Technological Institute, RUDN University, 8/2 Miklukho-Maklaya st., Moscow, 117198, Russian Federation; e-mail: astarkhanova-ts@rudn.ru
ORCID: 0000-0002-1431-9309

Эффективность биоинсектицида на основе энтомопатогенных бактерий *B. thuringiensis* для защиты винограда

В.И. Долженко[1], О.В. Шаповал[2], Ю.Н. Плескачев[3], Т.С. Астарханова[4] ✉

[1]Всероссийский научно-исследовательский институт защиты растений, *г. Санкт-Петербург, Пушкин, Российская Федерация*
[2]Всероссийский научно-исследовательский институт агрохимии им. Д.Н. Прянишникова, *г. Москва, Российская Федерация*
[3]Федеральный исследовательский центр «Немчиновка», *Московская обл., Российская Федерация*
[4]Российский университет дружбы народов, *г. Москва, Российская Федерация*
✉ astarkhanova-ts@rudn.ru

Аннотация. Рассматривается актуальная проблема загрязнения сельскохозяйственной продукции токсичными веществами в производстве. К самым обрабатываемым химическими препаратами культурам относятся виноградники. В результате многократных обработок за один вегетационный период виноградные насаждения становятся аккумулятором небезопасных химических средств защиты растений. Соответственно, подбор экологически безопасных пестицидов, замена химических препаратов биологическими — задача производителей данной культуры. Актуальность обусловлена еще и тем, что виноград потребляется в пищу в свежем виде. Проведена оценка применения биоинсектицида на основе энтомопатогенных бактерий *B. thuringiensis* в системе защиты винограда от вредителей. По литературным данным 90 % зарегистрированных инсектицидов разработаны на основе *Bacillus thuringiensis*. Выделяемые из природы микроорганизмы в качестве средств защиты растений при обратном внесении в природные условия позволяют сохранять полезные виды в биоценозах, при этом, обладая селективностью и поражая определенные вредные объекты, не причиняют вреда человеку и окружающей среде. Цель исследования — изучение влияния биоинсектицида Биомеч Инсекто, СП на вредителей виноградной лозы: гроздевую листовертку (*Lobesia botrana Den. & Schiff.*) и листовую форму филлоксеры (*Viteus vitifolii Fitch.*). В исследовании использован биоинсектицид Биомеч Инсекто, СП на основе штаммов *Bacillus thuringiensis var. kurstaki* HG207 *и Beauveria bassiana* HG208. Влияние энтомопатогенов сравнивалось с действием инсектицида Биослип, БВ, Ж, действующим веществом которого является *Beauveria bassiana*. Установлена высокая биологическая эффективность биоинсектида Биомеч Инсекто, СП, достигающая 90…100 % в защите виноградной лозы от вредителей при обработке в период массового отрождения гусениц гроздевой листовертки и выхода личинок из галлов листовой формы филлоксеры всех поколений.

Ключевые слова: гроздевая листовертка, листовая форма филлоксеры, виноградная лоза, Биомеч Инсекто, *Beauveria bassiana*

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